Contents

ORIGINAL RESEARCH

Nutritional status in conflict

Food production and processing

Indicators of nutritional status

Economic and social factors in malnutrition

Fortification

Food and Nutrition Bulletin, vol. 28, no. 1

United Nations University Press
Published by the International Nutrition Foundation for The United Nations University
53-70 Jingumae 5-chome, Shibuya-ku, Tokyo 150-8925, Japan
Tel.: (03) 3499-2811 Fax: (03) 3406-7345
E-mail: mbox@hq.unu.edu
ISSN 0379-5721
Design and production by Digital Design Group, Newton, MA USA
Printed on acid-free paper by Webcom, Toronto, ON Canada

Nutritional status in postconflict Afghanistan: Evidence from the National Surveillance System Pilot and National Risk and Vulnerability Assessment

Wendy A. Johnecheck and Diane E. Holland

Abstract

Background. *Two large-scale studies, the National Surveillance System (NSS) Pilot Study (2003–2004) and the National Risk and Vulnerability Assessment (NRVA) 2003, were conducted by government, United Nations, and nongovernmental organizations in Afghanistan, as part of wider efforts characterizing Afghan livelihoods in relation to particular outcomes of interest: vulnerability to poverty, food insecurity, and malnutrition.*

Objective. *To present the data from these two surveys with nutrition as the key outcome of interest, and to further construct the understanding of the underlying causes of malnutrition, thus providing public health practitioners and other sector specialists with insight into how a variety of sectoral programs can impact nutritional outcomes in Afghanistan.*

Methods. *The NSS gathered information on livelihoods, food security, and nutrition from 20 to 40 randomly selected households in each of 26 purposively selected sentinel sites (representative of livelihood zones) during November–December 2003 and May–June 2004. The NRVA gathered information nationally from households selected with a two-stage sampling (based on livelihood zone and then socioeconomic group) during July–September 2004.*

Results. *Acute malnutrition is below emergency levels for children under five. The level of chronic malnutrition in children under five indicates a problem of public health importance. Dietary diversity in Afghanistan is not as low as expected but still shows room for improvement, particularly in remote areas and with respect to food groups associated with adequate micronutrient intake. The findings also suggest that in addition to lack of adequate household food intake, recurrent illness and suboptimal infant and young child feeding and hygiene practices contribute to poor nutritional outcomes in this age group. The survey also found poor access to health care, markets, and water for household use.*

Conclusions. *Improving nutritional status requires a multipronged approach, directly targeting malnutrition, coupled with economic growth, household livelihood security, social protection, access to public health services, and water and sanitation. Nutrition policy, programming, and monitoring need to reflect the immediate and underlying causes of malnutrition. Future research needs to be designed to quantify the relative contribution of underlying causes of poor nutrition, allowing practitioners to prioritize responses aimed at improving nutritional outcomes.*

Key words: Afghanistan, conceptual framework of malnutrition, diet quality, dietary diversity, livelihoods, malnutrition, maternal nutrition status, survey, morbidity, nutritional risk, postconflict, seasonality, surveillance, vulnerability

---

Wendy A. Johnecheck is affiliated with the Gerald J. and Dorothy R. Friedman School of Nutrition Science and Policy, Tufts University, Boston, Massachusetts, USA. Diane E. Holland was affiliated with Concern Worldwide Burundi at the time of submission; she is now working as a public nutrition consultant.

Please direct queries to the corresponding author: Diane E. Holland, 54 Robinson Road, Lexington, MA, 02420, USA; e-mail: diane_e_holland@yahoo.com.

The primary author (Johnecheck) was involved in the inception and implementation of both the National Surveillance System Pilot and the National Risk and Vulnerability Assessment in her capacity as Livelihoods Advisor with Save the Children US, and later as a Consultant with UNDP as Advisor to the Ministry of Rural Rehabilitation and Development. She also framed and carried out further analysis as a consultant with the Feinstein International Center (formerly named The Feinstein International Famine Center). The secondary author (Holland) provided logistical support and technical feedback to the primary author in her role as Assistant Program Manager of Capacity Development in Public Nutrition and contributed to the discussion and overall document in her capacity as a consultant with the Feinstein International Center. Further details can be found in Johnecheck W, Holland D. Nutritional risk in Afghanistan: Evidence from the NSS Pilot Study (2003-2004) and NRVA 2003. Medford, Mass, USA: Feinstein International Center, Tufts University, 2005.

Introduction

Government agencies and public health professionals have devoted a significant amount of work since 2000 toward understanding the nutrition situation in Afghanistan. These efforts have focused on identifying nutritional outcomes in their various forms and patterns throughout the country, as well as constructing a picture of the underlying causes of malnutrition. Early quantitative efforts provided localized insights into nutrition status; however, the National Surveillance System (NSS) Pilot Study (2003–2004) and the National Risk and Vulnerability Assessment (NRVA) 2003 attempted to develop national-level information for a variety of nutritional and related statistics.

The primary objective of this paper is to present findings from the NSS Pilot Study (2003–2004) and the NRVA 2003, with nutrition as the outcome of interest. Both of these studies were carried out with the aim of further developing an understanding of livelihoods in Afghanistan in relation to particular outcomes of interest: vulnerability to poverty, food insecurity, and malnutrition and the risks associated with these particular outcomes.

The national-level NRVA 2003 was conducted by the World Food Program (WFP), government ministries, and nongovernmental organizations and covered rural populations in every province [1]. It grew out of the annual food needs assessments conducted since 1999 by WFP and government ministries and was used to guide their spatial and temporal allocation of food aid throughout the country. The NRVA 2003 signaled a significant shift by demonstrating increased governmental ownership of national food security, poverty vulnerability, and data collection, as well as the introduction of data collection methods generally used in more stable contexts for developing longer-term policy objectives to complement rapid assessment techniques used during emergencies.

The NSS was carried out by three nongovernmental organizations (Save the Children US, Action Contre La Faim, and GOAL Ireland) and provincial ministries, piloted in seven provinces [2]. This data collection system was developed through consultation between government ministries and other stakeholder agencies in May 2002, based on a health center surveillance system that monitors trends in disease occurrence. The NSS was designed to systematically collect information on livelihood strategies and shocks, coping strategies, food security perceptions, and a variety of nutritional risk factors, to be interpreted in conjunction with agro-meteorological and price information, in order to monitor trends in key indicators and predict early signs of change and deterioration in livelihoods, food security, and nutrition. The NSS research activities included two rounds of data collection in the fall of 2003 and spring of 2004.

Unlike other analyses of the NSS and NRVA data,* this paper aims to present the findings with nutrition as the primary outcome of interest, in order to explicitly look at the relationship between direct and indirect causes (**fig. 3**). Nutritional status is closely associated with childhood mortality, early childhood cognitive development, immune functions and growth, and ultimately with higher productivity and its associated economic ramifications. Although policies and programs that promote economic growth, food security, and poverty reduction may lead to improved nutritional outcomes, experience has shown that explicit consideration of nutritional outcomes is needed in order to ensure that these policies and interventions equate to adequate gains in cognitive development and other functions directly related to nutritional status. The analytic framework used by the authors presents nutritional outcomes and determinants of nutritional status in Afghanistan, with the objectives of providing public health professionals a perspective on the causes of nutritional deficiencies and providing professionals engaged in infrastructure development, economic growth, and livelihood security with an understanding of their direct impact on nutritional outcomes.

Methods

Study setting and design: NSS

Two cross-sectional surveys were conducted in sentinel sites in eight provinces and 22 districts: 30 sentinel sites in November–December 2003 and 26 sentinel sites in May–June 2004 (**fig. 1**). Sentinel sites were purposively selected to reflect the majority of the communities in a given area (livelihood/agro-ecological zone) with respect to agro-ecological features, economic activities, available services, and demographics in areas where the nongovernmental organization piloting agencies had ongoing activities. Within each sentinel site, 20 to 40 households were randomly selected based on the population of the sentinel site. A subset of households changed between the fall and spring rounds. All individuals in each household selected were included in the sample. A total of 911 and 910 households (5,936 and 5,935 individuals) were sampled in the fall 2003 and spring 2004 rounds of the NSS.

Data collection: NSS

Surveyors used community and household questionnaires to collect data on food security (e.g., characterization of market access, market prices of staple goods, dietary diversity, crop production and perception of

* Other reports can be found at http://www.mrrd.gov.af/vau/NRVA_2003.htm

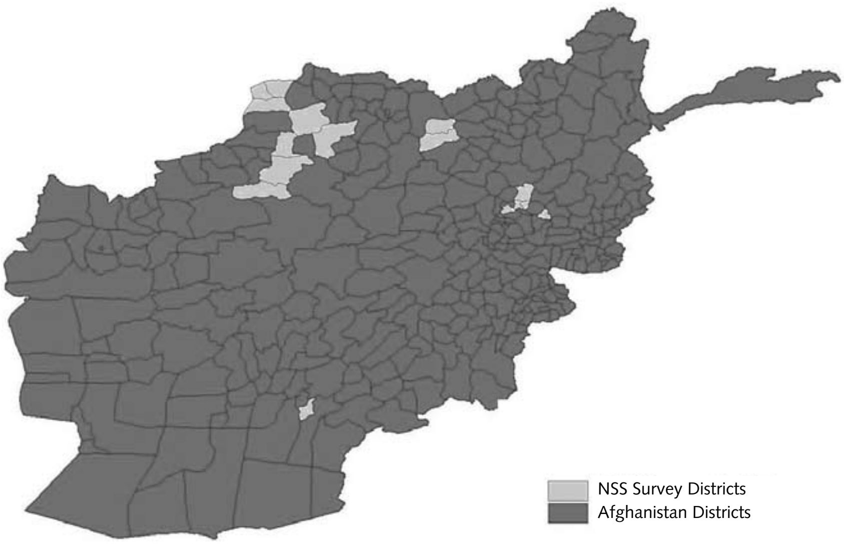

FIG. 1. Map of NSS coverage. Data from Afghanistan Information Management Service (AIMS) 2003. Produced by Tufts University 2005

food security), livelihoods (income sources, levels, shocks, and coping strategies), as well as risks to lives (morbidity and mortality data, anthropometric measures of children 6 to 59 months of age and women of reproductive age from 15 to 49 years), access to health services, iodized salt, and adequate water and sanitation). The questionnaire had a standard introduction explaining the purpose of the survey and request for informed consent before the administration of the survey.

Data analysis: NSS

Data from only 26 sentinel sites are reported in order to allow for direct comparison between seasons. This paper presents aggregated data from households and individuals in all 26 sites in order to gain an understanding of nutritional outcomes in relation to direct and underlying causes of malnutrition in a broader context than in one sentinel site or its corresponding livelihood zone. The aggregated information, however, is not statistically representative of any region, group, or subgroup in the population, and should not be used for prevalence estimates.

Study setting and design: NRVA

Households were selected from all 32 provinces and the majority of districts using a two-stage sampling procedure, based on identification of agro-ecological zones, from which two or three communities were purposively selected from within each zone. Within each community, focus groups of village leaders (known as the *shura*) identified households for the household-level questionnaire. These households were purposively selected to represent a particular socioeconomic group status. All individuals within a selected household were included in the sample. A total of 11,227 households (85,577 individuals) were sampled in the 2003 NRVA.

Data collection: NRVA

Data were collected by household-level and district-level questionnaires that gathered a range of information, including market access, productive capacity, market price data, household education, employment, assets, shocks, perceptions of food security, productive capacity, and dietary diversity.

Data collection occurred in July through September 2003. Eleven of 375 districts were not included in the survey because they were inaccessible due to insecurity (**fig. 2**). The questionnaire had a standard introduction explaining the purpose of the survey and request for informed consent before the administration of the survey.

Data analysis: NRVA

The findings presented were weighted so that households selected to represent a particular socioeconomic group adequately reflected the proportion of the population falling within each group at the community level. This same correction could not be applied at the district or provincial level due to lack of population estimates at those levels, and as a result national and provincial estimates must be used with caution.

Data from the NSS and NRVA were analyzed within the context of the conceptual framework of malnutri-

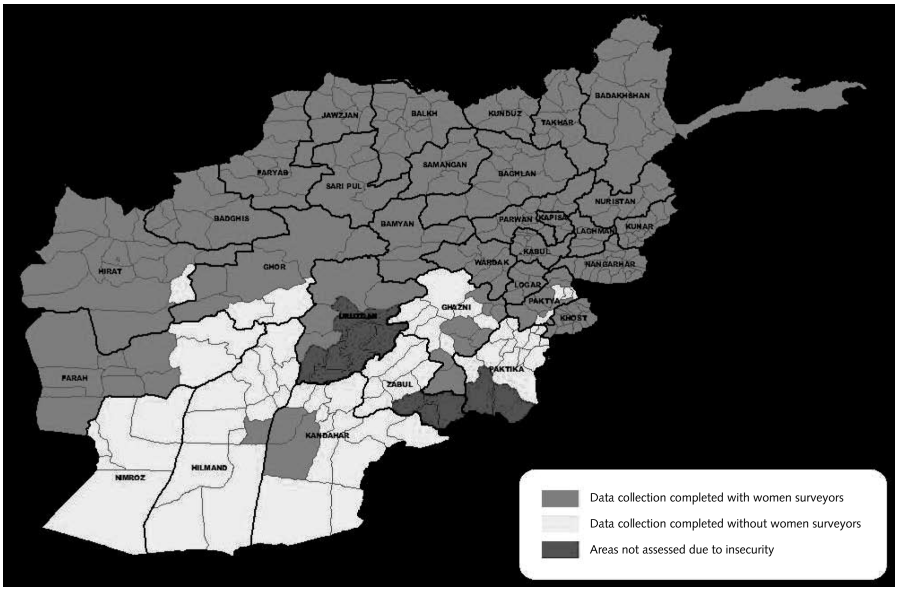

FIG. 2. Map of NRVA 2003 coverage according to province. The boundaries and names on the map do not imply official endorsement or acceptance by the United Nations World Food Programme Afghanistan Vulnerability Analysis and Mapping (VAM) Unit, 21-10-03. Source: World Food Programme VAM Supervisory Team

tion developed by UNICEF, and interpretation was complemented by comparing data with existing nutrition information from other surveys. **Figure 3** identifies the indicators that are presented in this article.

Results

Nutritional outcomes

Acute malnutrition (wasting), a measure associated with recent weight loss for children under five, was below alert or emergency levels defined by SPHERE and the Afghan Ministry of Public Health (MOPH) (less than 10% global acute malnutrition [weight-for-height < –2 z-scores for children 6 to 59 months old] in the absence of aggravating factors, e.g., insecurity or population displacement). The aggregated information from the NSS found acute malnutrition levels of around 8% (< 80% percent of median weight-for-height, 6–59 months of age) in children during the fall round and 9% in children during the spring round (**table 1**). The percentage of children 12 to 59 months old with mid-upper-arm circumference (MUAC) measurements < 13.5 cm was 11% during the fall round and 13% during the subsequent spring round.

In both the fall and spring rounds of NSS data, younger children (6 to 29 months old) were almost three times more likely to exhibit acute malnutrition than older children (30 to 59 months). Socioeconomic status was not significantly correlated with malnutrition. Households from the middle socioeconomic group had the poorest acute nutritional outcomes (e.g., the lowest average percentage of the median) in both rounds of data collection relative to both high- and low-income groups.

TABLE 1. Prevalence of acute malnutrition according to season and age in the National Surveillance System (NSS) Pilot Study[a]

Season and age	Severe malnutrition	Moderate malnutrition	Total
Fall 2003 (*n* = 793)			
6–29 mo	1.89	4.29	6.18
30–59 mo	1.01	1.26	2.27
6–59 mo	2.90	5.55	8.45
Spring 2004 (*n* = 545)			
6–29 mo	3.30	2.94	6.42
30–59 mo	0.55	2.02	2.75
6–59 mo	3.85	4.95	9.17

a. Prevalence values are given as percentages. Severe malnutrition is defined as a weight-for-height < 70% of the median together with edema, and moderate malnutrition as a weight-for-height between ≥ 70% and < 80% of the median.

Although chronic malnutrition was not an explicit outcome of interest for the NSS or the NRVA, this information was calculated from collected age and height data for children 24 to 59 months of age in the fall round of the NSS. The levels of chronic malnutrition or stunting (height-for-age < –2 z-scores) indicated a problem of public health importance, as 64% of children sampled were severely stunted [3]. Interpretation of these figures (**table 2**) must be done with caution,

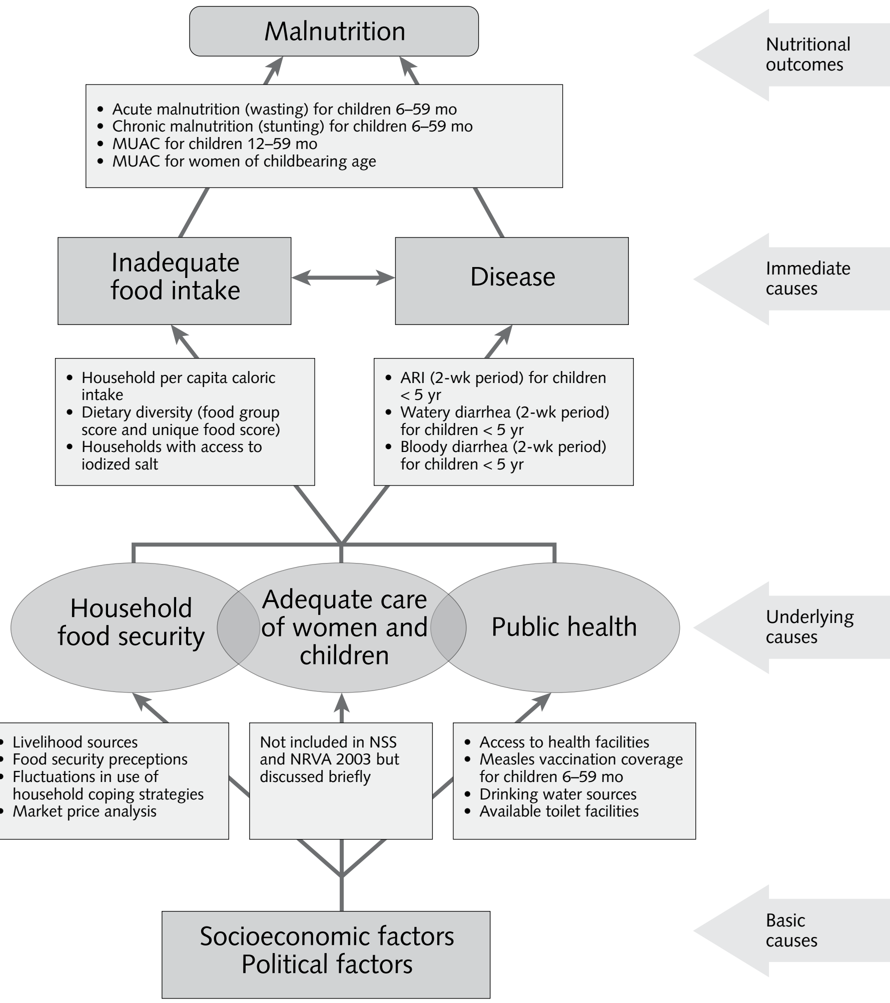

FIG. 3. NSS and NRVA data analysis arranged by conceptual framework for underlying determinants of malnutrition. MUAC, mid-upper-arm circumference; ARI, acute respiratory infection

since international standards are based on figures that include 6- to 23-month-old children. Older children, who are more likely to manifest the cumulative growth loss relative to younger children, were overrepresented relative to younger children in this prevalence figure.

Nutritional risk for women of reproductive age (15 to 49 years) measured by MUAC indicated that just under one-fourth of women were at risk for malnutrition according to a cut-off of 23.5 cm **(table 3)**. The more conservative cut-off of 21.0 cm identified around 5% of the women in this age group as at risk of malnutrition. These figures were consistent during both seasons and were not associated with socioeconomic class.

TABLE 2. Prevalence of chronic malnutrition among children 24 to 59 months of age as indicated by the prevalence of stunting in the National Surveillance System (NSS) Pilot Study, fall 2003

Degree of stunting	Prevalence (%) (n = 618)
None (HAZ ≥ –1)	17.2
Mild (HAZ ≥ –2 and < –1)	18.9
Moderate (HAZ ≥ – 3 and < –2)	23.3
Severe (HAZ < –3)	40.6

HAZ, height-for-age z-score

TABLE 3. Mid-upper-arm circumference (MUAC) measurements in women of childbearing age in the National Surveillance System (NSS) Pilot Study

MUAC	% of women	
	Fall 2003 (n = 1,122)	Spring 2004 (n = 1,178)
< 23.5 and ≥ 21 cm	22	24
< 21.0 cm	4	5

Immediate determinants of malnutrition

Morbidity

Almost one-third of children under five years sampled in the NSS had experienced an episode of watery diarrhea or acute respiratory infection (ARI) in the 2-week reference period (**table 4**). Five percent of children reported symptoms of bloody diarrhea in the 2-week reference period. The results were consistent in both the fall and spring round of data collection.

Dietary intake

Diet quality, as discussed here, refers to nutrient adequacy, or the intake of a diet that meets both caloric and essential nutrient requirements. These requirements are in turn related to dietary quantity and diversity (the number of different foods or food groups consumed over a given reference period). Caloric intake was below recommended age- and sex-adjusted levels (2,100 kcal) for about 20% of households sampled in the NRVA. Caloric intake was calculated by using a 7-day dietary recall for 64 individual foods, with consumed weights translated into caloric intake by FAO Food Composition Tables for the Near East, and then standardized for partial waste and refuse [4]. About 90% of middle or better-off households consumed more than 2,100 kcal/capita/day, whereas 75% of poor households and 62% of very poor households consumed more than 2,100 kcal/capita/day [5]. The NRVA data revealed a caloric reliance on wheat in general as a staple food (68% of mean kilocalories per capita per day came from cereals and 58% from wheat alone), which was more pronounced in poorer households, with 75% from cereals [6].

The NRVA results showed that increased household dietary diversity was positively associated with increased mean household energy intake (sex- and age-adjusted kilocalories), but the relationship was weaker than that reported in many other countries [7]. An analysis of 10 countries showed that all had a Spearman correlation coefficient of 0.20 or higher except for India (2002). The Afghan results were statistically significant but only showed an association of 10% (Spearman correlation coefficient, 0.1047; $p < .000$) [7]. In general, households with higher caloric intake per capita have higher dietary diversity, although households with low dietary diversity exhibit a broad range of energy intakes (data not shown).

In both studies, dietary diversity was positively associated with socioeconomic status, as the mean dietary diversity score increased with each increase in socioeconomic group (**table 5**). Dietary diversity was calculated by a 7-day recall of intake from nine different food groups for the NSS, and by categorizing responses from the 7-day recall on intake of 64 individual foods into the same nine food groups in the case of NRVA data. According to the NSS data, dietary diversity also exhibited a seasonal pattern, with increased mean dietary diversity in the spring relative to the fall (mean food score of 3.81 food groups per week in the spring, as opposed to 3.01 in the fall). Fruit consumption in the spring round was lower than in the fall round, while meat consumption remained stable, and consumption of dairy products and members of the "other vegetables" group more than doubled in May and June relative to November and December.

Table 6 shows that consumption patterns of specific foods varied between socioeconomic status groups. On average, better-off households ate meat at least once per week, whereas poor households consumed meat once a month or less. The number of households consuming fresh fruit, which provides essential nutrients such as vitamin C that are needed to prevent micronutrient deficiency diseases, increased with socioeconomic status. Consumption of all other food groups, except for pulses and nuts in the spring round of the NSS, demonstrated a similar trend to that of meat and fruit,

TABLE 4. Morbidity patterns in the National Surveillance System (NSS) Pilot Study

Condition and age group	% (*n*)	
	Fall 2003	Spring 2004
Watery diarrhea in past 2 wk, 0–59 mo	32 (920)	36 (826)
Bloody diarrhea in past 2 wk, 0–59 mo	5 (920)	5 (826)
Acute respiratory infection in past 2 wk, 0–59 mo	34 (917)	34 (812)
Measles vaccination, 6–59 mo	58 (805)	57 (741)

TABLE 5. Mean dietary diversity score according to household socioeconomic status in the National Surveillance System (NSS) Pilot Study and the National Risk and Vulnerability Assessment (NRVA)[a]

Socioeconomic status	NSS fall 2003 (*n* = 911)	NSS spring 2004 (*n* = 903)	NRVA 2003 (*n* = 11,227)
Very low	—[b]	—[b]	5.7
Low	3.00	3.46	6.3
Medium	3.34	4.21	7.1
High	4.36	5.01	—
All households	3.01	3.81	6.6

a. The dietary diversity score is the number of different food groups from which items are consumed at least once a week by a household. The nine food groups are meat, cereals, oils and fats, eggs, pulses and nuts, dairy products, green leafy vegetables, other vegetables, and fruit.

b. indicates that the NSS and NRVA used two different scales (NSS has low, medium, high, and all, but the NRVA has very low, low, medium and all); the dash is placed in the unused category in order to put the scales into the same table to see the trend with increasing socioeconomic status. The difference between NSS and NRVA data was not tested statistically.

with higher-income groups having a higher percentage of households consuming food items from each particular food group. It is possible that pulses were substituted for grains preceding the harvest if grain supplies were exhausted. Consumption of green leafy vegetables (spinach, wild greens, coriander, and mint) was low (19% to 29%), irrespective of socioeconomic status or seasonality.

The areas with the lowest levels of mean dietary diversity as identified by the NRVA data were also areas where symptoms of micronutrient deficiency diseases have been reported in the past (**fig. 4**). Areas with the highest mean dietary diversity scores were large food production centers. Reports of scurvy during the end of the drought suggest that these areas identified with poor dietary diversity are at particularly extreme nutritional risk during consecutive years of poor production [8, 9].

Improvements in caloric intake, but not necessarily dietary diversity, were linked to market access and infrastructure development. Mean household caloric intake decreased with increased distance to markets, suggesting a direct association between infrastructure development and nutritional outcomes (data not shown). However, diet quality (e.g., dietary diversity) did not necessarily improve with better access to markets. The unique food score from NRVA data, indicating the variety of foods consumed by households, decreased slightly with increasing distance to markets, but this relationship did not hold for food group scores from the same dataset. The dietary diversity score from the NSS decreased with increasing distance to permanent markets, and the decrease in dietary diversity score with increasing distance was more apparent in the spring relative to the fall.

Household use of iodized salt increased between the fall and spring rounds of the NSS, and on the basis of community- and market-level qualitative work this change was thought to relate primarily to increased market availability as the increased production of iodized salt through the national Universal Salt Iodization Program began to reach more remote markets.

Underlying determinants of malnutrition

Livelihoods and food security

Livelihoods in Afghanistan appear to be quite diverse but overall are highly dependent on agriculture through either direct production or wage labor. Most households sampled by the NSS reported three distinct sources of income, with income diversity increasing as socioeconomic status improved. Although affluent households in general invested in a variety of assets and engaged in different income-earning activities (making them less vulnerable to covariate shocks), they did not always have a larger number of income sources. In some NSS sentinel sites, low-income households on average had a larger variety of income sources than households classified in the medium socioeconomic status category. Households with no or limited assets in terms of land, livestock, shops, rental cars, etc. were highly reliant on labor as a source of income. Among poor households, those with fewer able-bodied workers were found to be more vulnerable to food insecurity

TABLE 6. Percentage of households consuming items from each food group at least once per week according to socioeconomic status in the National Surveillance System (NSS) Pilot Study and the National Risk and Vulnerability Assessment (NRVA)

Socioeconomic status	Cereals[a]	Oils and fats[a]	Meat	Dairy products	Eggs	Pulses and nuts	Green leafy vegetables	Other vegetables	Fruits
Fall NSS									
High	—	—	58	46	41	41	29	68	58
Medium	—	—	30	30	32	34	28	42	38
Low	—	—	15	17	21	31	19	31	25
All households			24	24	26	33	23	38	32
Spring NSS									
High	—	—	60	92	47	49	23	78	43
Medium	—	—	39	66	41	54	26	71	25
Low	—	—	20	40	30	51	23	65	16
All households	—	—	29	52	35	52	24	68	21
NRVA 2003									
Medium-high	100	95	82	84	70	82	24	99	72
Low	100	94	49	69	59	77	25	98	52
Very low	100	94	29	57	53	70	23	97	43
All households	100	95	63	75	64	79	24	98	61

a. Cereals and oils and fats were not included in the NSS questionnaire, because it assumed that all households consumed items from these food groups at least once a week. The NRVA results verified this assumption: 100% of households consumed cereals weekly and 95% consumed oils and fats weekly.

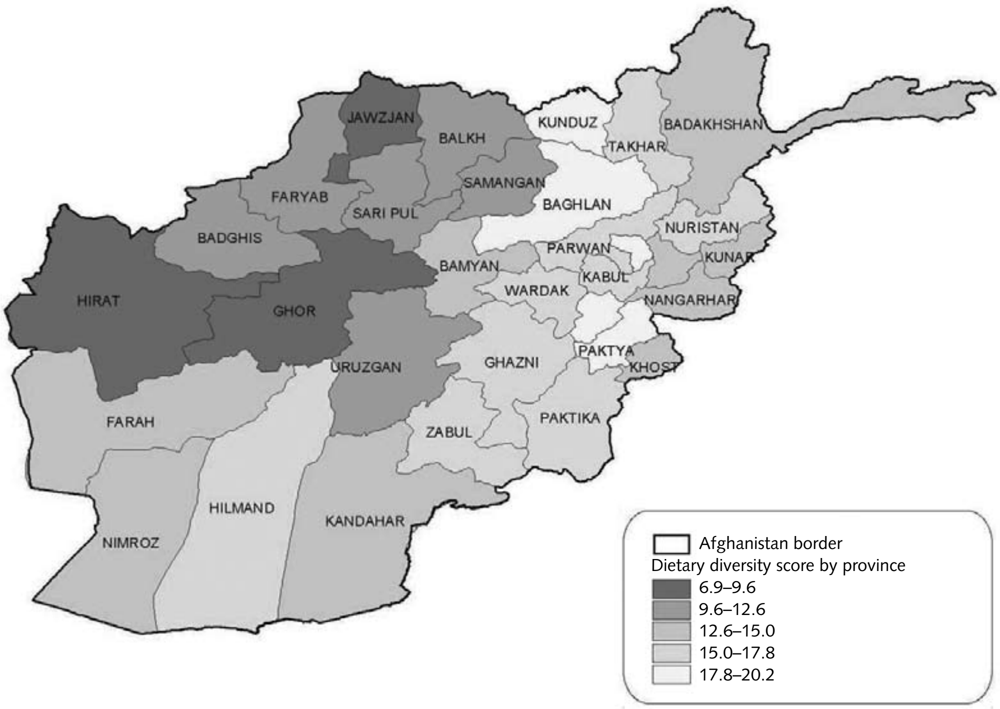

FIG. 4. Map of dietary diversity according to province (NRVA). Data from Afghanistan Information Management Service (AIMS) 2003 and NRVA 2004. Produced by the Feinstein International Famine Center 2005. The boundaries and names on the map do not imply official endorsement or acceptance by the United Nations World Food Programme Afghanistan VAM Unit 21-10-03

and poverty than those with greater labor capacity.

Agriculture provided income for a large proportion of the households sampled, but the importance and form of this income source varied seasonally. More households earned some income from their own production in the fall than in the spring (70% of households in the 4-month period prior to the fall vs. just over 40% for the 6-month period prior to the spring). A larger percentage of households also listed agricultural production as their primary income source in the fall than in the spring (30% vs. 4%). Engaging in agricultural labor as an income source was more prevalent in the spring than in the fall round of data collection (almost 50% in the fall and 60% in the spring) as well. In contrast, income from livestock production was stable between seasons, but this was a primary income source for a limited number of households (9% and 15% of households in the fall and spring rounds, respectively). The proportion of households that earned income from nonagricultural daily labor (20%) was consistent between both rounds of data collection.

Although men were the primary income earners, women did contribute to household economy beyond their role in the domestic arena. The percentage of households with women engaged in agricultural activities, weaving carpets, performing nonagricultural day labor, taking out loans, or performing activities related to accumulation of resources without productive assets, such as receiving gifts of resources or tithes, or gleaning crops, varied by season. Women were more involved in carpet weaving during November and December than in May and June when they spent more of their time working in agriculture and in nonagricultural day labor.

For the most part, increased ownership of livelihood assets such as land, cattle, and poultry was correlated with higher household caloric intake (data not shown) [10]. However, own production of irrigated and/or rain-fed wheat in 2003 did not show any significant relationship with caloric intake or dietary diversity. Thus, although land ownership signifies purchasing power and wealth, own production of crops did not directly yield diets that are more adequate in either quality or quantity. Households were more likely to consume specific foods if they also owned the food production asset (e.g., households that owned chickens consumed more eggs), with the exception of vegetable plots (**table 7**). In the case of goats and cows, which represent a higher socioeconomic standing, increased consumption was interpreted as indicative of higher purchasing power rather than direct consumption of assets.

Regarding perceptions of food security, around

TABLE 7. Relationship between consumption of items from particular food groups and household ownership of corresponding livelihood assets in the National Risk and Vulnerability Assessment (NRVA)

Food group	Livelihood asset	Weekly consumption (% of households)		p^a
		Asset owners	Asset nonowners	
Meat (beef)	Cows	38	32	< .001
Meat (goat)	Goats	7	4	< .001
Dairy products	Cows	74	70	< .001
Egg	Chickens	64	50	< .001
Fruit	Fruit orchard	67	55	< .001
Green leafy vegetables	Vegetable plots	17	24	.135
Other vegetables	Vegetable plots	99	98	.509

a. Pearson chi-square test.

30% of households did not find their food situation to be sufficient (in both quantity and quality) during both rounds of the NSS survey. Poor households were more likely than better-off households to report an insufficient food security situation. More households sampled in the fall reported that they always or often worried about where their food was coming from than households sampled in the spring (30% in the fall vs. 20% in the spring). By late spring, households had access to other sources of income, such as agricultural day labor, and therefore households were able to have consistent flows of income during this period. Still, around 10% of households in both rounds clearly thought that their situation was likely to be worse in the upcoming months. The NRVA data showed a similar trend and reported a high correlation between the frequency of household "worry" and asset ownership and socioeconomic status, wherein "worry" increased with decreasing socioeconomic status [5].

Coping strategies (defined based on extensive qualitative work by NSS pilot nongovernmental organizations and the WFP) employed at the time of the surveys did not appear to present a significant threat to livelihoods. Few households relied on erosive coping behaviors such as asset liquidation and the marrying of daughters at an early age [10]. More households received loans in the spring than in the fall (45% vs. 28%). However, many of these loans were obtained to purchase assets such as livestock, plows, and seeds, to rebuild dilapidated compounds, and to pay for weddings. In both rounds, households from all socioeconomic groups obtained loans, but the middle socioeconomic group had the highest percentage of households obtaining loans.

A direct link between poor nutritional outcomes (acute and chronic malnutrition) and higher wheat prices has not been established in Afghanistan. Because of the poor infrastructure and the different levels of production and imports in the various regions of Afghanistan, prices of the main cereal products vary substantially throughout the country, creating a situation where high food prices do not necessarily reflect a shortage of food. District-level wheat prices did not correlate well with mean caloric intake or dietary diversity aggregated at the district level. Wheat prices were lowest in areas of surplus production.

Public health environment

Access to health care and water continues to be limited for the rural Afghan population. According to the NRVA survey, 35% of households reported taking more than a half-day walk to reach a health facility [5]. Both the NSS and the NRVA found that the main sources of water were rivers, lakes, and canals. Data on household water use did not differentiate between clean and contaminated sources. Detecting contamination requires testing the water and is therefore rarely done in surveys that are not solely focused on water issues. Qualitative work from the NSS revealed that time for water collection and the limited amount of water for personal hygiene were specific concerns of those responsible (women) for water in the household.

Measles vaccination coverage rates were consistently below the government target of 80%, and on average actual measles coverage (delivered through the routine Expanded Program for Immunization for children under five) reported in the NSS was similar to the government-reported figures for the entire country (58% NSS vs. 50% MOPH in 2003, 57% NSS vs. 61% MOPH in 2004). In some areas, however, measles vaccinations fell below the local coverage rates [11].

Adequate care of women and children

Neither the NSS nor the NRVA included the caregiving behaviors usually associated with poor nutritional outcomes in their data collection.

Discussion and recommendations

This section presents data constraints and explores each indicator of underlying causes in order to analyze the relative impact of each factor on nutritional outcomes, and discusses the relevance of each indicator to individual outcomes of interest. This section also highlights areas where evidence from the NSS and NRVA 2003 should be considered during policy and program planning in order to strengthen future nutrition and nutrition-related policies.

Data interpretation and limitations

Interpretation of data from the NSS must be done in light of the fact that all of the sites included in the NSS analysis were located in the northern part of the country. Additionally, collection of NSS data in the spring round was delayed, so that the data might not adequately reflect the time of year when households had the greatest vulnerability to poor nutrition and food insecurity (specifically, in areas of low altitude where the crop was harvested as early as June due to unseasonably warm weather over the winter). Additionally, the survey occurred after a bumper harvest in 2003 (therefore a time of relative abundance compared with previous years) when livelihoods seemed to be improving after the drought. It was likely that both of the field visits occurred at a time of rebuilding and relative abundance for most of the communities interviewed.

Although the NRVA covered the entire country, the lack of a reliable sampling frame limits the generalizability of the findings to the entire Afghan population. Furthermore, in many parts of the country, particularly in the surplus production regions, 2002 also represented a better than normal harvest. Therefore, the food security and nutrition situation at the time of the survey was at the best possible time (post-harvest) for an agrarian society, in addition to occurring after two consecutive better than usual harvests.

Finally, this is a secondary analysis of data that were collected to meet the information needs of a variety of stakeholders who would subsequently analyze the data with their own outcomes of interest. It is recognized that the ideal would be a survey designed with the ability to statistically quantify the relative contributions of underlying causes. This analysis does make links where possible, given data constraints, thus providing potential indicators appropriate for a subsequent study with that aim.

Nutritional outcomes

The low levels of acute malnutrition (less than 10% < -2 weight-for-height z-scores for children aged 6 to 59 months) were consistent with existing nutrition data. Weight-for-height is an anthropometric measure of wasting, indicating recent weight changes. Various subnational-level surveys conducted since 2000 consistently reported a prevalence of acute malnutrition between 6% and 10% [12], and the 2004 National Nutrition Survey reported a prevalence of 6.7% [13]. The anticipated seasonal increase in malnutrition as a result of sharply decreased food security prior to the summer harvest was confirmed, due, potentially, to the delayed timing of the second round of NSS data collection (late enough in the spring for wild plants to be available and rural access to markets to improve).

The higher level of acute malnutrition found in younger children (three times as high for those aged 6 to 29 months as for those aged 30 to 59 months) is also consistent with the results of previous surveys. In these other studies, the relative risk of acute malnutrition was 2.0 to 7.31 for younger children compared with older children, suggesting that infant and young child feeding practices, specifically introduction of complementary foods, were the primary cause of acute malnutrition in the population [14–17]. The one nutrition survey that did not find a significant difference between those aged 6 to 29 months and those aged 30 to 59 months was conducted in an encashment center for returnees whose areas of origin and exposures to underlying causes are likely to have differed greatly [18]. The lack of association between socioeconomic status and average levels of acute malnutrition in the NSS data provides further evidence that socioeconomic status and dietary intake were not the only determinants of wasting in children under five years of age. Often acute malnutrition is worse in upper-income groups in developing countries because of the use of bottle-feeding in conjunction with poor hygiene practices, but this was not the case in these rural communities.

Efforts aimed at reduction of acute malnutrition in children under five years of age and its associated risk of high mortality, a primary objective of nutrition policy in Afghanistan, should concentrate on nutrition and health education campaigns that specifically address caregiving behaviors and hygiene issues (food safety, safe access to water, use of toilets) directly linked to the poor nutritional outcomes in this at-risk age group. The Afghanistan Ministry of Public Health Public Nutrition Policy and Strategy 2003–2006 is: “Strategy 1: Ensure that the prevalence of acute malnutrition or wasting (< -2 z-scores, weight for height) is reduced to and remains below 5% for all children less than five years old” [19]. These campaigns are not limited to one sector, and therefore relevant and consistent messages need to be incorporated into campaigns falling under various ministry mandates (e.g., MRRD Rural Water and Sanitation Department and Ministry of Woman’s Affairs Health Education activities).

It is well acknowledged among public health professionals that the level of stunting, a measure of chronic malnutrition, is of greater public health concern in Afghanistan than acute malnutrition at the national level, a position supported by the current data (64% of children 24 to 59 months of age had height-for-age < –2 z-scores). Earlier local-level surveys reported chronic malnutrition rates ranging between 45% and 59% (< –2 z-scores, height-for-age) in both urban and rural areas [12]. The 2004 National Nutrition Survey reported that 54% of children between the ages of 6 and 59 months were chronically malnourished (< –2 z-scores, height-for-age) at the national level [13]. Furthermore, 37% of children exhibited some level of stunting by 12 months of age, with the prevalence increasing in the second and third year of life [13].

Efforts aimed at reduction of chronic malnutrition require a multipronged strategy to overcome recurring disease (e.g., diarrhea and ARI) and reduced food intake, which are likely primary drivers of chronic malnutrition. This strategy would incorporate economic growth coupled with improved household livelihood security outcomes, social protection program coverage, and household access to public health services.

Maternal nutritional risk according to a conservative cut-off was slightly lower than that reported by other surveys (5% vs. 3% to 19% of women of childbearing age had MUAC < 21.0 cm) [14, 20–22]. The persistence of maternal nutrition risk, regardless of season or socioeconomic class, however, suggests that steps need to be taken to address the situation for Afghan women of childbearing age in the context of a more thorough understanding of the cross-cutting and specific risks.

Immediate determinants of malnutrition

Morbidity

Infectious disease reduces optimal absorption of nutrients into the body and depresses the appetite, negatively influencing nutritional outcomes, and appears to be widespread in Afghanistan throughout the year. The prevalence rates of watery diarrhea in children 6 to 59 months of age within a 2-week period found in the NSS (32% fall, 36% spring) were consistent with other national-level data. The Multiple Indicator Cluster Survey (MICS) carried out by UNICEF in 2003 reported a 30% prevalence of diarrhea within the past 15 days, and the 2004 National Nutrition Survey found a 46% prevalence of diarrhea within a 2-week period in children 6 to 59 months of age [13, 23]. Nutrition surveys carried out in Kabul have linked increases in acute malnutrition in children under 5 years of age to seasonal peaks in diarrheal disease [12, 20].

The prevalence of ARI during a 2-week period in children 6 to 59 months of age was consistently 34% in both the fall and the spring rounds of the NSS, suggesting that both fall and late spring have concerning, but similar, levels of disease. Nineteen percent of children under five sampled in the MICS survey reported an ARI in the previous 2 weeks, and 27% of children under five reported fever and/or cough within the previous 2 weeks [23].

It is likely that the consistently elevated prevalence of morbidity (specifically diarrhea and ARI) influences the prevalence of both acute and chronic malnutrition in children under five. Given the sustained prevalence of infection and the physiological links between repeated insults to the body as a result of illness and chronic malnutrition, it is necessary to more fully characterize the contributory role that formal health care, as well as adequate potable water for food preparation and sanitation, may play in addressing chronic malnutrition, relative to other underlying causes.

Dietary intake

Dietary intake in terms of caloric intake and dietary diversity is often considered to be a strong proxy for nutritional status, even though it is one of the immediate causes of malnutrition (**fig. 3**). The actual percentage of households with caloric intake of less than the recommended intake of 2,100 kcal may be higher than the 20% overall reported in the NRVA, due to the improved harvest and methodological constraints that did not account for guest consumption (in a culture in which guests are common) or intrahousehold distribution (which is thought to exhibit a gender bias against females). Additionally, even though most households were able to attain adequate calories, the higher percentage of poorer households not meeting their per capita caloric needs and the heavy reliance on cereals, and wheat in particular, suggest that adequate amounts of essential nutrients may be differentially obtained according to socioeconomic status.

The weak relationship between increased dietary diversity and caloric adequacy is masked (due to sample size) when means are looked at but becomes apparent when the dispersion of individual observations is taken into account. Therefore one must be cautious when assuming that dietary diversity reflects adequate caloric intake in Afghanistan and that the two measures have the potential to be used as a proxy for each other. Furthermore, it cannot be assumed that households consuming diets with adequate caloric intake are consuming nutritionally adequate diets.

Both caloric intake and dietary diversity were positively associated with socioeconomic status, suggesting that on average, households will increase their quality and quantity of intake if they have adequate purchasing power. Improvement in dietary diversity, however, will be influenced by the wider context of foods available by season, consumer preferences, and the function of rural livelihoods. Efforts to support general economic development and rural household livelihood security, as well as infrastructure development (e.g., improved access to markets), have an important role to play in improving dietary intake, bearing in mind the positive relationship found between caloric intake (but not necessarily dietary diversity) and improved market access. An increased understanding of the viability of subsistence agriculture household economies (e.g., poorer households practicing subsistence agriculture may not produce adequate food stocks for the year, and isolated circumstances support few other income-earning opportunities) would need to be incorporated into these efforts to ensure that interventions are appropriately designed to take into account shortfalls. Additionally, consumer preferences will need to be further characterized with a seasonal dimension, since the data at this time do not reveal why households able to obtain adequate calories may in fact have poor diet diversity.

The correlation between the occurrence of micronutrient deficiency diseases and low dietary diversity scores suggests that dietary diversity may be a potential marker for monitoring areas of nutritional risk and allocating programs directly aimed at improving nutritional adequacy, once the dietary diversity score is validated with nutrient adequacy and other measures. However, regular monitoring of areas of high risk for micronutrient deficiencies (indicated by low dietary diversity) and implementation of preventive measures may play a role in mitigating the negative effects of micronutrient deficiency diseases. Food-based approaches, however, may be limited, in particular with reference to the consumption of green leafy vegetables, regardless of socioeconomic group. Promotion of fruit production and consumption is one possible method of preventing certain micronutrient deficiency diseases, but further lesson learning is needed about the effectiveness and efficacy of food-based programming.

In the case of iodine-deficiency disorders, the improvement in household-level consumption of iodized salt over the course of a year is encouraging, but additional data from the 2004 National Nutrition Survey indicate that the overwhelming majority of the population have low (72% of children 7 to 11 years of age and 75% women of child-bearing age) or unacceptably low (50% of children 7 to 11 years of age and 58% of women of childbearing age) urinary iodine levels. The Universal Salt Iodization program will take many years of sustained and supported production and social marketing to make significant improvements in population-level health status [13].

Underlying determinants of malnutrition

Livelihoods and food security

Although rural livelihoods in Afghanistan are diverse, they are heavily dependent on agricultural activities as a direct source of food for consumption (ownership of poultry and goats, production of horticultural crops) and also as a means of income generation (agricultural labor, sale of produce), the importance of which changes seasonally [4, 10, 24].

Livelihoods in Afghanistan also have a gendered dimension, with both men (primarily) and women contributing to the household economy. It is critical to more fully understand the role of women in the household and formal economy in order to ensure that programs such as household income-earning activities or agricultural development are appropriately targeted and designed not to overburden household responsibilities, with a potential negative impact on time and energy available for adequate care of women and children.

Affluent households (for example, those with land, livestock, a shop, or a rental car) are more able to invest in and preserve their assets as well as to withstand covariate shocks, whereas poorer households are more reliant on labor as their means of income, which is in turn dependent on employment opportunities that can vary over time. Poor households with few able-bodied workers are therefore more vulnerable to food insecurity and poverty than those with more labor capacity, after accounting for similar work opportunities within the local area. The poorest communities were those that had limited work opportunities for all income groups and were living in remote areas with poor agricultural capacity.

The finding that ownership of specific assets (e.g., goats, cows, dairy cows, and poultry) was significantly associated with higher consumption may lead practitioners to consider prioritizing promotion of a diverse agriculture and animal husbandry base for rural households (for example, livestock restocking programs) as a means of improving household nutrition outcomes. Programming must be developed with the understanding that some assets, such as land, cattle, and goats, are more indicative of higher socioeconomic status and purchasing power, allowing households to purchase food commodities in the market rather than directly consuming or using their own assets. This suggests that market access facilitated by infrastructure development needs to be in place in order to convert this purchasing power into improved nutritional outcomes.

It is not clear that food-based approaches to increase consumption of dark-green leafy vegetables will be effective in Afghanistan, given that consumption was low across all socioeconomic groups throughout the year. This lack of association may be due to households selling their produce at the market, the produce not being ready for consumption during the 7-day period prior to data collection, or consumer preference, for example, the additional cost of time and resources (e.g., water) for preparation of green leafy vegetables. However, improvements in vegetable production may have positive indirect effects (rather than direct effects on consumption) through increased disposable income from the sale and subsequent purchase of other goods. Impact monitoring of agricultural support programs will need to take into account household utilization practices.

Perceptions of food insecurity by households indicate that not all households were benefiting from the improved economic situation in the country and that in fact, some households were actually worse off than during the previous year. Coping strategies seem to vary based on socioeconomic status, regional patterns, and idiosyncratic characteristics of the household [25]. Although most coping strategies have long-run adverse implications [10], it is unreasonable to expect that the newly formed government will be able to provide social protection to rural households in the short term to circumvent their use. Decreasing food consumption—the primary mechanism that households in Afghanistan use to cope with shocks (based on NRVA 2003 data), seasonal migration for labor, and taking out loans can have negative effects on human and household productivity [4, 10], but these actions will be better addressed by strategies promoting economic development in the mid to long term, and must be more fully understood (e.g., migration for labor or borrowing food is not always indicative of distress). A frequency aspect to utilization of coping strategies may need to be incorporated into monitoring systems. In the short term, it is essential to prioritize protection of households that have to rely on socially unacceptable risk management strategies or erosion of livelihood assets essential for income and survival in the long term.

Some of the indicators being proposed in Afghanistan for prioritization of social sectors resources are based on poverty (income) and food security measures (caloric intake, dietary diversity, and food security perceptions), and therefore it is necessary to understand whether these same measures will have a positive effect on nutritional status as well as reducing food insecurity. Caloric intake as the de facto definition of poverty is widely used in allocating program resources around the country. This definition of poverty needs to be expanded so that it reflects a more multifaceted definition of food security and vulnerability. Because caloric intake is not strongly correlated with dietary diversity, a first step in broadening the current definition would be to incorporate both dietary quantity and quality in future resource allocation efforts.

Adequate care of women and children

The disproportionate percentage of children between 6 and 29 months of age suffering from acute malnutrition, which is consistent with other available data, suggests that specific hygiene and caregiving behaviors are a primary cause of acute malnutrition in the younger age group. Quantitative and qualitative research on optimal infant and child feeding practices reveals that optimal feeding is rarely practiced in Afghanistan. For example, colostrum is often discarded, exclusive breastfeeding to 6 months is limited, and complementary foods, which are often inappropriate in terms of insufficient energy or nutrient density, are introduced early [26]. Cultural beliefs related to these practices, which need to be understood in order to effectively design programs and health education messages, are insufficiently characterized. Some research has begun to address this gap [12, 26].

Public health environment

Access to health facilities, clean drinking water, and use of sanitation facilities is extremely low in rural Afghanistan and is plausibly the most important factor in the high morbidity and mortality patterns present today. Efforts to reform health care services, in terms of increasing coverage and quality, have a strong policy foundation, but actual implementation and subsequent impact in rural areas will take years.

Water issues, in terms of quantity and quality for agricultural production as well as household use and personal hygiene, have always been a source of contention in Afghanistan. Many rural settlements and livelihoods have been built around access to water. The MICS 2003 data indicate that less than 40% of the population has access to safe water, suggesting that perhaps more than half are accessing unsafe water, which could be a contributor to the high morbidity and associated malnutrition seen in children under five years of age [23].

NSS and MICS data show that sanitation in rural communities is very basic in terms of sanitation facilities and personal hygiene practice, with nearly half of communities not having a latrine at all [23]. Additionally, personal practices related to hygiene are generally found to be insufficient (e.g., only 28% of people use soap and water after defecation [23] and only 16% of mothers of children under five wash their hands after defecation [23]), probably because of limited alternatives, cultural constraints, and low health literacy. Qualitative research from the NSS indicated a broad spectrum of understanding about the links between the spread of disease and poor nutrition and health outcomes among the NSS communities. When associated risks were understood, a range of actions were taken related to prioritization and ability to change practice. Continued efforts to improve access to adequate and safe water, as well as utilization practice related to personal hygiene, are needed.

Provision of adequate vaccination service is one role of government health services, aiding in the prevention of specific morbidities. Clinical symptoms of vitamin A deficiency, which is associated with a higher mortality risk for diseases (e.g., measles and diarrhea), have been reported in various areas of Afghanistan [17, 27, 28]. Therefore, improving coverage of national public health programs such as measles vaccination and those meant to improve vitamin A status (e.g., continuing supplementation campaigns in the short term while putting in place longer-term sustainable strategies) is essential for improving nutritional and mortality outcomes. Most likely, innovative approaches as well as regular monitoring of program coverage will be required in the case of remote, geographically isolated areas that naturally have limited access to health services.

Conclusions

In summary, these data suggest that although food insecurity does contribute to acute and chronic malnutrition, infrastructure development, improved access to health services, and efforts to increase health literacy and ameliorate infant and young child feeding practices may have a much larger role to play in Afghanistan in addressing population nutrition status. Therefore, improving nutritional status requires a multipronged approach, directly targeting malnutrition, coupled with economic growth, household livelihood security, social protection, access to public health services, and water and sanitation. Nutrition policy, programming, and monitoring needs to reflect the immediate and underlying causes of malnutrition.

Although there has been recent progress in understanding the nutritional situation as well as the immediate and underlying causes of malnutrition, the mechanisms by which they contribute, the relative importance of their contribution, and appropriate indicators for monitoring are not necessarily clear. Further work is required to elucidate the mechanisms and relative contribution of underlying causes to guide effective responses.

Acknowledgments

The authors would like to express their gratitude to both government and international agencies involved in collecting information with the National Surveillance System pilot work and the National Risk and Vulnerability Assessment 2003. A special note of appreciation goes to the two agencies that made the two assessments possible: the World Food Program (NRVA) and Save the Children/US (NSS). Financial support from the Feinstein International Center for the original analysis that forms the basis of this document is acknowledged.

References

1. Pinney A. National risk and vulnerability assessment 2003: A stakeholder-generated methodology. Kabul: Afghanistan Research and Evaluation Unit, 2004. Available at: http://www.mrrd.gov.af/vau/NRVA%2003%20Downloads/NRVA%202003%20Stakeholder%20methodology-AREU%20March%202004%20-%20Eng.pdf. Accessed 17 December 2006.
2. Johnecheck W. The evolution of food security information in Afghanistan: A case of limited availability, access and utilization. In: Pain A, ed. Light footprints at the crossroads: Learning from responses to food insecurity in Afghanistan? Rome and London: FAO & ITDG Publishing, 2006.
3. Kaplan Ramage A, Boyd E. Predicting childhood stunting and wasting in the transitional Islamic State of Afghanistan with 2003–2004 National Surveillance System Data. Boston: Tufts University, 2004.
4. Food and Agriculture Organization, U.S. Department of Agriculture. Food Composition Tables for the Near East. Rome: Food and Agriculture Organization, 1982. Available at: http://www.fao.org/docrep/003/X6879E/X6879E00.htm. Accessed 17 December 2006.
5. World Food Programme, Ministry of Rural Rehabilitation and Development. Report on findings from the 2003 National Risk and Vulnerability Assessment (NRVA) in rural Afghanistan. Kabul: MRRD, 2004. Available at: http://www.mrrd.gov.af/vau/NRVA%2003%20Downloads/NRVA%202003%20Livelihoods%20Report%20&%20Annexes%20Dec%2004.pdf. Accessed 17 December 2006.
6. USAID Famine Early Warning System Network, Ministry of Rural Rehabilitation and Development, Ministry of Agriculture and Animal Husbandry, Ministry of Health, World Food Programme, Food and Agriculture Organization. What are rural Afghans eating: Insights from the NRVA 2003. Available at: http://mrrd.gov.af/vau/NRVA%2003%20Downloads/AFSN%20Bulletin%20April%2004.pdf. Accessed 17 December 2006.
7. Hoddinott J, Yohannes Y. Dietary diversity as a household food security indicator. Washington, DC: Food and Nutrition Technical Assistance Project, Academy for Educational Development, 2002. Available at: http://www.fantaproject.org/downloads/pdfs/DietaryDiversity02.PDF. Accessed 17 December 2006.
8. Assefa F, Jabarkhil MZ, Salama P, Spiegel P. Malnutrition and mortality in Kohistan District, Afghanistan, April 2001. JAMA 2001;286:2723–8.
9. Action Contre la Faim. Nutrition anthropometric survey summary report, Sang Charak, Sar-e-Pul Province. Kabul: Action Contre la Faim, September 2002.
10. World Bank. Afghanistan poverty, vulnerability and social protection: An initial assessment. Human Development Unit. Report No. 29694-AF. Washington, DC: World Bank, 2005. Available at: http://www.mrrd.gov.af/vau/NRVA%2003%20Downloads/Afghanistan-poverty,%20vulnerability%20and%20social%20protection-an%20initial%20assessment%205%20March%2005%20WB.pdf. Accessed 17 December 2006.
11. Ministry of Rural Rehabilitation and Development, Ministry of Agriculture and Animal Husbandry, Ministry of Health, Save the Children–US. Tchachnyak Province: National Surveillance System. Kabul: MRRD, spring 2004.
12. Dufour C, Borrel A. Towards a public nutrition response in Afghanistan: Evolutions in nutritional assessment and response. In: Pain A, ed. Afghanistan in crisis? Rome:

Food and Agriculture Organization, 2005.
13. Ministry of Public Health of the Islamic Republic of Afghanistan, UNICEF, Centers for Disease Control and Prevention, National Institution for Research on Food and Nutrition, Tufts University. 2005. Summary Report of the National Nutrition Survey Afghanistan, 2004. Kabul: Ministry of Public Health of the Islamic Republic of Afghanistan/UNICEF, May 2005.
14. Action Contre la Faim. Nutrition and Mortality Survey, Panjsheer Valley–Shamali Plains, March–April 2002. Kabul: Action Contre la Faim, 2002.
15. Action Contre la Faim. Nutritional Anthropometric Survey, children under 5 years old, Mazar e Sharif, Afghanistan. August 2002. Kabul: Action Contre la Faim, 2002.
16. Save the Children–US, Centers for Disease Control and Prevention. Malnutrition and Mortality in Kohistan District, Afghanistan, April 4–10 2001. Pakistan/Afghanistan Field Office. Kabul: Save the Children–US, 2001.
17. UNICEF. Nutrition and Health Survey, Maslakh IDPs Camp, Herat, Afghanistan, April 2002. Kabul: UNICEF, 2002.
18. Action Contre la Faim. Nutritional Anthropometric Screening of Returnee Children under 5 at Pul E Charki Encashment Centre, July. 2003. Kabul: Action Contre la Faim, 2003.
19. Ministry of Public Health of the Islamic Republic of Afghanistan. Public Nutrition Department. Public Nutrition Policy and Strategy 2003–2006. Kabul: Ministry of Public Health of the Islamic Republic of Afghanistan, 2003.
20. Action Contre la Faim. Nutrition and Mortality Survey, Kabul City, Afghanistan, March 2001. Kabul: Action Contre la Faim, 2001.
21. Action Contre la Faim. Nutrition and Mortality Survey in Panjsheer Valley, March 2001. Kabul: Action Contre la Faim, 2001.
22. Action Contre la Faim. Nutritional Anthropometric Survey, Afghanistan. Southern Kapisa. Children under 5 years. August 2003. Kabul: Action Contre la Faim, 2003.
23. Central Statistical Office of the Islamic Republic of Afghanistan (CSO)/UNICEF. Progress of provinces: Multiple Indicator Cluster Survey. Kabul: Central Statistical Office of the Islamic Republic of Afghanistan (CSO)/UNICEF, 2003.
24. Grace J, Pain A. Re-thinking rural livelihoods in Afghanistan. Kabul: Afghanistan Research and Evaluation Unit, 2004.
25. Pain A. Livelihoods under stress in Faryab Province. Save the Children–US, 2001. Available at: www.areu.org.af/index.php?option=com_docman&Itemid=&task=doc_download&gid=319. Accessed 17 December 2006.
26. Save the Children–US. Breastfeeding and weaning beliefs and practices in parts of northern Afghanistan (Andkhoy, Khancharbagh, Qaramqul, and Qurghan districts, Jawsjan Province). Kabul: Save the Children–US, 2003.
27. Action Contre la Faim. Anthropometric nutrition survey, Mazar City, Afghanistan. March 2002. Kabul: Action Contre la Faim, 2002.
28. Save the Children–US. Nutrition Report in Andkhoy District. Kabul: Save the Children–US, 2003.

Effect of household processing on the *in vitro* bioavailability of iron in mungbean (*Vigna radiata*)

Lotika Barakoti and Kiran Bains

Abstract

Background. *Mungbean (*Vigna radiata*) is a major source of energy and protein in developing countries, especially for the vegetarian population. Improvement of the bioavailability of iron in mungbean by common household processes could make a significant contribution to the nutrition of people in countries where iron-deficiency anemia is widespread.*

Objective. *The study was conducted to determine the effect of common household processes on nutritional and antinutritional factors as well as* in vitro *bioavailability of iron in mungbean.*

Methods. *Mungbean was subjected to various domestic processes such as dehulling, pressure cooking, germination, and fermentation. The effects of these processes on proximate composition, antinutritional factors (phytin phosphorus, polyphenols, and neutral detergent), and iron, including ionizable iron, were determined.*

Results. *No significant change in crude protein content was observed. There was a significant ($p \leq .05$) increase in fiber content after germination. Ash content decreased significantly ($p \leq .05$) after all processing methods. The processing methods resulted in a significant ($p \leq .05$) reduction in phytin phosphorus and polyphenols. Pressure cooking significantly ($p \leq .05$) decreased the neutral detergent fiber, whereas fermentation and germination increased it. The phytate:iron molar ratio of processed mungbean revealed that the maximum reduction was in germinated and fermented samples. Ascorbic acid content increased significantly ($p \leq .05$) after germination. The* in vitro *bioavailability of iron in raw mungbean was 7.32%. All of the processing methods resulted in an increase in iron bioavailability* in vitro*; the maximum bioavailability was in germinated cooked mungbean (12.52%), followed by fermented cooked mungbean and germinated raw mungbean (both 11.04%).*

Conclusions. *Suitable processing techniques can improve the* in vitro *bioavailability of iron from mungbean. If mungbean products with enhanced iron bioavailabilty are developed, they could help improve the iron status of the population.*

Key words: Fermentation, germination, iron bioavailability, mungbean

Introduction

Legumes are an excellent source of protein and a fair source of iron, but their contribution to the nutrition of the consumer is limited, primarily due to the presence of antinutritional factors. It is widely recognized that minerals are generally less bioavailable from foods of plant origin. Dietary fiber, phytic acid, and polyphenols in legumes are thought to be the major contributors to the reduction in availability of minerals from these foods [1]. Excessive dietary intake of phytates affects the bioavailability of important polycations such as Fe^{3+} and Zn^{2+} by the formation of insoluble metal–phytate complexes [2]. Therefore, removal of these antinutrients by processing methods is necessary for effective utilization of legumes [3].

The reliable method of determining the bioavailability of iron from diets is to measure iron absorption in humans by the extrinsic tag method [4]. The *in vivo* method is accurate but it is time-consuming and expensive. An *in vitro* method, based on the release of ionizable iron from foods subjected to treatment with pepsin–hydrochloric acid at pH 1.35 and subsequent adjustment of the pH to 7.5, simulating conditions in the stomach and the intestine, respectively, can be used to predict iron bioavailability. Ionizable iron from foods and diets determined by this method is highly correlated with the actual percentage of absorption in the

The authors are affiliated with the Department of Food and Nutrition, Punjab Agricultural University, Ludhiana 141004, Punjab, India.

Please direct queries to the corresponding author: Kiran Bains, Department of Food and Nutrition, Punjab Agricultural University, Ludhiana 141004, Punjab, India; e-mail: kiranbains68@hotmail.com.

diets of adult male subjects observed by the extrinsic tag method (correlation coefficient = 0.94) [5].

The commonly consumed legumes in Indian diets are red kidney beans, bengal gram, mungbean, cowpea, redgram, mothbean, and lentils. Among them, mungbean (*Vigna radiata*) ranks third in production after bengal gram and red gram. Among the grain legumes, it is known for its easy digestibility and low flatulence potential in addition to having a high protein content and being a fair source of dietary minerals [6]. Mungbean is a major source of energy and protein in developing countries, especially for the vegetarian population, and improvement of the bioavailability of iron in mungbean could significantly improve its nutritional value.

Materials and methods

Preparation of samples

Mungbean variety ML-613 was procured from the legumes section of the Department of Plant Breeding, Punjab Agricultural University, Ludhiana, Punjab, India. A portion of the variety was dehulled. Whole and dehulled mungbean was subjected to the following household processing methods three times for the preparation of samples in the laboratory.

Pressure cooking

Whole and dehulled mungbean samples were washed thoroughly in running tap water and pressure cooked at 15 lb pressure for 15 and 5 minutes, respectively. Pressure cooking is the most commonly used cooking method for legumes by most people in India.

Germination

Whole mungbean was soaked for 12 hours in tap water and then allowed to germinate for 24 hours. The germinated mungbean was then pressure cooked in tap water (seed:water ratio, 1:2 w/v) for 4 minutes.

Fermentation

Dehulled mungbean was soaked overnight in tap water and ground to a coarse paste in an electric grinder. The batter was fermented for 12 hours at 37°C and then beaten for 7 to 10 minutes. Refined oil was heated to 190°C, and balls of the batter were deep fried in hot oil for 1 minute. The processed samples were spread on sterile petri dishes, dried in a hot-air oven at 60 ± 5°C, and stored in an airtight container for chemical analysis. Fresh samples were used for determination of ascorbic acid.

Chemical analysis

Moisture, crude protein, ash, crude fat, and crude fiber were estimated by the AOAC method [7]. Ascorbic acid was estimated by the AOVC method [8]. Antinutritional factors (phytates, polyphenols, and neutral detergent fiber) were estimated by the methods of Haug and Lantzsch [9], AOAC [7], and Georing and Van Soest [10], respectively. Minerals (iron and zinc) were estimated by an atomic absorption spectrophotometer (Varian model). The molar ratio of phytate to iron was calculated.

In vitro availability of iron was determined by the method of Rao and Prabhavathi [5]. Food was extracted with pepsin–hydrochloric acid at pH 1.35, and subsequently the pH was adjusted to pH 7.5. The extract was filtered. Ionizable iron in the filtrate was determined by the alpha-alpha dipyridyl method. The ionizable iron at pH 7.5 is used as a reliable measure of bioavailable iron. The following prediction equation for iron absorption was used:

$$Y = 0.4827 + 0.4707\ X$$

where Y is the percentage iron absorption in the adult human and X is the percentage of ionizable iron at pH 7.5.

Statistical analysis

Means and standard deviations were calculated. Analysis of variance was used to determine the variation between the samples. The critical difference (CD) at 5% where the F-ratio was significant was calculated by the CPCS computer program I [11].

Results and discussion

The proximate composition of the samples is shown in **table 1**. The crude protein content in the processed sample varied between 19.68% and 23.41%, with the highest content in dehulled cooked mungbean and the lowest in whole cooked mungbean. The fat content of the samples varied between 0.88% and 1.55%, with the highest content in germinated raw mungbeans and the lowest in fermented raw and dehulled cooked samples. A significant ($p \leq .05$) difference was observed between the crude fat content of germinated raw and germinated cooked mungbean and of raw, whole mungbean. The highest crude fiber content (9.16%) was in germinated raw mungbeans, and the lowest was in dehulled raw mungbeans (1.70%). A significant ($p \leq .05$) difference was observed between the crude fiber content of germinated raw and germinated cooked mungbean and of raw, whole mungbean. Giami [12] also reported that germination resulted in a significantly higher crude fiber content. The ash content of processed samples ranged between 1.02% and 3.45%. There was a significant ($p \leq .05$) reduction in ash content after dehulling, cooking, and germination of mungbean. Studies have

TABLE 1. Effect of processing method on proximate composition of mungbean

Processing method	g/100 g dry matter				
	Moisture	Crude protein	Crude fat	Crude fiber	Ash
Whole raw	0.50	21.21	0.92	5.16	5.87
Whole cooked	0.53	19.68	0.93	5.70	1.41
Dehulled raw	3.85	22.26	1.02	1.70	1.78
Dehulled cooked	6.53	23.40	0.88	1.74	2.93
Germinated raw	6.00	21.35	1.55	9.16	1.02
Germinated cooked	1.50	22.79	1.36	8.52	2.42
Fermented raw	0.62	20.06	0.88	3.23	3.45
Fermented cooked	0.60	22.44	1.16	4.16	2.05
F-ratio	30.45*	1.28	415.16*	6.17*	3.23*
CD (5%)	1.24	NS	0.36	2.99	2.39

CD, critical difference; NS, not significant
*Difference significant at $p \leq .05$

found that there is a significant reduction in ash content after scarification, fermentation, and germination of pulses [13–15].

All of the processing methods—dehulling, cooking, germination, and fermentation—resulted in a significant ($p \leq .05$) decrease in the phytin phosphorus content as compared with raw whole mungbean. A significant decrease in phytate content by domestic processing methods was reported in literature [14]. Germination reduced the phytic acid content of chickpea and pigeonpea seeds by more than 60%, and that of mung bean, urd bean, and soybean by about 40%. Fermentation reduced phytic acid content by 26% to 39% in all these legumes with the exception of pigeonpea, in which it was reduced by more than 50% [16]. The decrease in phytin phosphorus content of legume seeds during soaking and sprouting can be attributed to leaching of phytates into the water under the influence of the concentration gradient. The decrease in phytic acid content due to cooking may be attributed to the formation of insoluble complexes between phytates and other components, and also to the breakdown of phytic acid content at high temperatures. The breakdown of phytates by domestic processing techniques helps to enhance mineral bioavailability [17]. The polyphenol content of processed mungbean varied between 189 and 442 mg/100 g. The maximum reduction was obtained in fermented cooked mungbean samples followed by germinated cooked samples, whereas the least reduction was in the dehusking process. However, all processing techniques reduced the polyphenol content significantly. Cooking has been reported to reduce polyphenol content significantly, possibly due to the formation of some insoluble complexes between proteins and tannins [18]. The neutral detergent fiber values of samples ranged between 16.6% and 27.7%. There was a significant reduction in neutral detergent fiber content after cooking and dehulling. On the other hand, a significant increase in neutral detergent fiber content after germination and fermentation was observed (**table 2**). The neutral detergent fiber content of cooked lentils was significantly lower than that of

TABLE 2. Effect of processing method on phytin phosphorus, polyphenol, and neutral detergent fiber (NDF) contents of mungbean

Processing method	Phytin phosphorus (mg/100 g dry matter)	Polyphenols (mg/100 g dry matter)	NDF (g/100 g dry matter)
Whole raw	140	528	21.9
Whole cooked	112	322	18.8
Dehulled raw	124	442	16.6
Dehulled cooked	108	219	17.5
Germinated raw	93	257	27.5
Germinated cooked	82	210	27.7
Fermented raw	112	216	26.2
Fermented cooked	106	189	26.6
F-ratio	235.27	11623.16	124.00
CD (5%)	3.46*	3.46*	1.28*

CD, critical difference
* Difference significant at $p \leq .05$

raw legume, indicating that cooking caused a considerable decrease in neutral detergent fiber content [19]. On the other hand, germination of legumes increased their neutral detergent fiber content [20, 21].

Processing of mungbean by germination significantly ($p \leq .05$) increased its ascorbic acid content to 9.06 mg/100 g. However, cooking the germinated samples resulted in a decrease in ascorbic acid content to 6.98 mg/100 g (**table 3**). An increase in ascorbic acid content has been reported in legumes after germination. The ascorbic acid content increased to 6.86 mg/100 g in blackgram after germination agreed with the corresponding values observed in the present study [22]. Ascorbic acid is a strong promoter of iron absorption [23]. The ability of ascorbic acid to reduce iron and thus to prevent the formation of less-soluble ferric compounds is an important mechanism for the absorption-promoting effect of ascorbic acid [24].

The iron content of the samples ranged from 4.25 to 6.90 mg/100 g. Dehulling reduced total iron by 7.2%, whereas germination increased it by 31.8%. The percentage bioavailability of iron from processed samples varied from 8.0% to 12.5%, with the highest value in germinated cooked samples and the lowest in dehulled raw samples. Dehulling of whole mungbean enhanced *in vitro* iron bioavailability by 9%. Pressure cooking of whole mungbean increased iron bioavailability by 21%, whereas pressure cooking of dehulled mungbean increased it by 35%. Germination increased iron bioavailability by 50%, whereas cooking the germinated mungbean increased it by 71%. Fermentation of dehulled mungbean resulted in an increase of 19% in iron bioavailability. Frying the fermented sample further increased it by 38%. The phytate:iron molar ratio as a predictor of iron bioavailability ranged between 3.49 and 7.97. The maximum reduction of the phytate: iron ratio was in germinated samples (**table 4**). Cooking, germination, and fermentation processes decrease the phytate:iron ratio of legumes from that of raw legumes as a result of massive breakdown of phytates.

TABLE 3. Effect of processing methods on ascorbic acid content of mungbean

Processing method	Ascorbic acid (mg/100 g fresh weight)
Whole raw	0.62
Whole cooked	0.42
Dehulled raw	0.67
Dehulled cooked	0.42
Germinated raw	9.06
Germinated cooked	6.98
Fermented raw	12.24
Fermented cooked	8.86
F-ratio	13,425.65*
CD (5%)	0.16

CD, critical difference
* Difference significant at $p \leq .05$

The household processing techniques of dehulling, cooking, germination, and fermentation reduced phytates and polyphenols. Phytates and polyphenols are reported to be inhibitors of iron absorption. Addition of different known amounts of phytates and tannic acid to a meal resulted in a linear reduction in iron absorption [25, 26]. Processing by germination resulted in an increase in ascorbic acid in addition to reduction in phytates and polyphenols. Hallberg et al. [24] reported that the addition of 100 mg of ascorbic acid to a semisynthetic liquid formula increased iron absorption by 4.14 times. The addition of 100 mg of ascorbic acid to another similar liquid formula containing 85 mg of phytin-phosphorus increased iron absorption by 3.14 times. Therefore, the cumulative effect of two approaches, i.e., degradation of phytates and increase in ascorbic acid content, proved helpful

TABLE 4. Effect of processing method on total iron, ionizable iron, phytate:iron molar ratio, and *in vitro* iron bioavailability

Processing method	Total iron (mg/100 g dry matter)	Ionizable iron (mg/100 g dry matter)	Phytate:iron	*In vitro* iron bioavailability (%)
Whole raw	5.10	0.74	8.33	7.32
Whole cooked	4.52	0.80	7.50	8.86
Dehulled raw	4.74	0.75	7.97	7.99
Dehulled cooked	4.35	0.87	7.53	9.95
Germinated raw	6.72	1.50	4.16	11.04
Germinated cooked	6.90	1.76	3.49	12.52
Fermented raw	4.25	0.81	7.89	9.55
Fermented cooked	5.23	1.17	6.13	11.04
F-ratio	126.27*	14.07*	†	392.84*
CD (5%)	0.27	0.36	†	0.26

CD, critical difference
* Difference significant at $p \leq .05$
† Phytate:iron is a calculated value; statistical tests were not employed

in enhancing the *in vitro* bioavailability of iron from mungbean. Although mungbean is considered a major source of calories and protein in developing countries, especially for the vegetarian population, mungbean products with enhanced iron bioavailability, if they are developed, may help improve the iron status of the vegetarian population, since iron-deficiency anemia is widespread in these countries.

References

1. Hazell T, Johnson IT. In vitro estimation of iron bioavailability from a range of plant foods: Influence of phytate, ascorbate and citrate. Br J Nutr 1986;57:223–33.
2. Graf E, Eaton JW. Effects of phytate on mineral bioavailability in mice. J Nutr 1984;114:1192–8.
3. Paramjyothi S, Mulimani VH. Effect of sprouting, cooking and dehulling on polyphenols of red gram (Cajanus cajan L.). J Food Sci Tech 1996;33:259–60.
4. Hallberg L, Hulthen L. Methods to study dietary iron absorption in man—An overview. In: Hallberg L, Asp N-G, eds. Iron nutriture in health and disease. London: John Libbey & Company, 1996:81–95.
5. Rao BSN, Prabhavathi T. An in vitro method for predicting the bioavailability of iron from foods. Am J Clin Nutr 1978;31:169–75.
6. Doughty J, Walker A. Legumes in human nutrition. FAO Food and Nutrition Paper No. 20. Rome: Food and Agriculture Organization, 1982:46.
7. Association of Official Analytical Chemists (AOAC). Official methods of analysis. Washington, DC: AOAC, 1985.
8. Association of Vitamin Chemists (AOVC). Methods of vitamin assay. New York: Inter Science Publishers, 1996:306–12.
9. Haug W, Lantzsch HJ. Sensitive method for the rapid determination of phytate in cereals and cereal products. J Sci Food Agric 1983;34:1423.
10. Georing HK, Van Soest PJ. Forage fibre analysis (apparatus, reagents, procedure and some applications). In: US Department of Agriculture Handbook, Series No. 379. Washington, DC: US Department of Agriculture, 1970.
11. Cheema HS, Singh B. CPCS-1: A computer programme package for the analysis of commonly used experimental designs. Ludhiana, India: Punjab Agricultural University, 1990.
12. Giami SY. Effect of processing on the proximate composition and functional properties of cowpea (Vigna unguiculata) flour. Food Chem 1993;47:153–8.
13. Singh U, Rao PV, Seetha R, Jambunathan R. Nutrient losses due to scarification of pigeonpea (Cajanus cajan L.) cotyledons. J Food Sci 1989;54:974–6.
14. Kaur M, Mehta U. Effect of partial and full substitution of bengal gram and black gram dhals with ricebean on essential amino acid and mineral content of vada and pakoda. J Food Sci Tech 1993;30:454–6.
15. Igbedioh SO, Shaire S, Aderye BJI. Effects of processing on total phenols and proximate composition of pigeonpea (Cajanus cajan) and climbing bean (Vigna umbellata). J Food Sci Tech 1995;32:497–500.
16 Chitra U, Singh U, Rao PV. Phytic acid, in vitro protein digestibility, dietary fibre and minerals of pulses as influenced by processing methods. Plant Foods Hum Nutr 1996;49:307–16.
17. Grewal A. Effect of processing on nutritional quality of products from newly released varieties of green gram. MSc Thesis, CCS Haryana Agricultural University, Hisar, India, 2003.
18 Jood S, Bishnoi S, Sehgal S. Effect of processing on nutritional and anti-nutritional factors of mungbean cultivars. J Food Biochem 1998; 22:245–57.
19. Vidal VC, Frias J. Dietary fibre in processed lentils. J Food Sci 1992;57:1161–3.
20. Kaur M. Effect of domestic processing on mineral bioavailability from ricebean (Vigna umbellata). PhD Dissertation, Punjab Agricultural University, Ludhiana, India, 1997.
21. Bhama S. Effect of various processing methods on the availability of nutrients from Bengal gram products. MSc Thesis, Punjab Agricultural University, Ludhiana, India, 1999.
22. Bednarsi W, Tomasik J, Piatkowska B. Processing, suitability and nutritive value of field bean seeds after germination. J Sci Food Agric 1985;36:745–51.
23. Cook JD, Monsen ER. Vitamin C, the common cold, and iron absorption. Am J Clin Nutr 1977;30:235–41.
24. Hallberg L, Brune M, Rossander L. Effect of ascorbic acid on iron absorption from different meals. Study with ascorbic acid rich foods and synthetic ascorbic acid given in different amounts with different meals. Hum Nutr Appl Nutr 1986;40:97–113.
25. Brune M, Rossander L, Hallberg L. Iron absorption and phenolic compounds: Importance of different phenolic structures. Eur J Clin Nutr 1989;43:547–58.
26. Hallberg L, Brune M, Rossander L. Iron absorption in man: Ascorbic acid and dose dependent inhibition by phytate. Am J Clin Nutr 1989;49:140–4.

Improvement of the nutritional quality of a traditional complementary porridge made of fermented yellow maize (*Zea mays*): Effect of maize–legume combinations and traditional processing methods

Jeanne Ejigui, Laurent Savoie, Johanne Marin, and Thérèse Desrosiers

Abstract

Background. *Blends with a cereal–legume ratio of 70:30 have been introduced in many communities for use in the preparation of complementary foods with augmented protein quality. These foods should meet World Health Organization estimated energy and nutrient needs from complementary foods.*

Objective. *To increase energy and nutrient densities and nutrient availability in a traditional complementary porridge.*

Methods. *Yellow maize was processed by lactic acid fermentation. Peanuts (*Arachis hypogea*) and beans (*Phaseolus vulgaris*) were processed by germination, roasting, dehulling, and a combination of germination and roasting. Blends were prepared from processed peanuts and beans and cooked into porridges with viscosities less than 3,000 cp. Traditional porridge was the control and consisted of fermented yellow maize only. The porridges were analyzed for their physicochemical and nutritional properties.*

Results. *Blends increased energy and nutrient densities in porridges compared with the control (p < .05). The maize–peanuts combination yielded porridges with higher energy densities and improved nutritional quality compared with the maize–beans combinations.* In vitro *availability of iron did not change (p > .05) with formulation of the blends except for porridges made from maize and germinated peanuts, but there was a significant increase in zinc* in vitro *availability, whereas a decrease was observed for calcium* in vitro *availability. The energy densities of maize–peanuts porridges were sufficient to cover energy required from complementary foods for infants aged 6 to 11 months receiving four meals of complementary foods per day and an average amount of energy from breastmilk.*

Conclusions. *Maize-legume blends can efficiently improve the nutritional quality of traditional porridge. Peanuts are the best legume complements.*

Key words: Bean, blend formulation, complementary porridge, energy density, maize, nutritional value, peanut, traditional processing, viscosity

Introduction

The search for low-cost, nutritious, and easy-to-prepare locally available complementary foods will continue as long as protein–energy malnutrition still prevails in developing countries. Protein–energy malnutrition contributes to more than half of the deaths of children under five years of age throughout the world [1]. In 2004, the mortality rates of children under 5 years and under 1 year of age in sub-Saharan Africa were estimated at 17.1 and 10.2 per 100 live births, respectively [2]. In Cameroon in 2004, the mortality rates in these two age groups were 14.9 and 8.7 per 100 live births, respectively [2]. In addition, the prevalence of moderate underweight, severe underweight, moderate or severe wasting and moderate or severe stunting among children under five years of age in Cameroon was 18%, 4%, 5% and 32%, respectively [3]. Since nonexclusive breastfeeding, early weaning, nutritionally inferior diets, and improper complementary feeding are major contributing factors to the development of protein–energy malnutrition, one proposed approach could be the improvement of the nutritional value of traditional complementary foods. In fact, African traditional complementary foods, which are usually prepared from starchy flours only, have very low energy and nutrient densities [4].

The concept of cereal–legume complementation has been particularly applied to the development of

The authors are affiliated with Université Laval, Faculté des Sciences de l'Agriculture et de l'Alimentation, Département des Sciences des Aliments et de Nutrition, Québec, Canada.

Please direct queries to the corresponding author: Jeanne Ejigui, Université Laval, Faculté des Sciences de l'Agriculture et de l'Alimentation, Département des Sciences des Aliments et de Nutrition, Pavillon Paul-Comtois, Local 1312, Québec G1K 7P4, Canada; e-mail: ejigui@hotmail.com.

infant complementary foods with augmented protein quality [5–8]. Nutritionally, the high protein content of legumes increases the protein content of cereal-based complementary foods and supplements the deficient amino acids. However, to be considered suitable for infants, complementary foods must meet other standards in addition to protein content and quality. Research efforts should therefore be concentrated on the best ways of meeting World Health Organization (WHO) energy and nutrient estimated needs from complementary foods with the recommended 70:30 cereal–legume ratio and should focus on factors affecting energy intake, such as energy density and viscosity of the complementary foods and the frequency of feeding [9]. Higher energy density is proportional to higher energy intake, and low energy intake is inversely proportional to viscosity [10].

On the other hand, the use of unrefined maize flour, red sorghum, and legumes may be limited due to inherent antinutritional factors such as amylase inhibitors, protease inhibitors, phytates, condensed tannins, and polyphenols, which lower the utilization of starch, protein, and minerals unless they are processed [11–13]. Phytate also chelates divalent cations, thus reducing their bioavailability.

Traditional processing methods, such as fermentation, germination, and roasting, are simple and inexpensive and have been practiced for many years in developing countries. These methods have often been used separately or in combination for preparation of infant complementary foods [5–8] and the nutritional profile of these foods has been reported. Traditional processing may produce foods with many positive attributes, such as favorable texture, good organoleptic quality, reduced bulk, enhanced shelf life, partial or complete elimination of antinutritional factors, reduced cooking time, and improved nutritional value [5].

The objectives of this study were to test complementary foods made from recommended blends with a 70:30 cereal–legume ratio (weight/weight) by comparing their energy density and nutrient content with WHO estimated needs from complementary foods; to evaluate the effect of traditional processing methods and blend formulation on the physicochemical and nutritional characteristics of complementary porridges in terms of pH and viscosity; to compare the nutritive value of peanuts and beans as legume complements; and to determine the traditional processing method with the best potential for improving the nutritional quality of the traditional porridge.

Materials and methods

Materials

Fermented maize flour was obtained by fermentation of yellow maize (*Zea mays*) following the steps in the traditional procedure described by Ejigui et al. [13], which consists of steeping whole maize seeds in water and allowing them to ferment for 96 hours, with an extraction rate of approximately 75% to 80%. Small red peanuts (*Arachis hypogea*) and small red kidney beans (*Phaseolus vulgaris*) were purchased at a local market at Yaounde, Cameroon. The seeds were germinated, roasted, and dehulled according to the methods described by Ejigui et al. [11].

Formulation of blends and cooking of the porridges

Blends were formulated as 70% cereal to 30% legume, the recommended ratio [14] for correcting for lysine deficiency in maize. This would, theoretically, be nutritionally equivalent to casein [15]. The control porridge, fermented maize porridge (FMP), was made from maize flour only, whereas the six porridges made from blends of maize and processed peanut paste or maize and processed beans were fermented maize with germinated peanuts (FMGEP), fermented maize with roasted peanuts (FMRP), fermented maize with germinated and roasted peanuts (FMGRP), fermented maize with germinated beans (FMGEB), fermented maize with roasted beans (FMRB), and fermented maize with germinated and roasted beans (FMGRB). The ratio of blend to cooking water (w/v) was chosen so that the porridge viscosity would not exceed 3,000 cp, which is the highest reference value [14]; this ratio was 11.5:100 for blends of fermented maize flour and processed peanuts and 9.5:100 for blends of fermented maize flour and processed beans. The blends were cooked into porridges by the traditional method used by mothers in Cameroon, adapted for the laboratory, which consists of mixing the blends with cold distilled water and allowing the mixture to hydrate for 3 hours, and then bringing it to a boil and allowing it to cook for 20 minutes. After 15 minutes of cooking, 13.6 g of sugar (SOSUCAM, Cameroon) for each 600 mL of cooking water was added.

Energy and nutrient densities were compared with WHO estimated needs for children aged 6 to 11 months, receiving an average amount of energy from breastmilk and four meals per day. We chose to compare the adequacy of the Fe, Ca, and Zn intakes to the WHO estimated needs for infants fed four meals a day rather than the customary three for several reasons. First, porridges in the present study are made with plant foods, which are not nutrient dense. It would be difficult to meet the WHO estimates from complementary foods without considering frequency of feeding. Second, the objective of this study was to improve the nutritional quality of a traditional porridge while looking at factors affecting energy and nutrient intakes, which includes feeding frequency. And finally, we should encourage mothers to change certain nutritional habits when there is a clear advantage to them and their infants. Mothers in developing countries usually feed their children three times a day, but it is not unrealistic to suggest that they should increase the frequency of feeding.

Laboratory analyses

All experiments were done in triplicate, and analyses were done in triplicate or quadruplicate.

There was no need to perform analytical quality control for macronutrient, moisture, and ash analyses since they were determined by AOAC (Association of Official Analytical Chemists) routine and standard methods. However, analyses were repeated if a difference of more than 10% was observed. Carbohydrate and fiber, amino acid, and chemical score were computed. Complex methods such as *in vitro* protein digestibility and *in vitro* mineral availability were both performed by using digestion cells, which has shown good performance, reproducibility, and ease of use. The method has been proved to be accurate and efficient [16]; therefore, investigations on its precision were not done in the present study, since analyses in the present research were performed in the same laboratory, using the same equipment and protocols.

To determine mineral content, standard curves were used for calibration, and the readings were done twice. A sample with a known concentration was used to verify the precision of the method, and the differences were 2, 7, and 3% for iron, zinc, and calcium, respectively.

Viscosity

Viscosity was measured in freshly cooked sample complementary porridges with a digital Brookfield viscosimeter (model DV-II Brookfield Engineering Laboratories) using spindle RV no. 4 at 50 rpm (40°C). The pH was measured with a Fisher Scientific 925 Accumet pH meter.

Chemical analyses

Prior to chemical analysis, the porridges were freeze-dried with a Virtis freeze-dryer, milled in a Tecator 1093 cyclotec sample mill to pass through a 1-mm sieve, and stored in a desiccator in a cool room at 4°C until further analysis. Moisture and ash contents were determined by standard AOAC method 925.10 [17]. Nitrogen was determined by AOAC Kjeldahl method 979.09 [17] using a nitrogen autoanalyzer (Foss Electric), and crude protein content was calculated as %N × 6.25. Fat content was determined by AOAC Goldfish method no. 945.16 [17], and total carbohydrate and fiber was calculated by difference.

The method of Savoie and Gauthier [16] was used to measure *in vitro* digestibility of proteins; 93.75 mg of protein (15 mg N) was homogenized in 16 mL of 0.1 M HCl in a flat-bottomed glass tube and stirred magnetically for 10 min at 37°C. The pH was adjusted to 1.9 with deionized water, and 1 mL pepsin solution was added. After 30 min, the pH was adjusted to 7.5. The pepsin hydrolyzate was poured into the dialysis tube (Spectra/Por 6, Spectrum Medical Industries) with 1 kDa molecular weight cutoff, which was contained in the digestion cell. Pancreatin digestion was initiated by adding 1 mL of pancreatin solution. The products of digestion were continually collected by the circulation of $NaH_2PO_4.H_2O$ (0.01M, pH 7.5). The dialyzed digests were collected after 2, 4, and 6 hours. Protein digestibility was calculated by the equation: protein digestibility (%) = [nitrogen in the dialyzed digests (mg)/15 mg N] × 100.

Amino acid composition of each dialyzed digest was measured by ion exchange chromatography using a Beckman amino acid analyzer, model 6300. The samples were first hydrolyzed in 2 mL of 6 M HCl under vacuum for 24 hours at 100°C. Norleucine was used as standard. Amino acid release was calculated by the equation: amino acid release (%) = [dialyzed amino acid (mg)/amino acid in the protein sample (mg)] × 100. Essential amino acid scores were calculated by dividing the amino acid contents by the values in the reference protein [18]. The chemical score of the complementary porridges corresponded to the lowest value of these essential amino acid scores.

Iron, zinc, and calcium contents were simultaneously determined by flame atomic absorption spectrophotometry (Perkin-Elmer Analysis 100, USA) after wet digestion of the sample with HNO_3 and $HClO_4$ [19]. *In vitro* extractability of iron, zinc, and calcium was evaluated by pepsin–pancreatin digestion as described by Miller et al. [20]. It is well known that *in vitro* methods of analysis have some limitations in comparison with *in vivo* methods, which are considered to be more accurate. *In vitro* methods were used in order to develop improved complementary porridges by using WHO estimated needs as reference. Several combinations of foods and various processing methods could be studied at once in a short period of time without being influenced by the nutritional or medical status of infants and compliance of mothers. We could also obtain information on the effect of processing on various nutrients. This methodology is useful to determine which complementary porridges and processing methods have a good potential to be tested in a real situation.

Statistical analyses

Unless otherwise stated, all experiments were conducted in triplicate and means and standard deviations were reported. Triplicate experiments were used to detect exprimental errors. Analysis of variance (ANOVA) was performed with $p < .05$. The least-squares means test in a factorial design was used to look at the effect of blend formulation, processing, and their interactive effect on the nutritional characteristics of the complementary porridges made with blends of fermented maize with processed peanuts and fermented maize with processed beans. The statistical analyses were performed by SAS version 6.12 (SAS Institute Inc.).

Results and discussion

Nutritional properties of complementary porridges

Energy density

Energy density was significantly higher in all of the complementary porridges than in the control FMP ($p < .05$) due to blend formulation (**table 1**). Energy density in maize–peanuts complementary porridges was higher than energy density in maize-beans complementary porridges ($p < .05$). Maize–peanuts complementary porridges provided about 15% more kilocalories than maize–beans complementary porridges because of their higher percentage of fat (**table 2**). The minimum desirable energy densities recommended by WHO [9] for well-nourished infants receiving four meals per day and high energy from breastmilk are 24 kcal/100 g porridge for those aged 6 to 8 months and 38 kcal/100 g porridge for those aged 9 to 11 months (**table 1**). The energy density of the experimental complementary porridges ranged from 45 to 61 kcal/100 g porridge and was more than the 24 kcal and 38 kcal/100 g porridge recommended by WHO. However, it may be unrealistic to expect a well-nourished child to receive high energy from breastmilk daily. We then assumed that the energy density of the complementary foods should be compared with the WHO [9] estimated needs from complementary foods for well-nourished infants receiving average instead of high energy from breastmilk and four meals per day. The maize–peanuts complementary porridges met the WHO estimated needs for energy from complementary foods for the targeted age group of 9 to 11 months, whereas the maize–beans complementary porridges did not (**table 1**). The energy density of the control porridge (FMP) was the lowest (37 kcal/100 g porridge) and did not meet the WHO estimated need from complementary foods for well-nourished children aged 6 to 11 months receiving an average amount of energy from breastmilk.

The method of processing had a significant effect on energy density in the maize–peanuts and the maize–beans complementary porridges (**table 1**). The combination of germination and roasting yielded complementary porridges with the highest energy density in both legumes. The higher energy value of porridges made from maize with germinated and roasted legumes could be attributed to the apparent increase in protein and fat, probably due to dehulling and to the utilization of carbohydrate as a source of energy. This

TABLE 1. Effect of blends and processing methods on some nutritional properties of complementary porridges[1]

Nutritional properties	FMP	Porridges made with fermented yellow maize flour-processed peanut blends			Porridges made with fermented yellow maize flour-processed bean blends			Reference values[2]					
								6–8 mo			9–11 mo		
		FMGEP	FMRP	FMGRP	FMGEB	FMRB	FMGRB	L	A	H	L	A	H
Energy density (kcal)	37.45 ± 1.06^g	61.42 ± 0.05^c	62.53 ± 0.2^b	63.44 ± 0.36^a	46.41 ± 0.14^e	45.02 ± 0.2^f	49.3 ± 0.11^d	64	44	24	77	58	38
pH	3.16 ± 0.13^d	4.72 ± 0.02^c	4.76 ± 0.05^{bc}	4.72 ± 0.02^c	4.86 ± 0.01^a	4.85 ± 0.01^{ab}	4.88 ± 0.01^a	4.0–4.5	4.0–4.5	4.0–4.5	4.0–4.5	4.0–4.5	4.0–4.5
Viscosity (cp)	1654.3 ± 51.2^e	2530 ± 55.4^c	2898.7 ± 29^{ab}	2382.3 ± 27.3^d	2913.3 ± 11.6^{ab}	2960 ± 26.5^a	2833.3 ± 11.6^b	1000–3000[3]	1000–3000	1000–3000	1000–3000	1000–3000	1000–3000

FMP, porridge with fermented yellow maize flour (control porridge containing only maize); FMGEP, porridge with 70% fermented yellow maize flour and 30% germinated peanuts; FMRP, porridge with 70% fermented yellow maize flour and 30% roasted peanuts; FMGRP, porridge with 70% fermented yellow maize flour and 30% germinated and roasted peanuts; FMGEB, porridge with 70% fermented yellow maize flour and 30% germinated beans; FMRB, porridge with 70% fermented yellow maize flour and 30% roasted beans; FMGRB, porridge with 70% fermented yellow maize flour and 30% germinated and roasted beans.

1. Results are the means of three determinations. Plus–minus values are means ± SD. All values are given per 100 g of porridge. Means within the same row followed by the same letter are not significantly different by the Duncan's new multiple range and the LSMEANS tests ($p > .05$).
2. The reference values are from WHO/UNICEF [9], and the energy values were estimated for well-nourished children 6 to 8 months and 9 to 11 months of age, with estimated gastric capacity of 285 and 345 g, respectively, at four servings a day and at the WHO low (L), average (A), and high (H) amounts of breastmilk energy. Energy needs from complementary foods were set at total energy requirement + 2 SD (i. e., + 25%).

TABLE 2. Effect of blend and processing methods on the chemical composition of complementary porridges[1]

Nutrients	FMP	Porridges made with fermented yellow maize flour-processed peanut blends			Porridges made with fermented yellow maize flour-processed bean blends			WHO estimated needs from complementary foods[2]					
								6–8 mo			9–11 mo		
		FMGEP	FMRP	FMGRP	FMGEB	FMRB	FMGRB	L	A	H	L	A	H
Moisture (%)	91.82 ± 0.23^{a}	87.93 ± 0.04^{d}	87.89 ± 0.03^{d}	87.56 ± 0.66^{d}	89.55 ± 0.1^{bc}	89.9 ± 0.22^{b}	88.95 ± 1.15^{c}						
Ash (%)	0.023 ± 0.01^{c}	0.07 ± 0.01^{b}	0.08 ± 0.01^{b}	0.12 ± 0.02^{a}	0.12 ± 0.01^{a}	0.12 ± 0.01^{a}	0.12 ± 0.01^{a}						
Carbohydrate and fiber (%)	7.83 ± 0.2^{b}	8.63 ± 0.04^{ab}	8.58 ± 0.01^{ab}	8.94 ± 0.61^{a}	8.85 ± 0.1^{a}	8.50 ± 0.24^{ab}	9.23 ± 1.2^{a}						
Fat (%)	0.17 ± 0.05^{e}	1.57 ± 0.03^{b}	1.64 ± 0.02^{a}	1.51 ± 0.04^{c}	0.1 ± 0.02^{f}	0.11 ± 0.01^{f}	0.23 ± 0.03^{d}						
Crude Protein (% N × 6.25)	0.69 ± 0.03^{e}	1.8 ± 0.0^{b}	1.8 ± 0.0^{b}	1.87 ± 0.01^{a}	1.38 ± 0.01^{d}	1.36 ± 0.01^{d}	1.48 ± 0.01^{c}	0.5	0.2	0	0.8	0.3	0

Abbreviations are defined in table 1 footnotes.

1. Results are the means of three determinations. Plus–minus values are means ± SD. All values are given per 100 g of porridge. Means within the same row followed by the same letter are not significantly different by the Duncan's new multiple range and the LSMEANS tests ($p > .05$).

2. The reference values are from WHO/UNICEF [9], and the values were estimated for well-nourished children 6 to 8 months and 9 to 11 months of age, with estimated gastric capacity of 285 and 345 g, respectively, at four servings a day and at the WHO low (L), average (A), and high (H) amounts of breastmilk energy. Energy needs from complementary foods were set at total energy requirement + 2 SD

could have changed the nutrient distribution within the seeds [11].

The increase in energy was also associated in part with an increase in the dry matter content as a result of hydrolysis of starch by amylase.

pH levels

A significant increase in pH was observed in complementary porridges due to blend formulation (**table 1**). The control porridge (FMP) had a lower pH (3.16) than the cereal–legume complementary porridges, with pH values of 4.72 to 4.88. The low pH observed in FMP was due to lactic acid fermentation of yellow maize with the production of lactic acid. Nago et al. [21] observed a pH of 3.7 after a 1-week fermentation of maize. Processing did not significantly affect pH in maize–peanuts complementary porridges or in maize–beans complementary porridges. However, the pH in complementary foods was slightly higher than the safe hygienic pH of 4.0 to 4.5 [9].

Viscosity

Processing methods, blend formulation, and their interactive effects yielded significantly different ($p < .05$) viscosity levels (**table 1**). Except for FMRP, maize–peanuts complementary porridges (FMGEP and FMGRP) had lower viscosity than maize–beans complementary porridges (FMRB, FMGEB, and FMGRB). Griffith et al. [7] reported a viscosity of 1,515 cp in peanut-based weaning blends compared with 2,717 cp in cowpea-based blends formulated as a 60% cereal to 40% legume combination. Assuming that the solid content could have affected complementary porridge viscosity, a higher viscosity should have been expected from maize–peanuts complementary porridges because of their higher dry matter content estimated from moisture values (**table 2**). However, this was not the case; higher viscosity was observed in maize–beans complementary porridges than in maize–peanuts complementary porridges, except for FMGRB. The lower viscosity of maize–peanuts complementary porridges compared with maize–beans complementary porridges could be explained by the fat content of peanuts. A lowering effect of fat on complementary porridge viscosity was reported by Griffith et al. [7] and Ward and Ainsworth [22].

Processing methods had the most obvious effect on viscosity. In maize–peanuts complementary porridges, the combination of germination and roasting (FMGRP) produced a significantly ($p < .05$) lower viscosity than either germination (FMGEP) or roasting (FMRP). Roasting produced a complementary porridge with the highest viscosity in maize–peanuts complementary porridges, whereas the differences in viscosity between FMGEB and FMRB and between FMGEB and FMGRB were not significant (2,913 vs. 2,960 and 2,913 vs. 2,833, respectively). The difference in viscosity observed among the treatment groups could be explained by the increase in α-amylase content of germinated legumes. Germination of legumes increases the amount of α-amylase, which hydrolyzes starch into dextrin and maltose and thus reduces viscosity [9].

The viscosity of both maize–peanuts and maize–beans complementary porridges, however, was higher than that of the control porridge FMP (**table 1**), showing also the significant ($p < .05$) effect of blend formulation on the viscosity of complementary porridge compared with the control porridge. This could be attributed to the difference between the moisture content of the complementary porridges (87.6% to 89.9%) and that of the control porridge (91.8%) (**table 2**). However, the viscosity of the control porridge could have been above the recommended values of Mosha and Svanberg [14] if more than 9.5% maize flour had been used.

Chemical composition of the complementary porridges

Carbohydrate and fiber

Table 2 shows the mean chemical composition of the complementary porridges. The content of carbohydrate and fiber, which were the principal nutrients in all the complementary porridges, significantly increased with blend formulation from 11.52% to 15.2% compared with FMP, but no significant change ($p > .05$) was observed among maize–peanuts complementary porridges or among maize–beans complementary porridges. Processing did not affect the carbohydrate and fiber content of maize–peanuts complementary porridges (FMGEP, FMRP, and FMGRP) or maize–beans complementary porridges (FMGEB, FMRB, and FMGRB). On a dry matter basis (**table 3**), the carbohydrate and fiber content of complementary porridges was comparable to the values reported by Egounlety [8] in legume-fortified complementary food made with fermented maize. However, the fiber values in the present study were calculated by difference and were not obtained by chemical analysis.

Fat content

The fat content in maize–peanuts complementary porridges significantly increased ($p < .05$) compared with the control porridge (FMP), but a decrease was observed in maize–beans complementary porridges (**table 3**). Maize–peanuts complementary porridges were significantly higher in fat than maize–beans complementary porridges. As an oil seed, peanuts provided more fat to complementary porridges than beans. A similar trend was observed by Griffith et al. [7], who compared peanut-based blends with cowpea-based blends. The use of peanuts in blend formulation not only increased the fat but also provided a more concentrated energy source rich in the essential fatty acid linoleic acid [7].

Processing had a significant ($p < .05$) influence on fat content in maize–peanuts and maize–beans complementary porridges. Fat content was slightly higher ($p < .05$) in FMRP than in FMGEP and FMGRP (1.64% compared with 1.57% and 1.51%, respectively) and significantly higher ($p < .05$) in FMGRB than in FMRB and FMGEB (0.23% compared with 0.11% and 0.1%, respectively). On a dry matter basis (**table 3**), fat provided an average of 22.7% of the total kilocalories of peanut-based complementary porridges, compared with 2.7% for bean-based complementary porridges. Dewey and Brown [23] suggested that fat should provide 30% to 45% of total dietary energy in children 6 to 23 months of age. However, in developing countries, because of low fat intakes from mixed diets and the difficulty of achieving adequate micronutrient densities with high-fat diets, it is recommended that complementary foods provide 25% of total energy from fat, regardless of maternal milk fat content [9] and 3% of total energy from the essential fatty acids, linoleic acid (C18:2ω6) and linolenic acid (C18:3ω3) [1, 7]. None of the complementary porridges met the WHO estimated

TABLE 3. Proximate composition of the blends on a 100 g dry matter basis

Elements	FMP	FMGEP	FMRP	FMGRP	FMGEB	FMRB	FMGRB
Moisture (%)	2.46	2.80	3.32	2.56	3.29	4.90	3.32
Energy density (kcal)	439.03	508.67	516.32	509.62	444.06	445.74	446.18
Ash (%)	0.30	0.58	0.64	0.92	1.11	1.13	1.04
Carbohydrate and fiber (%)	89.13	68.72	67.56	69.32	81.46	79.37	80.23
Fat (%)	2.10	13.00	13.56	12.15	0.95	1.09	2.05
Crude protein [% (N × 6.25)]	8.01	14.91	14.92	15.05	13.18	13.50	13.36
Iron (mg)	2.99	2.22	2.56	4.03	3.74	3.93	3.83
% protein kcal	7.29	11.72	11.56	11.81	11.87	12.11	11.98
% fat kcal	4.3	23	23.63	21.46	1.9	2.2	4.1
% Carbohydrate and fiber kcal	81.20	54.04	52.34	54.40	73.4	71.22	71.93
Zinc (mg)	1.05	1.33	1.64	1.78	1.73	1.58	1.93
Calcium (mg)	19.26	25.60	29.17	24.90	27.16	27.98	33.79

Abbreviations are defined in table 1 footnotes.

needs for fat intake from complementary foods. However, there is little scientific information to support the need for fat to provide 30% to 45% of total dietary energy , as long as the needs for essential fatty acids are met and the energy density of the diet exceeds minimal criteria [7]. Griffith et al. [7] reported a mean energy distribution of fat in complementary blends made with 60% millet–40% peanut or 60% millet–40% cowpea. Fat provided an average of 37% of the total kilocalories in peanut-based blends, compared with 2.8% in cowpea-based blends. In the present study, the blends contained 70% cereal and 30% legume. This composition may explain the low fat content of the blends compared with those of Griffith et al. [7]. The low fat content of the complementary porridge blends could place a child at risk for essential fatty acid deficiency.

Protein content

WHO [9] estimated needs from complementary foods for protein were met by all the complementary porridges including the control porridge (**table 2**). Protein content significantly increased ($p < .05$) with blend formulation compared with FMP. The maize–peanuts complementary porridges provided about 23% more protein than the maize–beans complementary porridges and the protein content of the maize–peanuts complementary porridges was slightly higher ($p < .05$) in FMGRP (1.87%) than in FMGEP and FMRP, which both contained 1.8% protein. A similar trend was observed in bean-based complementary porridges, where the protein content was slightly higher in FMGRB (1.48%) than in FMGEB (1.38%) and FMRB (1.36%).

Protein–energy ratios on a dry matter basis (**table 3**) provide an indication of the protein sufficiency of complementary porridges and illustrate an appropriate balance of protein to energy. This ratio represents the energy contribution of protein in relation to the total energy value of the complementary porridges (% protein kcal) [7]. Protein–energy ratios of 10% to 11% are recommended for complementary foods, especially those of entirely vegetal origin [24]. **Table 3** shows that except for FMP, the protein–energy ratios were above the recommended levels for all the complementary porridges. The values ranged from 11.6% to 12.1%. Maize–beans complementary porridges had slightly higher protein–energy ratios than maize–peanuts complementary porridges, due to the difference in energy density on a dry matter basis. Proteins should also be viewed in light of their quality, illustrated by protein digestibility, chemical score, and amino acid release, to ensure that the proteins will be used efficiently.

Iron, zinc, and calcium content and in vitro *availability*

Despite significant increases observed in some complementary porridges due to blend formulation and processing, iron and calcium did not meet WHO estimated needs from complementary foods, whereas zinc met the estimated needs of infants and younger children receiving four meals a day and an average amount of energy from breastmilk (**table 4**).

Even though it is important to have sufficient amounts of minerals in complementary foods, it is more important to have them in a form in which they are easily digestible and available to the body. *In vitro* availability of minerals under "simulated" gastrointestinal conditions is an indicator of bioavailability [6]. In the present study, we have assumed that low iron availability should be considered as the reference, since all ingredients in the blends are plant foods. WHO references for foods of low iron availability are therefore 2.1 and 1.9 mg/100 g food for infants aged 6 to 8 months and 9 to 11 months, respectively, with four meals of complementary foods per day. Iron contents in complementary foods were less than the WHO estimated needs from complementary foods (**table 4).** *In vitro* iron availability increased by 60% in FMGEP compared with FMP (**table 4**). On the other hand, processing did not have a significant effect on *in vitro* iron availability, probably due to the low content of that element in complementary porridges. *In vitro* availability of zinc significantly increased in complementary porridges compared with the control porridge FMP. The difference was not observed in the maize–peanuts complementary porridges (FMGEP, FMRP, and FMGRP), whereas in the maize–beans complementary porridges, FMGEB and FMRB showed higher zinc *in vitro* availabilities, 43.62% and 37.04%, respectively, than FMGRB (18.2%). The difference between FMGEB and FMRB was not significant. The increases observed were mainly attributed to a decrease in phytic acid (phytate), which was also observed after processing of maize, peanuts and beans [11, 13]. Phytate may bind strongly to minerals and trace elements in the foods, rendering them unavailable for absorption [9]. Phytate content may be decreased by the action of the enzyme phytase, which is activated during soaking, germination, and lactic acid fermentation [11, 13].

Unlike calcium content, *in vitro* calcium availability decreased with blend formulation, showing that more phytate remained after processing of peanuts and beans than after processing of maize. This may explain the higher phytate inhibitory effect in complementary porridges than in FMP.

Protein quality

The quality of a protein must be viewed in the context of how well the protein can meet essential amino acid and amino nitrogen requirements (amino acid pattern). The nutritive value of a food protein is then a function of its protein content, essential amino acid composition, protein digestibility, and amino acid release.

TABLE 4. Effect of blend and processing methods on some mineral content and availability[1]

Micronutrients	Porridges made with fermented yellow maize flour-processed peanut blends				Porridges made with fermented yellow maize flour-processed bean blends			WHO estimated needs from complementary foods[2]					
								6–8 mo			9–11 mo		
	FMP	FMGEP	FMRP	FMGRP	FMGEB	FMRB	FMGRB	L	A	H	L	A	H
Iron (mg)	0.26 ± 0.04^{d}	0.27 ± 0.03^{d}	0.31 ± 0.04^{cd}	0.5 ± 0.09^{a}	0.39 ± 0.04^{bc}	0.4 ± 0.05^{bc}	0.42 ± 0.05^{a}	2.1* 1.1** 0.7***	2.1 1.1 0.7	2.0 1.1 0.7	1.8 0.9 0.8	1.9 0.9 0.6	1.8 0.9 0.6
Zinc (mg)	0.1 ± 0.01^{c}	0.16 ± 0.02^{b}	0.2 ± 0.02^{ab}	0.22 ± 0.03^{a}	0.18 ± 0.02^{ab}	0.16 ± 0.01^{b}	0.21 ± 0.04^{a}	0.3	0.2	0.2	0.2	0.2	0.2
Calcium (mg)	1.85 ± 0.1^{c}	3.09 ± 0.4^{ab}	3.53 ± 0.68^{ab}	3.1 ± 0.23^{ab}	2.84 ± 0.65^{b}	2.83 ± 0.2^{b}	3.74 ± 0.76^{a}	42	34	25	42	31	22
Iron *in vitro* availability (%)	28.31 ± 2.74^{bc}	45.34 ± 8.26^{a}	32.77 ± 6.2^{abc}	20.27 ± 4.02^{c}	38.17 ± 9.9^{ab}	39.94 ± 3^{ab}	36.34 ± 15.1^{ab}						
Zinc *in vitro* availability (%)	5.98 ± 0.34^{c}	22.49 ± 5.26^{b}	17.41 ± 6.85^{b}	19.23 ± 3.84^{b}	43.62 ± 3.71^{a}	37.04 ± 4.5^{a}	18.2 ± 9.7^{b}						
Calcium *in vitro* availability (%)	62.32 ± 6.3^{a}	23.2 ± 5.2^{d}	21.1 ± 9^{d}	31.9 ± 7.5^{cd}	51.5 ± 5.4^{ab}	39.56 ± 11.5^{bc}	17.62 ± 1.4^{d}						

Abbreviations are defined in table 1 footnotes.

1. Results are the means of three determinations. Plus–minus values are means ± SD. Except for iron, zinc, and calcium availability, values are given per 100 g of porridge. Means within the same row followed by the same letter are not significantly different by the Duncan's new multiple range and the LSMEANS tests ($p > .05$).
2. Data are from WHO/UNICEF [9], and the values were estimated for well-nourished children 6 to 8 months and 9 to 11 months of age, with estimated gastric capacity of 285 and 345 g, respectively, at four servings a day and at the WHO low (L), average (A), and high (H) amounts of breastmilk energy. Energy needs from complementary foods were set at total energy requirement + 2 SD (i.e., + 25%).

*Low, **medium, and ***high iron availability.

In vitro *protein digestibility*

The overall *in vitro* protein digestibility of complementary porridges after 6 hours of digestion ranged from 36.6% to 58.1% due to blend formulation and processing (**fig. 1**). Compared with FMP with an *in vitro* protein digestibility of 51.2%, *in vitro* protein digestibility increased in maize–peanuts complementary porridges (58.1% in FMGEP, 56.3% in FMRP, and 54% in FMGRP), whereas a decrease was observed in maize–beans complementary porridges (50.8% in FMGEB, 36.6% in FMRB, and 51% in FMGRB). These values were comparable to those of Griffith et al. [7], which were obtained by a similar method, but after 4 hours of digestion rather than 6 hours as in the present study. In Griffith et al. [7], *in vitro* protein digestibility ranged from 47% to 54% according to blend formulation (pearl millet–peanut or pearl millet–cowpea with or without teff) and from 48% to 55% according to processing method (roasting, germination, or fermentation). Protein digestibility varied significantly ($p < .05$) in complementary porridges, due to both blend formulation and processing methods. Maize–peanuts complementary porridges were 18% higher in protein digestibility than maize–beans complementary porridges, probably due to the higher fat content of peanuts (**table 2**) than of beans [11]. A comparison of true digestibility indicated that protein digestibility of peanut flour was better than that of soy flour and was comparable to animal protein digestibility [25]. In peanut-based complementary porridges, germination, roasting, and the combination of germination and roasting improved *in vitro* protein digestibility by 5.5%, 10% and 13.6%, respectively, over the control FMP (**fig. 1**). Griffith et al. [7] reported an improvement of 12% to 14% in digestibility in pearl millet–peanuts blends and pearl millet–cowpea blends due to germination and fermentation. In another study, *in vitro* protein digestibility of wheat flour was 30% and it increased to 40% when the flour was supplemented with peanut meal, indicating a considerable improvement in protein digestibility [26]. Alonso et al. [27] reported an increase in bean (*Phaseolus vulgaris*) *in vitro* protein digestibility after dehulling, soaking, and germination compared with the unprocessed seeds. The reduction of antinutritional factors such as phytate and trypsin inhibitors during processing may have contributed to the improved digestibility in maize–peanuts complementary porridges [11, 13] compared with FMP. The decrease in *in vitro* protein digestibility observed in maize–beans complementary porridges compared with FMP was probably due to the higher content of antinutritional factors in processed beans than in the processed maize reported in previous studies [11, 13]. *In vitro* protein digestibility in maize–peanuts and maize–beans complementary porridges showed a significant negative correlation with viscosity (r = –0.54 and –0.79, respectively) (results not shown). This suggests that a higher *in vitro* protein digestibility can be expected from complementary porridges with lower viscosity.

The method used to measure protein digestibility in the present study provided relative values expressed as percent digestibility. The method allowed the effect of addition of food on protein digestibility to be considered, with FMP as the control.

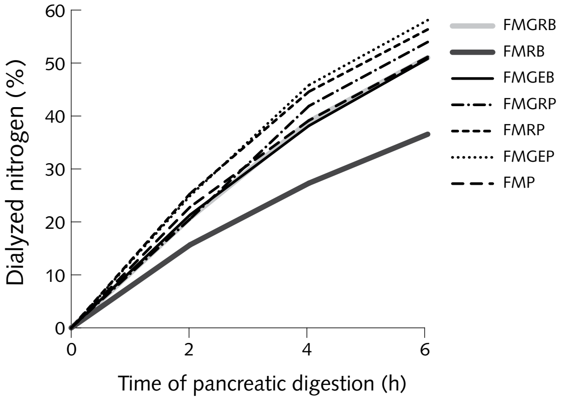

FIG. 1. Effect of blends and traditional processing methods on pancreatic hydrolysis of complementary porridges made with blends of 70% maize and 30% legume as measured in dialyzed nitrogen (means of four repetitions)

FMP, porridge with fermented yellow maize flour (control porridge containing only maize); FMGEP, porridge with 70% fermented yellow maize flour and 30% germinated peanuts; FMRP, porridge with 70% fermented yellow maize flour and 30% roasted peanuts; FMGRP, porridge with 70% fermented yellow maize flour and 30% germinated and roasted peanuts; FMGEB, porridge with 70% fermented yellow maize flour and 30% germinated beans; FMRB, porridge with 70% fermented yellow maize flour and 30% roasted beans; FMGRB, porridge with 70% fermented yellow maize flour and 30% germinated and roasted beans

Essential amino acid release and amino acid score of complementary porridges

Essential amino acid release from the complementary porridges is presented in **figure 2**. There were a variety of significant changes in *in vitro* individual amino acid release in complementary porridges compared with the control porridge. Except for FMGRP and FMGRB, in which the release of methionine was > 60%, the release of methionine was less than 30% in all complementary porridges and varied with blend formulation and processing. *In vitro* release of the aromatic amino acids tyrosine and phenylalanine was above 70% and 50% respectively and did not significantly increase ($p > .05$) with blend formulation and processing, except for phenylalanine in maize–beans complementary porridges, where the release was significantly higher in FMGRB than in FMRB (80% vs. 71%). *In vitro* release of lysine, the limiting amino acid in cereals, increased with blend formulation and was above 50% except in FMRB, where the change was not significantly different ($p > .05$) from that of the control porridge FMP. Infants require histidine, but it is not required by older

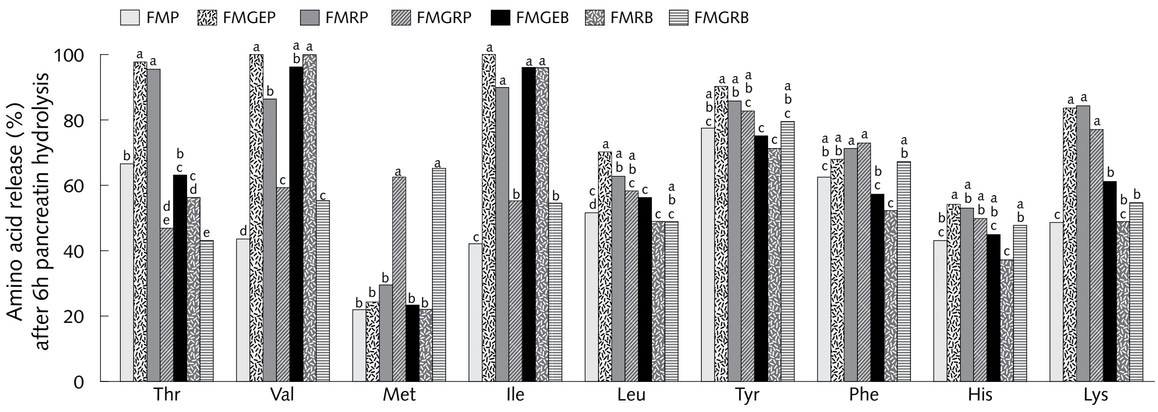

FIG. 2. Percent release of essential amino acids of complementary porridges made with blends of 70% maize and 30% legumes (means of four repetitions). Means within the same essential amino acid with the same letter are not significantly different ($p > 0.05$) according to Duncan's multiple range test

FMP, porridge with fermented yellow maize flour (control porridge containing only maize); FMGEP, porridge with 70% fermented yellow maize flour and 30% germinated peanuts; FMRP, porridge with 70% fermented yellow maize flour and 30% roasted peanuts; FMGRP, porridge with 70% fermented yellow maize flour and 30% germinated and roasted peanuts; FMGEB, porridge with 70% fermented yellow maize flour and 30% germinated beans; FMRB, porridge with 70% fermented yellow maize flour and 30% roasted beans; FMGRB, porridge with 70% fermented yellow maize flour and 30% germinated and roasted beans. Thr, threonine; Val, valine; Met, methionine; Ile, isoleucine; Leu, leucine; Tyr, tyrosine; Phe, phenylalanine; His, histidine; Lys, lysine individuals. The release of histidine did not change significantly due to blend formulation compared with that in FMP, whereas the increase was significant in FMGEP compared with that in FMP.

In the overall *in vitro* amino acid release, the effect of processing was more obvious than the effect of blend formulation in threonine, valine, methionine, and isoleucine release, in which germination and roasting produced higher ($p < .05$) *in vitro* amino acid release than the combination of germination and roasting (**fig. 2**). The same trend was observed for protein *in vitro* digestibility in complementary porridges made from processed peanuts. The effect of blend formulation was observed in leucine and lysine release ($p < .05$). Complementary porridges made from processed peanuts showed the greatest leucine and lysine release ($p < .05$) compared with the porridges made from processed beans.

The Food and Agriculture Organization (FAO) essential amino acid profile [18] was used as a basis for calculating amino acid scores. Despite the significant increase ($p < .05$) observed with blend formulation (0.43, 0.47, 0.50, 0.57, 0.40, and 0.71 for FMGEP, FMRP, FMGRP, FMGEB, FMRB, and FMGRB, respectively), compared with FMP (0.35), the protein chemical score remained below the recommended value of 1.0, indicating that all essential amino acid requirements were not met satisfactorily (**table 5**). Lysine generally had the lowest score in maize–peanuts complementary porridges, indicating that it was the first limiting amino acid, whereas valine had the lowest score in FMGEB and FMPRB. Histidine, an essential amino acid for infants, as well as leucine and phenylalanine + tyrosine, had a satisfactory score greater than 1 in all complementary porridges.

Conclusion

An infant complementary food of higher energy and nutrient density, formulated and prepared from a combination of 70% yellow maize and 30% peanuts or 30% beans, had the strongest impact on nutritional quality and should be viewed as an option in the development of infant complementary foods. Blend formulation had the most important effects on energy density, percent fat, and percent protein. The complementation of fermented yellow maize flour with processed peanuts increased the energy density and improved *in vitro* protein digestibility over complementary porridges with processed beans.

The optimal WHO [9] estimated energy needs from complementary foods for children 6 to 11 months of age, receiving four servings per day and an average amount of energy from breastmilk, was met by maize–peanuts complementary porridges, whereas maize–beans complementary porridges could only

TABLE 5. Essential amino acid scores and amino acid content of complementary porridges[1]

Amino acid	FMP		FMGEP		FMRP		FMGRP		FMGEB		FMRB		FMGRB		FAO/WHO [18]
	Score	Content	Score	Content	Score	Content	Score	Content	Score	Content	Score	Content	Score	Content	content
HIS	1.27	2.40	1.31	2.49	1.35	2.56	1.37	2.61	1.63	3.10	1.68	3.20	1.53	2.91	1.90
ILE	0.49	1.39	0.43	1.21	0.63	1.76	1.04	2.92	0.58	1.63	0.51	1.42	1.12	3.13	2.80
LEU	1.84	1.12	1.27	8.37	1.33	8.81	1.40	9.23	1.44	9.49	1.45	9.54	1.65	11.87	6.60
LYS	0.35	2.04	0.46	2.69	0.47	2.73	0.50	2.90	0.63	3.66	0.74	4.29	0.71	4.14	5.80
MET + CYS	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	2.50
PHE + TYR	1.69	10.65	1.62	10.21	1.57	9.89	1.52	9.57	1.65	10.41	1.68	10.58	1.50	9.43	6.30
THR	0.71	2.40	0.57	1.93	0.50	1.70	1.04	3.54	0.86	2.93	0.81	2.77	1.27	4.30	3.40
TRY	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	1.10
VAL	0.57	1.99	0.44	1.53	0.64	2.25	1.03	3.62	0.57	1.99	0.40	1.41	1.13	3.95	3.50
Chemical score	0.35		0.43		0.47		0.50		0.57		0.40		0.71		

nd, not determined; other abbreviations are defined in table 1 footnotes.
1. Essential amino acid scores were calculated by dividing the amino acid amounts by the values in the reference protein (FAO/WHO [18]), contents are expressed as grams per 100 g of protein.

meet energy recommendations for children aged 9 to 11 months (**table 1**) Mineral content, except for zinc in maize–peanuts complementary porridges, did not meet WHO [10] estimated needs from complementary foods. The complementary porridges were limited in lysine for maize–peanuts complementary porridges and valine for maize–beans complementary porridges, but the histidine score was satisfactory. There was an obvious effect of germination, roasting, and fermentation on viscosity, mineral availability, protein digestibility, and amino acid digestibility, probably due to the reduction of antinutritional factors [11, 13].

On the basis of a comprehensive chemical analysis, this study has demonstrated that some important nutritional benefits could be obtained by combining fermented maize with processed beans or peanuts. The decrease in pH and antinutritional factors of maize after fermentation, coupled with the beneficial effects of processing on peanuts or beans, could contribute to the suitability of these complementary foods. There were some similarities in the effect of blend formulation on complementary porridges, but the overall nutritional value was better in maize–peanuts complementary porridges, making beans apparently less advantageous than peanuts in the formulation of complementary cereal–legume blends.

These findings showed that blend formulation improved the nutritional quality of porridges compared with that of the control FMP made with maize only, but we recommend that the 70:30 ratio of cereals to legumes should be adjusted according to the nutrient and micronutrient contents of each food source used in the formulation of blends and according to the WHO estimated needs from complementary foods. On the other hand, it will be difficult to meet the WHO estimated needs for iron and calcium in complementary foods with blends of cereals and legumes alone. The use of commonly consumed local food supplements of vegetal or animal origin, instead of cereal–legume blends, coupled with public education programs, could help families and communities make better use of their products and at the same time improve nutrient density and meet micronutrient needs.

Acknowledgments

We gratefully acknowledge the Canadian National Sciences and Engineering Research Council (NSERC) for providing grant 18095 for this research.

References

1. Pelletier DL, Frongillo Jr EA, Schoeder DG, Habicht J-P. The effects of malnutrition on child mortality in developing countries. WHO Bull 1995;73(4):443–8.
2. UNICEF. La situation des enfants dans le monde. Table 1: Indicateurs de base. New York: UNICEF, 2006.
3. UNICEF. La situation des enfants dans le monde. Table 2: Nutrition. New York: UNICEF, 2006.
4. Trèche S, Mbome IL. Viscosity, energy density and osmolality of gruels for infants prepared from locally produced commercial flours in some developing countries. Int J Food Sci Nutr 1999;50:117–25.
5. Kannan S, Nielsen SS, Mason A. Protein digestibility-corrected amino acid scores for bean and bean-rice protein based infant weaning food products. J Agric Food Chem 2001;49:5070–4.
6. Mbithi-Mwikya S, Van Camp J, Mamiro PR, Ooghe W, Kolsteren P, Huyghebaert A. Evaluation of the nutritional characteristics of a finger millet based complementary food. J Agric Food Chem 2002;50:3030–6.
7. Griffith LD, Castell-Perez ME, Griffith ME. Effect of blend and processing method on the nutritional quality of weaning foods made from select cereals and legumes. Cereal Chem 1998;75(1):105–12.
8. Egounlety M. Production of legume-fortified weaning foods. Food Res Int 2002;35(2):233–7.
9. WHO/UNICEF. Complementary feeding of young children in developing countries: A review of current scientific knowledge. WHO/NUT/98.1 Geneva: World Health Organization, 1998.
10. Trèche S. Influence de la densité énergétique et de la viscosité des bouillies sur l'ingéré énergétique des nourrissons. Santé: Cahiers d'Etudes et de Recherches Francophones 1996;6(4):237–43.
11. Ejigui J, Savoie L, Marin J, Desrosiers T. Influence of traditional processing methods on the nutritional composition and antinutritional factors of red peanuts (Arachis hypogea), and small red kidney beans (Phaseolus vulgaris). J Biol Sci 2005;5:597–605.
12. Gahlawat P, Sehgal S. Protein and starch digestibility and iron availability in developed weaning foods as affected by roasting. J Hum Nutr Diet 1994;7:121–6.
13. Ejigui J, Savoie L, Marin J, Desrosiers T. Beneficial changes and drawbacks of a traditional fermentation process chemical composition and antinutritional factors of yellow maize (Zea mays). J Biol Sci 2005;5:590–6.
14. Mosha AC, Svanberg U. Preparation of weaning foods with high nutrient density using flour of germinated cereals. Food Nutr Bull 1983;5:10–4.
15. Bressani R. Grain quality of common beans. Food Rev Int 1993;9(2):237–97.
16. Savoie L, Gauthier SF. Dialysis cell for the in vitro measurement of protein digestibility. J Food Sci 1986;51: 494–8.
17. AOAC. Official methods of analysis of the Association of Official Analytical Chemists. 16th ed. Methods 979.09, 925.10, 945.16. Arlington, Va, USA: AOAC International, 1995.
18. FAO/WHO. Protein Quality Evaluation, Report of the Joint FAO/WHO Expert Consultation, Rome: FAO Food

and Nutrition Paper No. 51, 1991.

19. Barhisel R, Bertsch PM. Digestion with perchloric-nitric acids. *In* AL Page ed. Methods of soil analysis. SSSA Book Ser. 5. SSSA, Madison, 1982;2:279–80.
20. Miller DD, Schricker BR, Rasmussen RR,Van Campen D. An in vitro method for estimation of iron availability from meals. Am J Clin Nutr 1981;34:2248–56.
21. Nago CM, Télégan E, Matenciot F, Mestres C. Effects of maize type and fermentation conditions on the quality of Beninese traditional ogi, a fermented maize slurry. J Cereal Sci 1998;28(2):215–22.
22. Ward D, Ainsworth P. The development of a nutritious low cost weaning food for Kenyan infants. Afr J Health Sci 1998;5:89–95.
23. Dewey KG, Brown KH. Update on technical issues concerning complementary feeding of young children in developing countries and implications for intervention programs. Food Nutr Bull 2003;24:5–28.
24. Walker AF. The contribution of weaning foods to protein–energy malnutrition. Nutr Res Rev 1990;3(1):27–47.
25. Singh B, Singh U. Peanut as a source of protein for human foods. Plant Foods Hum Nutr 1991;41:165–77.
26. Brulé D, Savoie L. *In vitro* protein digestibility of protein and amino acids in protein mixtures. J Sci Food Agric 1988;43:361–72.
27. Alonso R, Aguirre A, Marzo F. Effects of extrusion and traditional processing methods on antinutrients and in vitro digestibility of protein and starch in faba and kidney beans. Food Chem 2000;68(2):159–65.
28. Steinke FH, Hopkins DT. Complementary and supplementary effects of vegetable proteins. Cereal Foods World 1983;28:338–41.

Acceptability and safety of novel infant porridges containing lyophilized meat powder and iron-fortified wheat flour

Helena Pachón, María Reyna Liria Domínguez, Hilary Creed-Kanashiro, and Rebecca J. Stoltzfus

Abstract

Background. *Lyophilized meat powder with iron-fortified wheat flour can be used to produce an infant porridge with bioavailable iron, but its acceptability and safety are unknown.*

Objective. *To evaluate the acceptability and safety of porridges containing lyophilized meat powder and iron-fortified wheat flour.*

Methods. *Peruvian mothers' input was used to develop porridges without (no meat) and with meat powder (low or high chicken liver, low or high chicken thigh). Acceptability was determined by maternal hedonic scoring, 9-day infant intake, and videotape analysis of how well infants liked each porridge. Dry and cooked porridges and meat ingredients were tested for microorganisms; meats were tested for pesticides.*

Results. *Mothers gave higher acceptability scores to the no-meat porridge, followed in order by low and high quantities of meat powder (e.g., mean ± SD "taste" scores were 4.5 ± 0.9 for the no-meat, 3.7 ± 1.1 for the low-liver, and 3.3 ± 1.1 for the high-liver porridges,* $p = .0001$*). Infants' porridge intake did not differ: 61.4 ± 47.1 g of no-meat, 62.1 ± 44.9 g of low-thigh, and 67.5 ± 42.0 g of low-liver (*$p = .7$*), as supported by the video analysis. Microbiologic safety was acceptable except for marginally acceptable molds and yeasts in dry ingredients. No pesticide residues were detected.*

Conclusions. *Despite mothers' clear preference for no-meat porridges, infants consumed equal amounts of porridges with and without meat. Thus, if mothers can be convinced to feed the meat-containing porridges to the infants despite their own preferences, the infants will consume these porridges. The mold and yeast content of the porridge ingredients must be reduced.*

Key words: Acceptability, bioavailability, complementary feeding, complementary food, infant feeding, intervention, iron, Peru, porridges, sensory characteristics

Introduction

Anemia, resulting from iron deficiency or other causes, affects an estimated 2 billion people around the world [1]. Anemia is of concern in infants because it has been associated with impaired mental and motor development [2] and retarded growth [3]. Infants 6 months of age and older in developing countries are especially vulnerable because the complementary foods offered to them are often not iron dense, and breastmilk alone does not provide sufficient quantities of iron to meet infants' requirements. Thus, the challenge in improving the iron status of infants from 6 months onward is to regularly provide them with *bioavailable* iron in sufficient quantities at mealtimes. This could be achieved by feeding infants iron-fortified foods coupled with an iron-absorption enhancer [4].

High-quality and low-cost iron-fortified infant foods are not common in developing countries [5]. Nevertheless, food-fortification efforts have made iron-fortified staple foods, not necessarily directed at children, available and at relatively low cost. A study from Peru has demonstrated the effectiveness of ferrous sulfate–fortified wheat products on iron status among 41 stunted and moderately anemic children 3 to 4 years of age [6]. These children were given wheat products (biscuits or noodles) with ferrous sulfate (3 mg iron per 100 g wheat product), plus or minus zinc fortificant, daily for 7 weeks. In the iron-only group, hemoglobin increased from 106.0 to 129.6 g/L ($p < .001$) and no child was

Helena Pachón and Rebecca J. Stoltzfus are affiliated with the Division of Nutritional Sciences, Cornell University, Ithaca, New York, USA; María Reyna Liria Domínguez and Hilary Creed-Kanashiro are affiliated with the Instituto de Investigación Nutricional, Lima, Peru.

Please direct queries to the corresponding author: Helena Pachón, Centro Internacional de Agricultura Tropical (CIAT), Apartado Aéreo 6713, Cali, Colombia; e-mail: h.pachon@cgiar.org.

anemic at the endpoint. In addition to 100 g of fortified wheat products daily, children ate their baseline diet throughout the study. The authors commented that it is "notable that children's hemoglobin concentrations increased in response to the dietary treatments with iron-fortified foods, even though these foods delivered a net increase of only ~1 mg/d [of iron]."

With any type of fortified food, the type of fortificant used can limit the amount of ingested iron that is absorbed; promoters such as meat or ascorbic acid must also be present to enhance iron absorption [4]. Meat, in addition to being an excellent enhancer of iron absorption, contributes nutrients that are lacking in the diets of infants in developing countries: iron, zinc, and vitamin B_{12} [7–9]. Meats are available in developed and developing countries [10]; however, in most settings, meats are given to young children infrequently and only in small quantities [11–18]. This is probably due to a combination of parents' beliefs and perceptions of appropriate first foods [19–20], the recommendations of health personnel [21] and family members [22] about what foods to introduce, cost [23], availability in the home [24], and infants' innate preference for sweet foods [25].

Recent research in human adults [26] has shown that adding lyophilized meat powder to an iron-fortified food increases by 85% the intake of absorbable iron from the diet. Further, these substantial improvements in iron absorption were observed with small amounts of lyophilized meat (equivalent to 20 g of meat) combined with small amounts of an iron-fortified food (48 g). Compared with nonlyophilized meat, lyophilized meat powder has a longer shelf life and is a more concentrated source of nutrients [27].

Before recommending that infants eat such a combination, it needs to be determined whether parents will feed these meat-fortified food combinations, infants will consume the combinations if offered them, and these foods are safe for infants to consume. Therefore, the objectives of this study were to evaluate mothers' and infants' acceptance of recipes containing lyophilized meat powder and iron-fortified wheat flour and assess the microbiologic and pesticide safety of the lyophilized meat powder and meat-flour recipes.

Methods

To meet the study objectives, five activities were carried out in June to August 2003 and September to December 2004: recipe-creation exercises, maternal acceptability trial, infant acceptability trial, infant video taping, and microbiological and pesticide-residue testing. The study activities were approved by the institutional review boards of the Instituto de Investigación Nutricional in Peru and Cornell University in the United States. Written informed consent was obtained from all women who participated in any study activity. All participants received a token gift.

Study setting

Study activities were carried out in an urban shanty town in southern Lima, Peru. Approximately 8,000 families live in the community, which was founded in January 2000. The community lacks basic services such as running water, indoor plumbing, paved roads, and a reliable electricity supply.

Recipe-creation exercises

Recipe-creation exercises [19] were held in 2003 and 2004. The purpose of the first exercise was for mothers of infants to create recipes that combined meats as they are sold in the community (Nestlé beef with vegetables, alpaca *charqui* (dried meat), chicken thigh, chicken liver, chicken spleen, chicken blood) with locally available iron-fortified food (Gerber infant rice cereal, Papimás infant wheat cereal, iron-fortified pasta, iron-fortified wheat flour). The purpose of the second exercise was for mothers to create porridges that combined lyophilized meat powder (chicken thigh, chicken liver) with iron-fortified wheat flour (donated by Alicorp SA, Peru); other ingredients were also provided (powdered whole milk (Leche Anchor, Nestlé Peru), brown sugar, cinnamon sticks or powder, whole or powdered cloves, vanilla extract). Mothers' input was used to develop the recipes tested during the maternal and infant acceptability trials.

Preparation of lyophilized meat powder

Raw chicken liver and heart and chicken thigh were purchased from a local chicken producer (San Fernando, Lima, Peru). At the Instituto de Investigación Nutricional (IIN), visible fat and the heart were removed from the chicken liver, and visible fat, skin and bone were removed from the chicken thigh. The chicken liver and thigh were cut into small cubes, and 1 kg of each was cooked for 12 minutes in 50 mL of water in a microwave oven (LG model MS-114YFA). After cooling at room temperature for 30 minutes, the cooked chicken was packaged in 0.5-mm-thick polyethylene bags and stored at –180°C with dry ice.

The cooked meat was transported to a commercial lyophilization plant (Liofilizadora Pacífico) where the meat was processed in a lyophilizer (Leybold Hereaus, Model GT12) at platen temperature of 38°–42°C at a pressure of 0.3 mBARS for 23 hours. The lyophilized meat was then ground to a fine powder with a company-made hammer mill using a 600-micron mesh.

Energy and nutrient profile of porridges

The porridges used during the maternal and infant acceptability trials contained ingredients available in Lima: fortified powdered whole milk, fortified wheat flour, brown sugar, meat, and vanilla powder (SD 18798 donated by Montana SA). The reported energy and nutrient contents of these ingredients were used to develop isocaloric recipes.

According to the label, the milk contains 500 kcal of energy, 16.2 g of protein, 26.6 g of fat, 9.2 mg of iron (as ferric pyrophosphate; Rudy Campos, Nestlé Perú, personal communication), 1.7 mg of zinc, 350 μg RE of vitamin A and 50 mg of ascorbic acid per 100 g. The donated wheat flour was formulated with the iron (5.5 mg as ferrous fumarate/100 g), thiamin (0.5 mg/100 g), riboflavin (0.4 mg/100 g), niacin (4.8 mg/100 g), and folic acid (120 μg/100 g) levels recently enacted by Peruvian national decree [28]; other values were taken from the IIN food composition table [29]: 359 kcal of energy, 10.5 g of protein, 2.0 g of fat, 0.7 mg of zinc, 0 μg RE of vitamin A, and 1.8 mg of ascorbic acid per 100 g. According to the IIN food composition table, the brown sugar contains 380 kcal of energy, 0 g of protein, 0 g of fat, 1.7 mg of iron, 0.2 mg of zinc, 0 μg RE of vitamin A, and 0 mg of ascorbic acid per 100 g. Given that the final weight of lyophilized liver powder is 19.5% of the weight of raw liver, and assuming that no nutrients were lost during the cooking and lyophilization process, the IIN food-composition table values for liver were multiplied by 5.13 (100/19.5) to obtain the liver powder values. By a similar process, the energy and nutrient profile of the chicken thigh powder was obtained (multiplication factor of 4.57). In summary, the liver powder was estimated to have 641 kcal of energy, 92 g of protein, 20 g of fat, 44 mg of iron, 16 mg of zinc, 31,626 μg RE of vitamin A, and 173 mg of ascorbic acid per 100 g and the thigh powder to have 544 kcal of energy, 90 g of protein, 18 g of fat, 5 mg of iron, 9 mg of zinc, 91 μg RE of vitamin A, and 14 mg of ascorbic acid per 100 g. No energy or nutrient contribution was estimated for the vanilla powder. The energy and nutrient profile of the porridges was estimated from these ingredient values (**table 1**).

TABLE 1. Energy and nutrient profile and ingredients (per 100 g of cooked porridge) of five recipes tested during the acceptability trials with mothers and infants[a]

	Porridge[b]				
Component	No meat	Low liver	High liver	Low thigh	High thigh
Energy and nutrients					
Energy (kcal)	115.1	115.0	115.1	115.5	115.4
Protein (g)	2.9	3.5	3.8	5.0	5.6
Fat (g)	3.4	3.1	3.0	3.2	3.0
Calcium (mg)	120.9	107.3	98.2	98.1	84.4
Iron (mg)[c]	1.5	1.8	2.0	1.4	1.3
Zinc (mg)	0.3	0.4	0.5	0.5	0.5
Vitamin A (μg RE)	42.0	353.0	507.6	36.0	31.7
Ascorbic acid (mg)	6.2	7.1	7.5	5.3	4.7
Ingredients					
Fortified whole milk powder (g)	12.0	10.5	9.5	9.5	8.0
Fortified wheat flour (g)	9.0	8.0	8.0	7.0	7.0
Brown sugar (g)	6.0	7.2	7.7	7.0	7.5
Vanilla powder (g)	0.02	0.06	0.08	0.04	0.06
Liver powder (g)	0	1.0	1.5	0	0
Thigh powder (g)	0	0	0	3.0	4.0

a. The energy and nutrient values for these foods were obtained from multiple sources: the food label for milk powder; the national decree for wheat flour iron, thiamin, riboflavin, niacin, and folic acid levels [28]; and the Instituto de Investigación Nutricional (IIN) food-composition table for other nutrients [29]. No energy or nutrient contribution was estimated for the vanilla powder.

b. The no-meat porridge contained ferrous fumarate–fortified wheat flour, ferric pyrophosphate–fortified whole milk powder, brown sugar, and vanilla powder. The liver and thigh porridges contained the same ingredients plus lyophilized meat powder, as follows: low liver (1.0 g liver powder/100 g cooked porridge), high liver (1.5 g liver powder/100 g cooked porridge), low thigh (3.0 g chicken thigh powder/100 g cooked porridge), and high thigh (4.0 g chicken thigh powder/100 g cooked porridge).

c. Iron-fortified whole milk powder, iron-fortified wheat flour, meat powders, and brown sugar contributed the iron in the porridges. Specifically, for the no-meat porridge, the iron came from the milk (1.2 mg), flour (0.3 mg) and sugar (0.1 mg). For the low-liver porridge, the iron came from the milk (1.0 mg), liver powder (0.4 mg), flour (0.3 mg), and sugar (0.1 mg). For the high-liver porridge, the iron came from the milk (0.9 mg), liver powder (0.7 mg), flour (0.3 mg), and sugar (0.1 mg). For the low-thigh porridge, the iron came from the milk (0.9 mg), flour (0.3 mg), thigh powder (0.1 mg), and sugar (0.1 mg). For the high-thigh porridge, the iron came from the milk (0.7 mg), flour (0.3 mg), thigh powder (0.2 mg), and sugar (0.1 mg).

Maternal acceptability trial

From a master list of community families with infants, all women with an infant 5 to 11 months of age (n = 274) were assessed for eligibility to participate in the acceptability trial. Mothers were excluded (n = 80) if their children were < 6 months or > 11 months old (n = 67) or had a condition that interfered with their ability to eat such as cleft lip (n = 1), or if the mother lived outside the community (n = 12). Among the women eligible to participate (n = 194), 43 were not located; therefore, 151 women were invited to participate. Of these, 61 did not participate because of refusal (n = 9), because they did not show up on the testing date (n = 44), or for an unknown reason (n = 8). A total of 90 women participated in the acceptability trial.

Women were randomly divided into two groups (liver-tasters and thigh-tasters); each woman tasted and evaluated three porridges: liver tasters tried the no-meat, low-liver (1.0 g liver powder/100 g cooked porridge), and high-liver (1.5 g liver powder/100 g cooked porridge) porridges, and the thigh tasters tried the no-meat, low-thigh (3.0 g chicken thigh powder/100 g cooked porridge), and high-thigh (4.0 g chicken thigh powder/100 g cooked porridge) porridges.

The acceptability form had been used previously with women living in a Lima shantytown. The form required that the woman be able to write. For any woman who requested help (n = 10), a member of the study team sat with her and filled out the form with her. The form asked women to evaluate with numbers and words the smell, taste, texture, color, consistency, and overall characteristics of each porridge. The numerical scales included a five-item Likert-type scale where cartoon faces and words ranged from "dislike very much" (1) to "like very much" (5). In addition, the women were asked to rank the porridges from most to least liked. It took the women about 30 minutes to complete the form.

The trial was carried out in the large living room of a home located near the entrance to the community. Two pieces of plywood were arranged on top of a picnic-size table to create cubicles for four women. The cubicles were created on the two large tables in the room, creating space for eight women to carry out the trial simultaneously, without influence from the other women.

The porridges were prepared at the testing site; all three porridges were served at one time to each woman. There were six possible combinations for serving three porridges to each woman: no meat, low meat, high meat; no meat, high meat, low meat; low meat, high meat, no meat; low meat, no meat, high meat; high meat, low meat, no meat; and high meat, no meat, low meat. Each of these combinations was randomly counterbalanced among the women. Food samples of about 30 g were served to women in 90-ml transparent, disposable plastic cups that were marked with a three-digit code that identified the porridge and another three-digit code that identified the mother.

Infant acceptability trial

An updated list of community women with an infant 5 to 11 months of age (n = 277) was reviewed to identify eligible participants for the infant acceptability trial. For a variety of reasons, 208 women were excluded: mother did not participate in recipe exercise or acceptability trial (n = 82), infant < 6 months or > 9 months (n = 81), no date of birth for infant (n = 34), no address (n = 6), infant with birthweight < 2.5 kg (n = 3), infant with Down syndrome (n = 1), and infant with cleft palate (n = 1). Among the eligible women (n = 69) with infants 6 to 9.9 months of age, 8 were not located. Therefore, 61 women were invited to participate; of these, 12 declined to participate. A total of 49 mother–infant pairs participated in the home-based, 9-day, infant acceptability trial.

In the trial, the infants were given three porridges for 3 days each in their homes. The three porridges were each fed at one sitting on 3 consecutive days to the infant. The six possible orders of presenting the porridges (e.g., no meat, low liver, and low thigh) were randomly counterbalanced among the participants.

On days 1 and 9, a qualitative 24-hour dietary recall was administered to capture the infant's dietary pattern. At the first visit, the infant's health status in the previous week was also ascertained. In subsequent home visits, the infant's health status was determined prospectively by the mother's report.

Infants sick with diarrhea, vomiting, fever, or cough with phlegm that affected their appetite (by maternal report) were not fed porridge that day. They were visited daily (up to 6 days/week) and fed the porridge on the 9 days when not sick (although the infant could be recuperating). Some infants were visited up to 18 days to obtain the 9 days of intake data.

Prior to preparing the porridge, mothers were asked the last time the baby was fed (any drink, food, or breastmilk), and an attempt was made to wait at least 30 minutes from the previous feeding to the feeding of the baby. Mothers were also asked to assess the infant's appetite on that day. Mothers were instructed to mix the dry ingredients with 180 ml of water in a pot, remove lumps, and cook for about 3 minutes until the porridge had boiled and thickened. They were then asked to serve and feed the porridge to the infant. The weight of the cooked porridge and the amounts served and fed to the baby were weighed on a digital scale (Soehnle DC and Vera scales). When the mother noted that the baby was full, she was asked to wait about 5 minutes and to try feeding the baby again. At the end of the feeding session, the mothers were asked how well they thought the baby ate the porridge and whether or not this was the amount the baby typically consumed. Using a four-item scale, the fieldworker also independently evaluated how the infant ate the porridge (i.e., rejected, accepted with dislike, accepted with indifference, accepted with pleasure).

Videotaping infant acceptability trial

On the basis of logistics (when the camera and camera operator were available), a subset of 10 mother–infant pairs were selected to have their nine feeding sessions videotaped for subsequent analysis of how well the infant liked each of the recipes. The sessions were videotaped with a Hi-8 video camera (Sony CCD-TRV608 NTSC). Only the mother's code and the porridge code were written on the tapes so that the scorers would be blind to the day of the feeding session and the porridge being fed to the infant. In the United States, three scorers reviewed in real time the first 2 minutes of each feeding session to ascertain initial acceptance, as done by Mennella et al. [30]. The scorer coded the infant's acceptance of the porridge on a scale of 1 (infant consistently rejects food) to 5 (infant always accepts food), following the coding of Sullivan and Birch [31].

Microbiological and pesticide residue testing

Three groups of food were microbiologically tested: raw meat and cooked meat powder, porridge dry ingredients, and cooked porridge. Raw meat (chicken thigh, chicken liver) was refrigerated at 4°–8°C and sampled at one time point within approximately 24 h of purchase of the meat. The cooked, lyophilized, and ground meat powder was analyzed at days 5, 30 and 60 after having been cooked, lyophilized, ground, and stored at 4–8°C. The meat samples were tested at the IIN microbiological laboratory for *Escherichia coli*, *Clostridium perfringens,* and *Salmonella* by standard assays [32]. The dry ingredients for the porridges tested in the infant acceptability trial were packaged in 0.5-mm-thick polyethylene bags and stored at room temperature in the study community for 3 weeks. The dry ingredients were tested for aerobic mesophiles, molds and yeast, and *Salmonella* upon packaging and after 3 weeks of storage. The porridges tested during the infant acceptability trial were cooked and stored with a plate covering them (a common practice in the study community) at room temperature in the IIN laboratories and tested for aerobic mesophiles and *E. coli* at 2, 3, 5, and 7 hours post-cooking.

After about 1 year of cold storage, chicken liver and chicken thigh powders were sent to Covance Laboratories (Madison, WI, USA) for pesticide residue testing (FDA PAM 304 screen). The presence of organochlorinated or organophosphate compounds was assessed.

Statistical methods

For the maternal acceptability trial, the numerical scores women gave to the six sensory characteristics of three porridges were compared by ANOVA and the rank data were analyzed by rank-ordered logistic regression (Stata version 8.2). For the infant acceptability trial, the amount consumed in grams and the acceptability score assigned by the video coders to the three different porridges were compared by a repeated-measures ANOVA (SAS version 9). To assess intra- and interscorer reliability prior to video coding, a weighted kappa coefficient was calculated by Stata, which allowed for concordant scoring to include a deviation of ± 1 unit (e.g., scoring the same video as 3 and 4).

Results

Recipe-creation exercises

For the recipe-creation exercises, the convenience samples consisted of 25 women with infants 5 to 8 months in 2003 and 21 women with infants 4 to 17 months in 2004. In 2003, the participants did not combine savory foods (such as meats) with sweet foods (such as the Gerber and Papimás infant foods). However, they did combine the meats with iron-fortified pasta; the consistency of these combinations varied between liquid (such as soups) and thick (such as purées). Further, because infants about 5 to 8 months of age do not have teeth and because of fear of infants choking, the mothers preferred to feed them easily mashed food such as chicken liver and not sinewy food such as muscle tissue (e.g., chicken, beef).

With the use of mothers' input obtained through the recipe-creation exercises, five isocaloric recipes were developed in 2004: one porridge without meat and two recipes each for the chicken liver powder and the chicken thigh powder (**table 1**). A commercially produced vanilla powder (Vanilla SD 18798) made of natural ingredients (as required for cereal-based infant foods per Codex Alimentarius and Peruvian norms [33, 34]) was donated by a local producer (Montana SA).

On the basis of a previous study carried out in a Lima shantytown with 6-month-old infants consuming a wheat-flour-based infant porridge, infants could be expected to consume about 100 g per serving (Nelly Zavaleta, IIN, personal communication). For 100 g of the final cooked porridge, the goal was to maintain the vitamin A content below 600 µg RE, which is the tolerable upper intake level (UL) recommended for infants 7 to 12 months of age [35], and to have an energy density of at least 1 kcal/g of cooked porridge [36]. The vitamin A UL substantially limited the amount of liver powder that could be added to the porridge.

Participants in 2004 recipe exercise and maternal and infant acceptability

In the maternal acceptability trial in 2004, 90 women with infants 3 to 12 months of age participated. A total of 49 mother–infant pairs participated in the infant acceptability trials; the infants were 6 to 9 months old. The pairs yielded 504 infant-days of data. Of these, 151 infant-days were excluded from the analyses because the infant was sick with fever, cough with phlegm, diarrhea, loose stools, vomiting, or stomach infection or was convalescing (n = 66), the time since the last feeding was ≤ 30 minutes (n = 52), the infant was not at home (n = 26), the amount of porridge served was greater than the amount cooked (n = 2), the type of porridge served was unknown (n = 2), or the time since the last feed was unknown (n = 2). A total of 353 infant-days were analyzed. Since some women participated in multiple research activities (recipe exercise, maternal acceptability, infant acceptability), a total of 111 different women participated in 2004.

In 2004 the infants were 8.0 ± 2.4 (SD) months old on average, 45.1% were girls, 94.4% were currently breastfeeding, and 80.3% had eaten meat at least once prior to the study. On average, the women who participated in 2004 were 25.9 ± 5.9 years old, had a husband or partner (87.4%), had 2.1 ± 1.2 living children, and had 7.8 ± 3.4 years of formal education. Few (18.9%) reported working. The participants' homes primarily had dirt or sand floors (62.2%), plywood walls (76.6%), and tin roofs (80.2%).

Maternal acceptability

Of the 90 women, 39 tasted thigh-containing recipes and 51 tasted liver-containing recipes. The same numeric score was used by 39 women each for liver and thigh. All 90 used the same scale to rank their preferred porridges.

With the exception of porridge consistency, the women who tasted the liver-containing recipes gave the highest characteristic scores to the no-meat porridge, followed by the low-liver and the high-liver porridges (**table 2**). Similarly, the women who tasted the thigh-containing recipes gave the highest taste, texture, and overall scores to the no-meat porridge, followed by the low-thigh and the high-thigh porridges.

After factors that could influence the scores women gave to the porridge characteristics had been controlled for (order in which porridge was tested, woman's birthplace, whether or not the child had been fed meat already, whether or not the woman participated in the recipe-creation exercise), the porridge type consistently emerged as an important factor (data not shown). These covariates did not alter the conclusion that porridge type significantly influenced most of the women's sensory scores.

By simple rank-ordering, it was clear that addition

TABLE 2. Unadjusted scores (range, 1 to 5) of porridges tested during the maternal acceptability trial[a]

	No meat	Low liver	High liver	Unadjusted p
Liver tasting (n = 39)				
Smell	4.46 ± 0.96	3.64 ± 1.20	3.28 ± 1.32	.0001
Taste	4.51 ± 0.88	3.74 ± 1.12	3.38 ± 1.09	< .0001
Texture	4.10 ± 1.13	3.53 ± 1.27	3.11 ± 1.27	.0025
Color	4.28 ± 1.17	3.62 ± 1.18	3.56 ± 1.17	.0125
Consistency	4.05 ± 1.15	3.82 ± 1.14	3.82 ± 1.17	.6000
Overall	4.44 ± 1.07	3.90 ± 1.14	3.62 ± 1.21	.0069
Thigh tasting (n = 39)				
Smell	3.85 ± 1.20	3.49 ± 1.34	3.28 ± 1.23	.1393
Taste	4.41 ± 0.88	3.44 ± 1.35	3.00 ± 1.26	< .0001
Texture	4.24 ± 0.94	3.50 ± 1.08	3.05 ± 1.15	< .0001
Color	4.00 ± 1.10	3.85 ± 1.16	3.51 ± 1.32	.1885
Consistency	4.00 ± 1.08	4.03 ± 0.93	3.63 ± 1.24	.2118
Overall	4.33 ± 0.90	3.61 ± 1.26	3.38 ± 1.31	.0014

a. All values are means ± SD. Unadjusted p values were calculated by analysis of variance (ANOVA). Among 90 women, 39 tasted thigh-containing recipes and 51 tasted liver-containing recipes. Among the liver tasters, 39 used the same numeric scales to evaluate the porridge characteristics as the thigh tasters and 12 did not. Therefore, for the thigh- and liver-containing recipes, the porridge characteristics are based on 39 women each. The no-meat porridge contained iron-fortified wheat flour, whole milk powder, brown sugar and vanilla powder. The liver and thigh porridges contained the same ingredients plus lyophilized meat powder, as follows: low liver (1.0 g liver powder/100 g cooked porridge), high liver (1.5 g liver powder/100 g cooked porridge), low thigh (3.0 g chicken thigh powder/100 g cooked porridge), and high thigh (4.0 g chicken thigh powder/100 g cooked porridge). Estimate of texture was based on how the porridge felt on the tongue. Estimate of consistency was based on observation of a spoonful of porridge lifted out of the cup and allowed to drop back into the cup.

of either meat decreased mothers' ranking of the porridges. Thigh-tasters (n = 39) ranked the no-meat porridge as the porridge they liked the most (84.6%), followed by the low-thigh (12.8%) and the high-thigh (2.6%) porridges ($p < .001$). The liver-tasting women (n = 51) ranked the porridge they liked the most as follows: no meat (76.5%), low liver (19.6%), and high liver (3.9%) ($p < .001$).

Infant acceptability

Infants' unadjusted daily intake of the porridges was not statistically significantly different (p = .7): 61.4 ± 47.1 g of the no-meat, 62.1 ± 44.9 g of the low-thigh, and 67.5 ± 42.0 g of the low-liver. Porridge remained a statistically nonsignificant predictor (p = .69) of infant's intake when adjusted for time since last feed, mother's report of infant's appetite prior to feeding, infant's age, percentage of the final cooked porridge that was fed to the infant, the fieldworker, whether or not the infant had eaten meat before, mother's age, and the energy density of the porridge. Mothers' and field workers' assessments of how the infants liked the porridge were statistically significantly correlated with infant intake (Spearman's rho = −0.52, $p < .0001$; Spearman's rho = 0.49, $p < .0001$, respectively).

Infant videotaping

Intraobserver and interobserver reliability was high (the kappa coefficient ranged from 0.74 to 1.0). Unadjusted video scores (ranging from 1 to 5) given to infants' acceptance by scorers did not significantly differ according to porridge type (p = .99): 3.8 ± 1.0 for the no-meat, 3.8 ± 1.2 for the low-thigh, and 3.8 ± 0.8 for the low-liver porridge. After controlling for position of the feeder with respect to the infant, the number of spoonfuls offered to the infant in the 2-minute video segment, and the proportion of spoonfuls offered that the infant consumed, porridge type remained a nonsignificant predictor of the video score (p = .85). To explore relationships between intake by the infant (in grams) and the mothers' and fieldworkers' assessment of acceptance by the infant, the sample size was reduced to 57 child-days due to exclusions for infant intake described earlier. Infant intake and video score were not associated (Spearman's rho = 0.19, p = .16). Mothers' assessments of how well the infants liked the porridge were not associated with video acceptance scores (Spearman's rho = −0.20, p = .14); fieldworker assessments were associated with video scores (Spearman's rho = 0.35, $p < .01$).

Microbiological and pesticide residue testing

At all four time points (0 [raw meat], and days 5, 30, and 60 post-lyophilization), the chicken liver and chicken thigh had < 10 CFU/g of *C. perfringens* and no *Salmonella* present/25 g. The raw meat had higher *E. coli* counts (most probable number [MPN] 49/g and 71/g for chicken liver and chicken thigh, respectively) than the lyophilized meat powder that had been stored for 5, 30, or 60 days at 4°–8°C (MPN < 3/g). According to the Codex Alimentarius [33], supplementary foods for older infants and young children should be free of pathogenic microorganisms. The meat powders met this standard, since *C. perfringens* and *E. coli* were not detectable and *Salmonella* was absent.

In a three-class sampling plan, two values are used to distinguish acceptable from unacceptable microbiological quality: m and M, where > M is unacceptable, > m and < M is marginally acceptable, and < m is acceptable [37]. The porridge dry ingredients at packaging and after 3 weeks of room-temperature storage had no detectable *Salmonella* and met the Peruvian standards for acceptable aerobic mesophiles (< $m = 10^4$ CFU/g where m differentiates good quality from marginally acceptable quality) [38, 39] (**table 3**). In contrast, the porridge dry ingredients were marginally acceptable for molds and yeast ($m = 10^2$ CFU/g, M = 10^4 CFU/g where M distinguishes marginally acceptable from unacceptable quality) [38, 39].

The cooked porridges met Peruvian standards for

TABLE 3. Microbiological results from assay of porridge dry ingredients immediately after packaging and after 3 weeks of storage at room temperature in the study community

Porridge[a]	Aerobic mesophiles (CFU/g)[b]	Molds and yeasts (CFU/g)[b]	*Salmonella* (present or absent/25 g)
Standards used[c]			
m	10^4	10^2	Absent
M	10^6	10^4	
After packaging			
No meat	3×10^2	1.7×10^2	Absent
Low liver	4×10^2	3.6×10^2	Absent
Low thigh	3×10^2	1.7×10^2	Absent
After 3 wk storage			
No meat	2×10^2	2.3×10^3	Absent
Low liver	2×10^2	6×10^2	Absent
Low thigh	2×10^2	4.7×10^2	Absent

CFU, colony-forming unit

a. The no-meat porridge contained iron-fortified wheat flour, whole milk powder, brown sugar, and vanilla powder. The liver and thigh porridges contained the same ingredients plus lyophilized meat powder, as follows: low liver (1.0 g liver powder/100 g cooked porridge) and low thigh (3.0 g chicken thigh powder/100 g cooked porridge).

b. The assay cannot detect values < 10 CFU/g.

c. In a three-class sampling plan, two values are used to distinguish acceptable from unacceptable microbiological quality: m and M, where > M is unacceptable, > m and < M is marginally acceptable, and < m is acceptable [37]. The standards were obtained from the International Commission on Microbiological Specifications for Foods [39].

aerobic mesophiles (< $m = 10^4$ CFU/g) [39] (**table 4**) and had undetectable levels of *E. coli* after 2, 3, 5, and 7 hours of standing at room temperature after cooking.

No pesticide residues were detected in the chicken liver powder or the chicken thigh powder (< 0.2 ppm of organochlorinated residues and < 0.05 ppm of organophosphates).

Discussion

Porridges containing lyophilized meat powder were evaluated for safety and for acceptability by mothers and infants living in an urban shantytown in Lima, Peru. Consistently, mothers gave higher acceptability scores to organoleptic characteristics (smell, taste, texture, color, consistency, and overall acceptability) of the porridge containing no meat, followed by porridges containing low quantities of meat powder and higher quantities of meat powder. In comparison, infants' intake (used as a proxy of acceptance) was the same for porridges containing no meat, low quantities of chicken liver powder, and low quantities of chicken thigh powder. The infants' lack of a preference for a porridge was supported by the video analysis. The lyophilized meat powder was microbiologically safe up to 2 months after it was processed. Similarly, the cooked porridge met microbiological standards for up to 7 hours of storage at room temperature. However, the dry porridge ingredients had only marginal acceptability with respect to molds and yeasts upon packaging and 3 weeks post-storage at room temperature. No pesticide residues were detected in the meat powders.

TABLE 4. Microbiological results of porridges cooked and stored at room temperature for 2, 3, 5, and 7 hours after cooking

Porridge[a]	Storage time (h)	Aerobic mesophile (CFU/g)[b]	*Escherichia coli* (MPN/g)[c]
No meat	2	2×10^3	< 3
	3	1.5×10	< 3
	5	1.7×10^2	< 3
	7	5×10^2	< 3
Low liver	2	1.2×10^3	< 3
	3	< 10	< 3
	5	7.5×10	< 3
	7	2.5×10^2	< 3
Low thigh	2	2.5×10^2	< 3
	3	8×10	< 3
	5	1.7×10^2	< 3
	7	2.2×10^3	< 3

CFU, colony-forming unit; MPN, most probable number

a. The no-meat porridge contained iron-fortified wheat flour, whole milk powder, brown sugar, and vanilla powder. The liver and thigh porridges contained the same ingredients plus lyophilized meat powder, as follows: low liver (1.0 g liver powder/100 g cooked porridge) and low thigh (3.0 g chicken thigh powder/100 g cooked porridge).

b. The assay cannot detect values < 10 CFU/g. These results were compared against standards from the International Commission on Microbiological Specifications for Food [39]: $m = 10^4$ CFU/g and $M = 10^6$ CFU/g.

c. The assay cannot detect values < 3 MPN/g. These results were compared against standards from the International Commission on Microbiological Specifications for Food [39]: $m = < 3$ MPN/g and M = 10 MPN/g.

Recipe-creation exercises

A key insight from the recipe-creation exercises was that mothers did not mix savory with sweet foods, which is what was done to create the meat-containing porridges that were evaluated in this study. However the alternative, creating an infant food mixing iron-fortified pasta with meat in a soup, was not nutritionally acceptable to the researchers because of the low energy density. Therefore, mothers' preference for non-meat-containing porridges during the acceptability trial was not surprising. That this preference did not extend to infants is promising; these meat-containing sweet porridges could be promoted as a nutritious food that infants like.

Nutrient contribution of porridges

Because of the changes in the organoleptic properties of the base porridge caused by adding the chicken liver and thigh powders and because of the vitamin A UL for infants, smaller amounts of the meat powders were added to the porridge than originally planned. Subsequent analysis of the energy, macronutrient, and vitamin A contents of the meat powders by Covance Laboratories resulted in vitamin A values that were considerably lower for chicken liver powder than we had estimated (6,690 vs. 31,626 µg RE/100 g), within 8% of the energy, protein, and fat values that we had estimated for chicken liver and chicken thigh powders, and within 8% of the vitamin A value for chicken thigh powder. The vitamin A discrepancy was due to the value for raw chicken liver in the IIN food-composition table [29], which is about twofold larger (6,165 vs. 3,296 µg RE) than in the USDA nutrient database [40]. Whether the vitamin A UL level of 600 µg RE is accurate or too low for this age group is debatable [41].

The iron contribution of the porridges is small relative to international recommendations (1.3–2.0 mg/27 g dry product vs. 7.4 mg/27 g dry product recommended [42]). However, previous work in Peru suggests that these porridges could provide an important bioavailable source of iron to infants, and the addition of lyophilized meat powder would increase bioavailability.

In a poor community in Lima, Peru, 6-month-old infants were served approximately 200-g portions of porridge containing wheat flour, milk, sugar, and oil twice a day for 9 months (Nelly Zavaleta, IIN, personal communication). These infants were fed a porridge similar to the "no-meat" porridge that we evaluated, with one exception: the earlier study was conducted when the wheat flour in Peru was fortified with 3.0 mg of iron per 100 g as ferrous sulfate [43], and we used wheat flour fortified with 5.0 mg of iron per 100 g as ferrous fumarate (in addition to thiamin, riboflavin, niacin, and folic acid), as allowed by new Peruvian legislation [28]. No baseline hemoglobin data were available on the Peruvian infants who consumed a porridge comparable to the "no-meat" porridge for 9 months; however, another study in a similar Peruvian community revealed that 72.7% of infants 6 months old had a hematocrit < 33%, indicative of anemia (Mary Penny, IIN, personal communication). At the end of the 9-month study, the prevalence of anemia was about 25%, which is about one-third of what could have been expected at baseline and one-third of that in a national sample of children 12 to 15 months of age [44].

As described earlier, daily consumption of ferrous sulfate–fortified wheat products (3 mg of iron per 100 g of wheat product) for 7 weeks eliminated anemia among 41 stunted and moderately anemic children 3 or 4 years of age [6].

Maternal acceptability trial

Mothers preferred porridges with low or no meat to porridges with a higher meat content. Nevertheless, scores for the organoleptic properties of the porridges averaged 3.4 to 4.4 (ranging from 3, "neither like nor dislike," to 5, "like very much"), suggesting that all porridges were within an acceptable range for women. When the proportion of women giving porridges the lowest scores (1, "dislike very much," and 2, "dislike some") was compared among the porridges, the same trend was seen as for other analyses: the proportion disliking the porridges was highest for the porridges with most meat, followed by those with less meat and no meat (data not shown).

Infant acceptability trial and videotaping

Despite differing maternal perceptions of the foods, infants consumed the same amount of the three porridges offered to them (no meat, low liver, low thigh), suggesting that they had no preference for one over the other, even after factors that influence infants' intake of any food had been controlled for. In contrast, the mothers expressed a preference for the no-meat porridge over the meat-containing porridges. This difference is supported by the lack of association between the overall score women gave to the porridges and the amount of porridge the infants ate (data not shown). Because an infant's intake is influenced by so many factors beyond his or her preference for a particular food, we videotaped a subset of children to try to objectively determine their porridge preference. The videotape analysis supports the findings from the intake data: there was no difference in infants' initial (2 minutes) preference for the porridges based on blinded coding. Correspondence between infants' total intake and 2-minute video coding has been observed in another study [30].

Comparison with acceptability of porridges used in public health programs

Several porridges used in public health programs in different countries have undergone acceptability trials [45–50]. The methods used in Mexico for the national PROGRESA program are most comparable with those of our study [49, 50]. In Mexico, the acceptability of three PROGRESA porridges (vanilla, banana, and chocolate flavored) to children was evaluated with hedonic scores (how trained enumerators judged the child's acceptance) and total intake over 14 days (grams consumed); acceptability of the porridges by the caregiver was not assessed. Acceptability and consumption were evaluated with 108 children (75% were < 2 years old, and 25% were moderately underweight and 2 or 3 years old). For acceptability, the enumerators scored children's acceptance of each porridge on a scale of 1 to 5. The average score for the three PROGRESA porridges was 3.7 to 4.2, and the differences between porridges were not statistically significantly different. These results are comparable to the unadjusted overall scores (on a five-point scale) mothers gave the porridges tested in Peru: 4.4 ± 1.0 (SD) for the no-meat porridge, 3.9 ± 1.1 for the low-liver porridge, 3.6 ± 1.2 for the high-liver porridge, 3.6 ± 1.2 for the low-thigh porridge, and 3.4 ± 1.3 for the high-thigh porridge.

Consumption in Mexico was measured over 14 days when the children's favorite porridge (based on acceptability) was provided daily at a feeding center. On each day, 69 g of the porridge was offered to the child; the average daily intake of the porridges was 57.6 to 65.2 g, which represents 83.4% to 94.3% of the porridge served. Children ate more of the chocolate porridge than the vanilla porridge, and more of the vanilla porridge than the banana flavor ($p < .05$). Among breastfed children < 2 years of age, the mean consumption of any porridge was 56.5 ± 15.0 g (82% of the amount served). The amount of porridge consumed by breastfeeding children < 2 years of age in Mexico is comparable to the amount consumed by Peruvian infants 6 to 9 months of age; however the Peruvian children were offered a larger amount (up to 200 g) than the Mexican children (69 g). It is not surprising that more than 83% of the porridge fed to the Mexican children was consumed; these children in the second year of life probably could have consumed more than the 69 g offered to them in one sitting.

The hedonic scale used to evaluate children's acceptance in Mexico was similar to the videotape analysis we completed. In both cases, coders observed children during a feeding and scored acceptability according to predefined categories, with 1 representing the lowest acceptability and 5 representing the highest acceptability. However, in our case, the observation was recorded and scoring was limited to the first 2 minutes of the feeding, whereas in Mexico, the observation was made in real time and covered the entire feeding episode.

Microbiological and pesticide residue testing

These data show that it is possible to develop meat-containing porridges that are pathogen-free for up to 7 hours after cooking and storage at room temperature. In addition, the lyophilized meat powder alone was microbiologically safe for 60 days when refrigerated at 4°–8°C. However, upon packaging, the dry ingredients had marginally acceptable levels of mold and yeast, and these values increased after 3 weeks of room-temperature storage. Allthough these results are not surprising, given the humid conditions in Lima, they suggest that the porridge ingredients need to be stored in conditions that prevent or decrease mold and yeast growth *prior* to mixing and packaging, that refrigeration of the packaged dry ingredients may be warranted to slow mold and yeast growth, and that other packaging material needs to be explored. With respect to pesticide residues, neither organophosphate nor organochlorinated compounds were detected in the meat powders after approximately 1 year of cold storage.

The microbiological safety of a processed complementary food used in Ghana (Weanimix) was evaluated under two conditions: fermentation and storage in a vacuum flask [51]. The main outcome was the presence of coliforms (≥ 100/mL), an indicator of fecal contamination, in the cooked maize–peanut-soybean porridge approximately 9 hours after preparation. A strength of this study is that foods were prepared and stored in homes under real-life conditions by women with children 6 to 18 months of age (n = 50). Both experimental conditions reduced coliform contamination of the food (fermentation and storage in vacuum flask if the food temperature was > 50°C). In the public health setting, however, neither condition is presumably used by families. In fact, the unfermented, non-vacuum-stored food had the highest proportion of contaminated samples (48%).

Simplified methods to assess food acceptability among young children

Rapid and accurate methods to assess food acceptability could promote their more regular use in public health programs and research. The results of this study indicate that in lieu of measuring infants' intake as a proxy for acceptability, other less resource-intensive methods could be used. For example, we found that mothers' and field workers' assessments of how much infants liked the porridge was strongly correlated with the infants' porridge intake. In a subset of infants with real-time video analysis of the first 2 minutes of each feeding, the coders' scores yielded similar conclusions as the infant intake data, but the scores were not associated with infant intake. These data suggest that a fieldworker needs to observe only the first 2 minutes of a feed and not the feeding session in its entirety. Notably, the mean length of feeding sessions for the 67 infant-days videotaped was 13.7 ± 6.1 minutes. Alternatively or in addition, maternal assessment of how the infant liked the porridge can be used. In summary, these "subjective" measures do not require training of mothers, use of food scales, or extensive data collection in the field and may be sufficient to determine infant acceptability of a new or modified food.

Conclusions

With maternal input, we developed infant porridges containing fortified wheat flour, fortified powdered milk, brown sugar, vanilla powder, and lyophilized meat powder; the porridges were tested for maternal and infant acceptability, microbiological, safety and pesticide residues. According to hedonic rating of sensory characteristics, mothers preferred the porridge with no meat to any of the porridges with meat. Even so, their overall ratings of the meat-containing porridges ranged from "neither like nor dislike" to "like." In comparison, infants consumed the same amount of the meat-containing and non-meat-containing porridges. The acceptability data suggest that if mothers can be convinced to feed the meat-containing porridges to the infants despite their own preferences, the infants will consume them. Pesticide residues were not detected in the meat powders. The microbiological quality of the lyophilized meat powder, dry ingredients, and cooked porridge was acceptable, except for the numbers of molds and yeasts in the dry ingredients. Measures need to be taken to reduce the mold and yeast content of the porridge ingredients.

Acknowledgments

We acknowledge the contributions to the fieldwork of Katiushka Vaccari Silva, Liliana Cutipa Quispe, Liz Espinoza Oré, Sara Livia Echía, Carolina Yong Ruiz, Roberto Carlos Renjifo Silva, and students from the Universidad Nacional Mayor de San Marcos and the Cordon Bleu Institute. We thank the community and Ministry of Health leaders for facilitating our entry into the community, the staff of the IIN laboratories for conduct of the microbiological analyses, and Dr. Kathryn Boor of Cornell University for interpretation of the microbiological results. We also thank Ruben Frescas, Jr., Akua Gyamerah, and Julia Raymond for development of the video coding system and for coding the videos, and Drs. Patricia Kariger and Richard Canfield for assistance with the coding system. Funding was provided by Cornell University (Division of Nutritional Sciences, Latin American Studies Program/Tinker Foundation, Program in International Nutrition, Mario Einaudi Center for International Studies, Marion Fish Cox Fellowship); Gerber Foundation Predoctoral Fellowship; American Society for Nutritional Sciences; Hispanic Scholarship Fund/Pfizer Inc. Fellowship; National Institutes of Health Training Grant in Nutrition (5 T32 K07158-28); and Scientific Cooperation Research Program, USDA (58-3148-4-071). Fortified wheat flour was donated by Alicorp SA, Peru, and vanilla powder by Montana SA, Lima, Peru.

References

1. Stoltzfus RJ, Dreyfuss ML. Guidelines for the use of iron supplements to prevent and treat iron deficiency anemia. Washington, DC: International Life Sciences Institute, 1998:1. Available at: http://inacg.ilsi.org/file/b2_VUHUQ8AK.pdf. Accessed 22 December 2006.
2. Grantham-McGregor S, Ani C. A review of studies on the effect of iron deficiency on cognitive development in children. J Nutr 2001;131(2S-2):649S–68S.
3. Rivera JA, Hotz C, Gonzalez-Cossio T, Neufeld L, Garcia-Guerra A. The effect of micronutrient deficiencies on child growth: A review of results from community-based supplementation trials. J Nutr 2003;133(11 suppl 2):4010S–20S.
4. Lynch SR, Stoltzfus RJ. Iron and ascorbic acid: Proposed fortification levels and recommended iron compounds. J Nutr 2003;133:2978S–84S.
5. Lutter CK. Macrolevel approaches to improve the availability of complementary foods. Food Nutr Bull 2003; 24:83–103.
6. López de Romaña D, Salazar M, Hambidge KM, Penny ME, Peerson JM, Krebs NF, Brown KH. Longitudinal measurements of zinc absorption in Peruvian children consuming wheat products fortified with iron only or iron and 1 of 2 amounts of zinc. Am J Clin Nutr 2005; 81:637–47.
7. Dewey KG, Brown KH. Update on technical issues concerning complementary feeding of young children in developing countries and implications for intervention programs. Food Nutr Bull 2003;24:83–103.
8. Allen LH. Folate and vitamin B_{12} status in the Americas. Nutr Rev 2004;62(6 pt 2):S29–33.
9. Murphy SP, Allen LH. Nutritional importance of animal source foods. J Nutr 2003;133(11 suppl 2):3932S–5S.
10. Delgado CL. Rising consumption of meat and milk in developing countries has created a new food revolution. J Nutr 2003;133(11 suppl 2):3907S–10S.
11. Cowin I, Emond A, Emmett P; ALSPAC Study Group. Association between composition of the diet and haemoglobin and ferritin levels in 18-month-old children. Eur J Clin Nutr 2001;55:278–86.
12. Faber M. Complementary foods consumed by 6–12-month-old rural infants in South Africa are inadequate in micronutrients. Public Health Nutr 2005;8:373–81.
13. Fox MK, Pac S, Devaney B, Jankowski L. Feeding infants and toddlers study: What foods are infants and toddlers eating? J Am Diet Assoc 2004;104:S22–30.
14. Fox MK, Reidy K, Karwe V, Ziegler P. Average portions of foods commonly eaten by infants and toddlers in the United States. J Am Diet Assoc 2006;106(1 suppl 1): S66–76.
15. Hotz C, Gibson RS. Participatory nutrition education and adoption of new feeding practices are associated with improved adequacy of complementary diets among rural Malawian children: A pilot study. Eur J Clin Nutr 2005;59:226–37.
16. McConahy KL, Smiciklas-Wright H, Birch LL, Mitchell DC, Picciano MF. Food portions are positively related to energy intake and body weight in early childhood. J Pediatr 2002;140:340–7.
17. Ntab B, Simondon KB, Milet J, Cissé B, Sokhna C, Boulanger D, Simondon F. A young child feeding index is not associated with either height-for-age or height velocity in rural Senegalese children. J Nutr 2005;135:457–64.
18. Yeung GS, Zlotkin SH. Efficacy of meat and iron-fortified commercial cereal to prevent iron depletion in cow milk-fed infants 6 to 12 months of age: A randomized controlled trial. Can J Public Health 2000;91:263–7.
19. Creed Kanashiro H, Fukumoto M, Bentley ME, Jacoby E, Verzosa C, Brown KH. Use of recipe trials and anthropological techniques for the development of a home-prepared weaning food in the Central Highlands of Peru. J Nutr Educ 1991;23:30–5.
20. Graham MA. Food allocation in rural Peruvian households: Concepts and behavior regarding children. Soc Sci Med 1997;44:1697–709.
21. Serra-Majem LL, Calvo JR, Male ML, Ribas L, Lainez P. Population attitudes towards changing dietary habits and reliance on general practitioners in Spain. Eur J Clin Nutr 1999;53(suppl 2):S58–61.
22. Aubel J, Touré I, Diagne M. Senegalese grandmothers promote improved maternal and child nutrition practices: The guardians of tradition are not averse to change. Soc Sci Med 2004;59:945–59.
23. Gittelsohn J, Vastine AE. Sociocultural and household factors impacting on the selection, allocation and consumption of animal source foods: Current knowledge and application. J Nutr 2003;133(11 suppl 2): 4036S–41S.
24. Liria MR, Bartolini R, Abad MF, Creed-Kanashiro HM. Mothers' perceptions towards the feeding of animal source foods to infants, Lima, Peru. FASEB J 2005;19(4): A460 (abstract 285.6).
25. Desor JA, Maller O, Greene LS. Preference for sweet in humans: Infants, children, and adults. In: Weiffenbach JM, ed. Taste and development: The genesis of sweet

preference. DHEW Publication No. (NIH) 77-1068. Bethesda, MD, USA: US Department of Health, Education, and Welfare, Public Health Service, 1977:161–72.
26. Hallberg L, Hoppe M, Andersson M, Hulthén L. The role of meat to improve the critical iron balance during weaning. Pediatrics 2003;111(4 pt 1):864–70.
27. Gailani MB, Fung DY. Critical review of water activities and microbiology of drying of meats. Crit Rev Food Sci Nutr 1986;25:159–83.
28. El Peruano. Normas legales, 3 febrero 2005. Aprueban Reglamento de la Ley N 28314 que dispuso la fortificación de la harina de trigo con micronutrientes. Lima, Peru: El Peruano, 2005:286087–286089.
29. Instituto de Investigación Nutricional (IIN). Food composition tables IIN, 2001. Lima, Peru: Instituto de Investigación Nutricional, 2001.
30. Mennella JA, Jagnow CP, Beauchamp GK. Prenatal and postnatal flavor learning by human infants. Pediatrics 2001;107(6):e88. Available at: http://www.pediatrics.org/cgi/content/full/107/6/e88. Accessed 10 December 2006.
31. Sullivan SA, Birch LL. Infant dietary experience and acceptance of solid foods. Pediatrics 1994;93:271–7.
32. Food and Drug Administration. Bacteriological analytical manual, 7th ed. Arlington, VA, USA: AOAC International, 1992:17–234.
33. Codex Alimentarius. Guidelines on formulated supplementary foods for older infants and young children, CAC/GL 08-1991. Available at: http://www.codexalimentarius.net/web/standard_list.do?lang = en. Accessed 10 December 2006.
34. Instituto Nacional de Defensa de la Competencia y de la Protección de la Propiedad Intelectual (INDECOPI). Norma técnica Peruana, Alimentos cocidos de reconstitución instantánea, Papillas, Requisitos. NTP 209.260:2004. Lima, Peru: INDECOPI, 2004:15.
35. Institute of Medicine. Dietary reference intakes for vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel, silicon, vanadium and zinc. Washington, DC: National Academies Press, 2001:144.
36. Pan American Health Organization/World Health Organization (PAHO/WHO). Guiding principles for complementary feeding of the breastfed child. Washington, DC: PAHO, 2003:21. Available at: http://www.paho.org/English/AD/FCH/NU/Guiding_Principles_CF.htm. Accessed 22 December 2006.
37. Huss HH, Ababouch L, Gram L. Assessment and management of seafood safety and quality. FAO Fisheries Technical Paper No. 444. Rome: Food and Agriculture Organization, 2004:8. Available at: http://www.fao.org/DOCREP/006/Y4743E/Y4743E00.htm.. Accessed 22 December 2006.
38. Institute of Medicine. Scientific criteria to ensure safe food. Washington, DC: National Academies Press, 2003:142. Available at: http://www.nap.edu/books/030908928X/html. Accessed 22 December 2006.
39. International Commission on Microbiological Specifications for Foods (ICMSF). Microorganismos de los alimentos. Métodos de muestreo para análisis microbiólogicos: Principios y aplicaciones específicas, volumen II. Zaragoza, Spain: Editorial Acribia, 1981:110–3.
40. US Department of Agriculture (USDA). National nutrient database for standard reference, release 18. Available at: http://www.ars.usda.gov/main/site_main.htm?modecode=12354500. Accessed 10 December 2006.
41. Zlotkin S. A critical assessment of the upper intake levels for infants and children. J Nutr 2006;136:502S–6S.
42. Lutter CK, Dewey KG. Proposed nutrient composition for fortified complementary foods. J Nutr 2003; 133:3011S–20S.
43. Hertrampf E. Iron fortification in the Americas. Nutr Rev 2002;60(7 pt 2):S22-5.
44. Instituto Nacional de Estadística e Informática (INEI). Encuesta demográfica y de salud familiar 2000. Lima, Peru: INEI, 2001:186.
45. Tartanac F. Incaparina and other Incaparina-based foods: Experience of INCAP in Central America. Food Nutr Bull 2000;21:49–54.
46. Barenbaum M, Pachón H, Schroeder DG, Hurtado E. Use, acceptability, and cost of Incaparina, a commercially processed food in Guatemala. Food Nutr Bull 2001; 22:71–80.
47. Rozo C. Complementary foods in Colombia. Food Nutr Bull 2000;21:55–60.
48. Lartey A, Manu A, Brown KH, Peerson JM, Dewey KG. A randomized, community-based trial of the effects of improved, centrally processed complementary foods on growth and micronutrient status of Ghanaian infants from 6 to 12 mo of age. Am J Clin Nutr 1999;70:391–404.
49. Rosado JL, Rivera J, Lopez G, Solano L, Rodriguez G, Casanueva E, Garcia-Aranda A, Toussaint G, Maulen I. Desarrollo y evaluación de suplementos alimenticios para el Programa de Educación, Salud y Alimentación. Salud Publica Mex 1999;41:153–62. Available at: http://www.scielosp.org/pdf/spm/v41n3/41n3a02.pdf. Accessed 22 December 2006.
50. Rosado JL, Rivera J, Lopez G, Solano L. Development, production and quality control of nutritional supplements for a national supplementation programme in Mexico. Food Nutr Bull 2000;21:30–4.
51. Kimmons JE, Brown KH, Lartey A, Collison E, Mensah PP, Dewey KG. The effects of fermentation and/or vacuum flask storage on the presence of coliforms in complementary foods prepared for Ghanaian children. Int J Food Sci Nutr 1999;50:195–201.

Effect of iodine status and other nutritional factors on psychomotor and cognitive performance of Filipino schoolchildren

Ma. Sofia V. Amarra, Demetria C. Bongga, Leticia Peñano-Ho, Federico B. Cruz, José S. Solis, and Erniel B. Barrios

Abstract

Background. *Until 1998, iodine deficiency was a public health problem in the Philippines. A law entitled "An Act Promoting Salt Iodization Nationwide" (ASIN) has been passed and implemented by the government to eliminate iodine deficiency. The contribution of salt iodization, as well as dietary, health, and environmental factors, to improving the intellectual performance of Filipino schoolchildren remains to be determined.*

Objective. *The objectives of the study were to determine the relationship between iodine status and levels of psychomotor and cognitive performance in first-grade children aged 6 to 10 years, and to examine the extent to which dietary, biochemical, health, and environmental factors contribute to children's mental performance.*

Methods. *Two hundred ninety children in six classroom sections from a public school in Manila were examined by measurement of urinary iodine excretion (UIE) and thyroid palpation. The median UIE level for each section was determined. Sixty-five children classified as iodine deficient (UIE < 90 μg/L with grade 1 goiter,* n *= 34) and non-iodine deficient (UIE > 100 μg/L without goiter,* n *= 31) were given psychomotor and cognitive function tests (Bender–Gestalt and Raven's Colored Progressive Matrices). Scores from the two tests were used to determine each child's general ability percentile rank. Other variables examined were dietary intake (% RDA of nutrients ingested based on two nonconsecutive 24-hour recalls); deficiencies in iron, vitamin A, and selenium; parasitic infection; coliform contamination of drinking water; household use of iodized salt; illness in the past 2 weeks; and wasting and stunting.*

Results. *Children whose general ability scores were at or above the 50th percentile had higher UIE levels, but the relationship was not significant. Children from sections with higher median UIE levels had higher percentile ranks for general ability (*p *= .002). Backward logistic regression showed that the variance in deficient and adequate mental performance was explained by dietary intakes that met ≥ 80% of the RDA for energy, protein, thiamin, and riboflavin; the use of iodized salt; child's iodine status; and stunting (*R^2 *= .520,* p *= .0016). Higher class median UIE was associated with better psychomotor and cognitive performance in children who were tested. Factors that contributed to better performance include higher intakes of energy, protein, thiamin, and riboflavin; household use of iodized salt; normal iodine status; and absence of stunting or chronic malnutrition.*

Conclusions. *Salt iodization, accompanied by adequate intakes of energy, protein, and foods rich in thiamin and riboflavin, can contribute to improved mental performance in Filipino schoolchildren. Longer-term factors that can contribute to improved performance are achievement of normal iodine status and elimination of protein–energy malnutrition.*

Key words: Cognitive function, iodine deficiency, nutrient intake, psychomotor function, salt iodization, schoolchildren

Background

Iodine is an essential trace element required for the synthesis of thyroid hormones. These hormones are involved in normal growth and development, particularly in the development of the central nervous system. Thyroid hormone action is exerted through the binding of triiodothyronine to nuclear receptors, which regulate

Ma. Sofia V. Amarra is affiliated with the Food and Nutrition Research Institute, Department of Science and Technology, Metro Manila, Philippines; Demetria C. Bongga, Leticia Peñano-Ho, José S. Solis, and Erniel B. Barrios are affiliated with the University of the Philippines, Diliman, Quezon City. Federico B. Cruz is affiliated with the University of the Philippines, Diliman, Quezon City, and Manila.

Paper presented at the Parallel Symposium on Nutrition and Development, 18th International Congress of Nutrition of the IUNS, Durban, South Africa, 19–23 September 2005.

Please direct queries to the corresponding author: Ma. Sofia V. Amarra, 2C Freedom Lane, Interville 2, Culiat, Quezon City 1128, Philippines; e-mail: sofiaamarra@yahoo.com.

the expression of specific genes in different brain regions following a precise developmental schedule. Mental retardation and endemic cretinism result from insufficient supply of thyroid hormones to the developing brain. Mild deficiency leads to a global loss of 10 to 15 intelligence quotient (IQ) points at the population level. The ensuing brain damage and loss of intellectual potential due to iodine deficiency can hinder the socioeconomic development of populations [1].

In 1995, a law entitled "An Act Promoting Salt Iodization Nationwide" (ASIN) was passed and implemented by the Philippine Government for the elimination of iodine deficiency. The law mandates universal salt iodization and legislates guidelines for iodization technology, quality assurance, marketing, and advocacy and provides sanctions against those in industry who do not comply.

Until 1998, iodine deficiency was a persistent public health problem in the Philippines. Surveys in 1987 and 1993 among schoolchildren aged 6 to 12 years found goiter prevalence rates of 12.8% and 12.6%, respectively [2]. This is way above the 5% cut-off established by the World Health Organization (WHO). The country's 5th National Nutrition Survey [3] showed a goiter rate of 6.7% in the same age group and a median urinary iodine excretion (UIE) level of 71 µg/L, with 35.8% of values below 50 µg/L. The acceptable median UIE level for a population is above 100 µg/L, with not more than 20% of values falling below 50 µg/L [4]. It is only recently that improvements in the iodine status of Filipino children have been seen. The 6th National Nutrition Survey (2003) showed that the median UIE of children aged 6 to 12 years was 201 µg/L, with 11.4% of values below 50 µg/L [5]. These findings indicate that iodine deficiency has probably ceased to be a major health problem in the country.

Several factors influence the supply of thyroid hormones to the brain. Poor dietary quality (such as the lack of animal-source foods), the use of noniodized salt, and microbial contamination of drinking water limit absorption of iodine and render the mineral unavailable for use by the body [6, 7]. Host characteristics such as parasitic infestation, deficiencies in iron, vitamin A, and selenium, and protein–energy malnutrition adversely affect iodine uptake and utilization [7]. The presence of these factors may cause iodine deficiency to persist, with consequent effects on children's intellectual development. The objectives of this study were to examine the relationship between iodine status and psychomotor and cognitive performance in first-grade children aged 6 to 10 years, and determine the extent to which dietary intake, micronutrient status, and health and environmental factors contribute to children's mental performance.

Methods

Sample selection and sample-size estimation

Ethical approval for this study was granted by the Review Committee of the Office of the Vice-Chancellor for Research and Development, University of the Philippines, Diliman.

First-grade children aged 6 to 10 years (mean age, 6.8 ± 1.1 years) studying in a public elementary school located in one of the most depressed areas in Manila were examined. The student population was composed mainly of poor children whose families lived in shanty areas surrounding the school.

A two-stage sampling procedure was used to select the study sample. The first stage was the initial screening of 290 children from three classroom shifts based on UIE and thyroid palpation. Spot casual urine samples were collected during class hours, stored in ice, and transported to the Biological Research Laboratory of the Department of Health for determination of urinary iodine. Goiter was determined by thyroid palpation.

The second stage of the procedure was the selection of a purposive sample of 40 children classified as having adequate iodine status (> 100 µg/L UIE and grade 0 goiter) and 40 children with iodine deficiency (< 90 µg/L UIE and grade 1 goiter).

Consent was obtained from school authorities and parents prior to the implementation of the study. Authorization to conduct the study was first obtained from the Division of City Schools under the Department of Education. Letters addressed to parents were then given to all students belonging to the selected class sections. The objectives and methods of the study were explained in these letters using local language. Parents or guardians were asked to indicate their consent by signing an attached form. Children whose parents gave written consent for participation were included in the sampling frame.

As a result of attrition during the study period, only 38 children with adequate iodine status and 39 children with iodine deficiency participated for the entire duration of the study.

Determination of goiter and UIE level

Goiter was determined based on the following simplified criteria [4]: grade 0, no palpable or visible goiter; grade 1, goiter palpable but not visible when neck is in the normal position; grade 2, goiter visible when neck is in the normal position. Urinary iodine was determined using the method prescribed by WHO/UNICEF/ICCIDD [4]. Urine was digested with chloric acid under mild conditions, and the amount of iodine was determined by its catalytic role in the reduction of ceric ammonium sulfate in the presence of arsenious acid. Duplicate analyses were performed.

Measurement of dietary intake, microbial contamination of drinking water, and salt iodine content

Two nonconsecutive 24-hour food recalls were obtained by interviewing the children and their caretakers, who were usually their mothers. Snacks eaten by the subjects in school were observed and recorded for 2 days. The mean intakes of energy and nutrients were computed from Philippine Food Composition Tables [8]. Samples of drinking water were taken from households, placed in ice, and tested for coliform contamination by the most probable number (MPN) test. Household salt was tested for the presence of iodate with a rapid test kit.

Anthropometric measurements

The heights and weights of the children were measured with a Detecto physician scale. The measurements were taken by a single trained person to avoid interobserver errors. The subjects wore light clothing and no shoes during measurement. Height was measured to the nearest 0.1 cm and weight was measured to the nearest 0.1 kg. Children's birthdates were obtained from class records. Z-scores of height-for-age and weight-for-height were computed using the National Center for Health Statistics (NCHS) population as reference.

Measurement of iron, vitamin A, and selenium deficiencies

The presence of iron-deficiency anemia, vitamin A deficiency, and selenium deficiency was assessed by examining hemoglobin, serum retinol, and whole blood selenium, respectively. Five milliliters of blood was collected by venipuncture. Serum retinol was determined by high-pressure liquid chromatography [9]. Hemoglobin was determined by the cyanmethemoglobin method [10]. Whole blood selenium was examined by atomic absorption spectrophotometry [11]. Duplicate analyses were conducted for each test. The coefficient of variation between analyses fell within ± 5.0%. The cut-offs used were < 12 g/dL hemoglobin [12], < 0.7 μmol/L serum retinol, and < 60 μg/L whole blood selenium [13].

Intestinal parasites and morbidity

Parasitic infestation and morbidity in the past 2 weeks were determined. Fresh stool samples were collected, stored in ice, and immediately analyzed in the laboratory for the presence of *Ascaris, Trichuris,* and *Giardia lamblia*. Morbidity in the past 2 weeks was determined by interviewing the child's mother.

Psychomotor and cognitive measures

The children's psychomotor and cognitive functions were examined by Raven's Colored Progressive Matrices (CPM) and the Bender–Gestalt test. Two trained psychologists administered, analyzed, and interpreted each child's test results.

Raven's CPM is a test that measures cognitive function, for which multicultural application has been established [14]. The test consists of 36 colored matrix items or patterns constructed with a logical relationship. Each item contains a matrix with a missing piece. The task for the examinee is to determine which of several choices best fits the logical theme of the matrix and completes the pattern.

The Bender–Gestalt test measures psychomotor function [15]. The test is a complex visual-motor task, although the subject is instructed to copy relatively simple designs. A general ability percentile rank, reflecting age-specific intelligence and developmental level, was computed for each child based on his or her scores.

Data quality

In order to maintain data quality and reduce variability, individual trained personnel were assigned to collect specific data throughout the study period. A single trained examiner conducted all thyroid palpations during screening. All 24-hour recall interviews were administered by the same professional nutritionist–dietitian using a USDA-developed multiple pass approach [16]. Instruments used to facilitate respondents' recall memory were food photographs, measuring cups and spoons, rulers, and thickness cards to estimate the thickness of food slices. Samples of all food items bought in the local market and listed in the recalls were obtained and weighed. Recipes of mixed dishes cooked in the home were obtained, and their yields were estimated and used as the basis for computing the nutrient content of dishes.

Data analysis

Data were analyzed by SPSS for Windows version 7. The chi-square test (for categorical values) and Student's *t*-test (for continuous values) were used to test differences in subjects' characteristics and dietary intakes. The Mann–Whitney test was used to test differences in UIE between children with normal and low general ability scores. Differences in mean general ability scores among class sections were examined by Kruskal–Wallis analysis of variance. Backward stepwise logistic regression was used to examine the influence of dietary intake, micronutrient status, anthropometric status, contamination of drinking water, morbidity in the past 2 weeks, and household use of iodized salt on the child's psychomotor and cognitive function expressed as general ability percentile rank. Scores that were ranked in the 50th percentile or above were considered normal (coded as 0), and those below the 50th percentile were considered deficient or low (coded as 1). For the independent variables, deficient values were coded as 1; normal values were coded as 0. Likewise, the presence of health-promoting factors (i.e., household use of iodized salt, availability of clean drinking water, no illness in the past 2 weeks) was coded as 0 and their absence coded as 1.

TABLE 1. Education and occupations of parents of children with normal iodine status and with iodine deficiency

Variable	Normal iodine status—no. (%)	Iodine deficient—no. (%)
Parental education		
Father		
Below high school	17 (56.7)	22 (64.7)
High school and above	9 (30.0)	8 (23.5)
No schooling or deceased	4 (13.3)	4 (11.8)
Total	30 (100.0)	34 (100.0)
Mother		
Below high school	19 (63.3)	27 (79.4)
High school and above	11 (36.7)	7 (20.6)
Total	30 (100.0)	34 (100.0)
Parental occupation		
Father		
Unemployed	1 (3.3)	3 (8.8)
Tricycle driver	3 (10.0)	4 (11.8)
Construction worker	3 (10.0)	5 (14.7)
Driver (taxi/bus/jeepney)	6 (20.0)	4 (11.8)
Skilled laborer	5 (16.7)	3 (8.8)
Painter (house/car)	1 (3.3)	5 (14.7)
Vendor	2 (6.7)	4 (11.8)
Salaried worker (security guard, dicer[a])	2 (6.7)	1 (2.9)
Meat/fish dealer	2 (6.7)	0 (0)
Truck helper	1 (3.3)	1 (2.9)
Collects garbage for recycling	0 (0)	1 (2.9)
None or deceased	4 (13.3)	3 (8.8)
Total	30 (100.0)	34 (100.0)
Mother		
Housewife	18 (60.0)	20 (58.8)
Office worker	2 (6.7)	1 (2.9)
Factory worker	2 (6.7)	0 (0)
Vendor	2 (6.7)	6 (17.6)
Collects garbage for recycling	1 (3.3)	1 (2.9)
Manicurist or laundrywoman	1 (3.3)	3 (8.8)
Doormat maker	2 (6.7)	0 (0)
Stall owner	2 (6.7)	1 (2.9)
Housemaid or street sweeper	0 (0)	2 (5.9)
Total	30 (100.0)	34 (100.0)

a. Rolls dice in illegal gambling

Results

The socioeconomic profile of the subjects' families was fairly homogeneous. All subjects lived in a community that was classified as socioeconomic category D/E (lowest category) by the country's National Statistical Coordination Board. The fathers were engaged in various types of manual labor and low-paying jobs, while the mothers were usually housewives. Most parents had finished primary school (**table 1**).

Table 2 shows the characteristics of the subjects. There were no differences in age, sex distribution, or anthropometric or micronutrient status between children with normal iodine status and those with iodine deficiency. Deficiencies in iron, vitamin A, and selenium were high regardless of iodine status. Levels of stunting were high in both groups.

Problems were encountered in collecting fecal samples. Forty-eight percent of mothers refused to collect their children's feces, despite being given adequate supplies for ease of collection. Of the 41 samples that were submitted, almost half (49%) tested positive for parasites. The majority of children (70%) reported no acute illness in the past 2 weeks.

Most households (78%) used coliform-contaminated drinking water, which exceeded the cut-off point of 2.2 MPN per 100 mL, and 86% of households did not use iodized salt. There were no significant differences between iodine-deficient and normal iodine groups in morbidity, contamination of drinking water, and use of iodized salt.

Table 3 shows the mean nutrient adequacy of chil-

TABLE 3. Mean percentage of recommended dietary allowance (RDA) of nutrients consumed by children with normal iodine status and with iodine deficiency

Nutrient	Normal iodine status	Iodine deficiency	*p*
Energy	81.1	80.5	.918
Protein	109.1	95.9	.143
Iron	76.8	71.4	.538
Vitamin A	69.0	44.3	.066
Thiamin	58.2	47.8	.074
Riboflavin*	71.5	55.6	.012
Niacin	52.5	44.8	.108
Ascorbic acid	28.5	32.0	.681

*Difference significant at .05 level.

TABLE 2. Characteristics of children with normal iodine status and with iodine deficiency

Characteristic	Normal iodine status	Iodine deficiency	Total sample	*p*
Age—mean ± SD (yr)	6.8 ± 1.1	6.9 ± 1.1	6.8 ± 1.1	.644
Gender distribution—no.				
Males	18	23	41	.307
Females	20	16	36	
Anthropometric status—%				
Stunted (HAZ < −1 SD)	63.1	70.6	65.5	.287
Wasted (WHZ < −1 SD)	18.8	17.4	18.4	.797
Micronutrient status—%				
Iron deficient	37.5	25.8	31.7	.234
Vitamin A deficient	15.6	35.5	25.4	.064
Selenium deficient	68.7	51.6	60.3	.165
Morbidity and household variables—%				
Acute illness in past 2 wk	36.7	23.5	29.7	.251
Used contaminated drinking water	76.7	78.8	77.8	.840
Did not use iodized salt	80.0	91.2	85.9	.199

HAZ, height-for-age z-score; WHZ, weight-for-height z-score

dren's diets compared with local recommended dietary allowances (RDAs) [17]. Except for vitamin C, children with normal iodine status had slightly higher nutrient intakes. They also had significantly higher levels of riboflavin intake than those with deficiency. Further analysis showed that these children consumed larger amounts of animal protein (**table 4**).

There was no difference in mean UIE levels of children with normal and low general ability percentile ranks (**table 5**). The mean percentile ranks differed significantly among children from different class sections. Children in morning classes tended to have higher ranks than those in late afternoon classes (**table 6**). Comparison of the median UIE in each class with the general ability ranks of children belonging to the class showed that the median UIE increases as mean general ability percentile ranks increase (**fig. 1**).

Backward stepwise logistic regression was performed with general ability percentile rank as the dependent variable. Variables included in the analysis were child's age and gender, dietary intake (< or ≥ 80% of RDA), nutritional status (stunting, wasting), micronutrient status (iodine, iron, vitamin A, selenium), previous morbidity, coliform contamination of drinking water, and use of iodized salt. Fifty-two children whose data were complete were included in the analysis. Fac-

TABLE 4. Mean ± SD percentage of total calories contributed by different food groups in diets of children with normal iodine status and with iodine deficiency

Food group	Normal iodine status	Iodine deficiency	*p*
Cereals	68.9 ± 11.9	71.9 ± 15.3	.391
Sugars and syrups	5.4 ± 4.3	6.1 ± 6.6	.602
Fats and oils	1.31 ± 2.4	3.3 ± 5.5	.078
Animal protein*	12.6 ± 8.4	7.6 ± 6.4	.009
Dried beans/nuts/seeds	2.9 ± 4.3	3.3 ± 6.4	.841
Green leafy and yellow vegetables	0.2 ± 0.3	0.01 ± 0.2	.258
Sugared beverages	5.3 ± 3.9	4.7 ± 3.9	.733

*Difference significant at .05 level.

TABLE 5. Urinary iodine concentrations according to children's general ability percentile rank

General ability percentile rank	No. (%)	Mean ± SD urinary iodine (μg/L)[a]
Normal (≥ 50th %ile)	28 (43.1)	111.4 ± 49.9
Low (< 50th %ile)	37 (56.9)	99.2 ± 49.4

a. Mann-Whitney U value = 467.5; *p* = .503.

TABLE 6. General ability percentile rank according to children's class section

Class section	*N*	Mean general ability percentile rank[a]
A (6–10 a.m.)	5	41.4
B (6–10 a.m.)	8	43.7
C (10 a.m.–2 p.m.)	13	45.9
D (10 a.m.–2 p.m.)	15	31.8
E (2–6 p.m.)	15	20.3
F (2–6 p.m.)	9	23.17

a. Chi-square = 19.557; df = 5; *p* = .002.

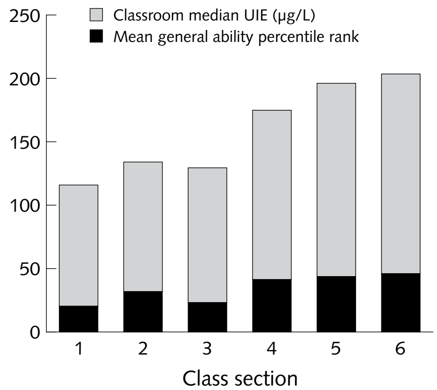

FIG. 1. Classroom median urinary iodine concentration (UIE) and mean general ability percentile ranks of students belonging to each section. Class sections: 1 = class E, 2 = class D, 3 = class F, 4 = class A, 5 = class B, 6 = class C

tors that significantly explained 52% of the variance (p = .0016) in general ability scores were adequacy of intake of energy, protein, thiamin, and riboflavin; household use of iodized salt; child's iodine status; and height-for-age (**table 7**). Deficient mental performance was more likely to be present in children whose dietary intakes failed to meet at least 80% of the requirements for energy, protein, thiamin, and riboflavin, whose households did not use iodized salt, or who were stunted or iodine-deficient.

Discussion

One limitation of the study was its small sample size, which was due mainly to resource constraints. Huda et al. [18] estimated that for cognitive function tests, a sample size of 170 is needed to show a significant difference of 0.5 SD between groups at the 5% level with 80% power, allowing a loss of 10%. In addition, other factors that affect child development, such as quality of parental care and psychosocial stimulation, were not examined. But in spite of these limitations, the study has shown that children with adequate psychomotor and cognitive performance had better-quality diets (in terms of energy, protein, thiamin, and riboflavin), had normal height-for-age and iodine status, and belonged to households that used iodized salt.

TABLE 7. Factors that influence schoolchildren's psychomotor and cognitive performance based on general ability percentile rank[a]

Variable	Regression coefficient ± SE	p
Dietary intake < or ≥ 80% of RDA		
Energy	−2.5143 ± 1.3934	.0712
Protein	3.0818 ± 1.4978	.0396
Thiamin	3.5957 ± 1.7542	.0404
Riboflavin	−2.4828 ± 1.4827	.0940
Household does not use iodized salt	−2.1965 ± 1.3659	.1078
Child has iodine deficiency	−1.6025 ± 0.8869	.0708
Child is stunted	−3.8010 ± 1.5303	.0130
Constant	1.0248 ± 0.7636	

RDA, recommended dietary allowance
a. R^2 of model = .520; p = .0016.

These findings are consistent with other studies showing that dietary adequacy, as measured by both quality and quantity, is important for the development of children's cognitive abilities [19, 20]. Studies in Kenya showed that nutrients, specifically animal-source iron and cobalamin, were highly associated with cognitive scores in school-aged Kenyan children. Diet quality (percentage of calories from animal sources) had direct effects on children's cognitive abilities, whereas energy intake affected abilities indirectly through children's levels of activity. The Kenyan studies showed that supplementation with animal-source protein had significant positive effects on cognitive performance, even if the effects were not equivalent across all domains of cognitive functioning. Children supplemented with meat outperformed nonsupplemented controls and milk-supplemented children on Raven's Progressive Matrices [19].

Low height-for-age or stunting had a significant effect on mental performance among children in the present study. Mendez and Adair [21] found that Filipino children who were stunted in the first 2 years of life had significantly lower cognitive test scores at 8 and 11 years than those who were never stunted. Among Kenyan children, stunting predicted performance in mathematics and in cognitive and motor function [19]. Children who experienced hunger did worse in school achievement tests, even if they were given a snack and were not hungry during the tests. The researchers suggested that shortage of food may be a serious obstacle to children's ability to achieve in school and that school feeding programs should be encouraged, especially where undernutrition is endemic. This is because cognition in undernourished children is more vulnerable to the detrimental effects of hunger than cognition in adequately nourished children [19].

Iodine status is a known predictor of mental performance. Normal iodine status contributed to better mental performance among children in this study. Inadequate thyroid hormone production due to iodine deficiency results in hypothyroidism, which is accompanied by lethargy, sleepiness, and apathy and is thought to be related to poor school performance in iodine-deficient areas. Huda et al. [22] found that Bangladeshi children with hypothyroidism secondary to iodine deficiency performed worse than euthyroid children on reading, spelling, and cognitive function tests.

The child's existing diet and environment are not the only factors that affect his or her iodine status and mental performance. Thyroid hormones transferred from the mother to the fetus are critical for normal brain development. It is now recognized that only a slight difference in the concentration of thyroid hormones during pregnancy can lead to significant changes in intelligence in children [23]. Prenatal exposure to environmental contaminants that interfere with the thyroid system has been linked with neurodevelopmental damage in humans. These so-called endocrine disrupters mimic the biological activity of a hormone and alter the structure or function(s) of the endocrine system, causing adverse health effects in an organism or its progeny [23, 24]. Chemicals shown to disrupt thyroid function include pesticides and chemicals widely used in cosmetics, perfumes, detergents, toys, and plastics. Scientists are now looking at the possible association between *in utero* exposure to these chemicals and the increasing number of children in developed countries who exhibit attention deficit disorder, autism, and associated neurodevelopmental and behavioral problems [23]. Given the unhygienic living conditions in the study area and its proximity to the city's dumpsite, maternal exposure to chemical contaminants during pregnancy is highly probable.

Although iodized salt may alleviate or prevent iodine deficiency, its use as the sole intervention may not be sufficient to address deficits in mental performance because of physiological interactions between iodine and other nutrients. Deficiencies in protein, iron, vitamin A, and selenium are known to impair thyroid metabolism. Iron-deficiency anemia reduced thyroid peroxidase activity in animals [25]. Thyroid peroxidases convert inorganic iodide into organic iodide to couple iodinated tyrosyl moieties in the synthesis of thyroid hormone. Selenium deficiency aggravates iodine deficiency by reducing the activity of selenoenzymes involved in iodine metabolism [26]. Selenium deficiency in rats decreased the conversion of thyroid hormone (thyroxine or T_4) to its metabolically active form (triiodothyronine or T_3) [27]. Deficiency of vitamin A inhibits the essential dimerization of thyroid hormone with retinols, a process essential for thyroid hormone expression [28]. Protein deficiency in rats impaired thyroidal transport of iodine, decreased its concentration in the thyroid, and enhanced the goitrogenic effect of antithyroid agents [29]. Thus, these nutrients must also be provided in order to maximize the benefits of iodine supplementation.

The present study has shown that children with iodine deficiency consumed significantly smaller amounts of riboflavin and animal protein. Regardless of iodine status, a high prevalence of protein–energy malnutrition as well as iron, vitamin A, and selenium deficiency existed in the study sample. These results suggest that salt iodization should be accompanied by dietary supplementation of animal protein and energy-dense foods fortified with multiple micronutrients. Feeding programs providing such foods would most likely benefit not only the mental abilities of these children but also their physical well-being.

Conclusions

Children with significantly higher levels of psychomotor and cognitive performance belonged to classroom sections with higher median UIE levels. Factors that predisposed to better mental performance in this group were adequate intakes of energy, protein, thiamin, and riboflavin; household use of iodized salt; adequate iodine status; and normal height-for-age. School and community feeding programs that aim to eradicate protein–energy malnutrition and provide poor children with high-quality protein (i.e., meat and milk that are rich in thiamin and riboflavin, respectively) and fortified foods are likely to have significant effects on students' mental functions. The government should therefore encourage such programs while vigorously pursuing its universal salt iodization campaign in order to improve Filipino children's academic capabilities.

Acknowledgments

The senior author wishes to acknowledge the International Nutrition Foundation and Unilever for providing the INF-Unilever Fellowship Grant that enabled the presentation of this study at the 18th International Congress of Nutrition of the IUNS held in Durban, South Africa on September 19–23, 2005. Financial support for the research was provided by the Office of the Vice-Chancellor for Research and Development and the College of Home Economics Foundation, University of the Philippines, Diliman; and the National Research Council of the Philippines.

References

1. Delange F. Iodine deficiency as a cause of brain damage. Postgrad Med J 2001;77:217–20.
2. Philippine National Micronutrient Operations Plan. 1996–1998. Philippine Plan of Action for Nutrition. Metro Manila: Department of Health, 1999.
3. Food and Nutrition Research Institute. Philippine Nutrition Facts and Figures. Bicutan, Taguig: Department of Science and Technology, 2001

4. World Health Organization/UNICEF/International Council for Control of Iodine Deficiency Disorders. Indicators for assessing iodine deficiency and their control through salt iodization. Geneva: WHO/UNICEF/ICCIDD, 1994.
5. Food and Nutrition Research Institute. 6th National Nutrition Survey results. Available at: http://www.fnri.dost.gov.ph/htm/nutstat.htm. Accessed 10 December 2006.
6. Hetzel BS. The concept of iodine deficiency disorders (IDD) and their eradication. In: Dunn JT, Pretell EA, Daza CH, Viteri FE, eds. Towards the eradication of endemic goiter, cretinism and iodine deficiency. Washington, DC: Pan American Health Organization/World Health Organization, 1986.
7. World Health Organization. Trace elements in human nutrition and health. Geneva: WHO, 1996.
8. Food and Nutrition Research Institute. Philippine Food Composition Tables. Bicutan, Taguig: Department of Science and Technology, 1997.
9. Gunter EW, Miller DT. Laboratory procedures used by the Division of Environmental Health Laboratory Sciences, Center for Environmental Health, Centers for Disease Control for the Hispanic Health and Nutrition Examination Survey (HHANES, 1986). Atlanta, GA, USA: U.S. Department of Health and Human Services, 1986.
10. International Nutritional Anemia Consultative Group (INACG). Measurements of iron status—A report of the International Nutritional Anemia Consultative Group. Washington, DC: INACG Secretariat, 1985.
11. Fidanza F, ed. Nutritional status assessment: A manual for population studies. London: Chapman and Hall, 1991.
12. World Health Organization. Nutritional anemias. Technical Report Series No. 503. Geneva: WHO, 1972.
13. Neve J. New approaches to assess selenium status and requirement. Nutr Rev 2000;58:363–9.
14. Suzuki LA, Meller PJ, Ponterotto JG. Handbook of multicultural assessment. San Francisco, CA, USA: Jossey-Bass, 1996.
15. Bleichrodt N, Shrestha RM, West CE, Hautvast JG, van de Vijver F, Born MP. The benefits of adequate iodine intake. Nutr Rev 1996;54(4 pt 2):S72–8.
16. US Department of Agriculture. Design and operation: The continuing survey of food intakes by individuals and the diet and health knowledge survey, 1994–96. Bethesda, MD, USA: USDA, 1997.
17. Food and Nutrition Research Institute. Recommended dietary allowances for Filipinos. Bicutan, Taguig: Department of Science and Technology, 1987.
18. Huda SN, Grantham-McGregor SM, Tomkins A. Cognitive and motor functions of iodine-deficient but euthyroid children in Bangladesh do not benefit from iodized poppy seed oil (Lipiodol). J Nutr 2001;131:72–7.
19. Whaley SE, Sigman M, Neumann C, Bwibo N, Guthrie D, Weiss RE, Alber S, Murphy SP. The impact of dietary intervention on the cognitive development of Kenyan school children. J Nutr 2003;133(11 suppl 2):3965S–71S.
20. Sigman MP, Espinosa MP, Whaley SE. Mild undernutrition and the cognitive development of Kenyan schoolchildren. In: Nutrition, health and child development. Research advances and policy recommendations. Washington, DC: PAHO/World Bank, 1998.
21. Mendez MA, Adair LS. Severity and timing of stunting in the first two years of life affect performance on cognitive tests in late childhood. J Nutr 1999;129:1555–62.
22. Huda SN, Grantham-McGregor SM, Rahman KM, Tomkins A. Biochemical hypothyroidism secondary to iodine deficiency is associated with poor school achievement and cognition in Bangladeshi children. J Nutr 1999;129:980–7.
23. Colborn T. Neurodevelopment and endocrine disruption. Environ Health Perspect 2004;112:944–9. doi:10.1289/ehp.6601. Available at: http://dx.doi.org/. Accessed 10 December 2006.
24. Bigsby R, Chapin RE, Daston GP, Davis BJ, Gorski J, Gray LE, Howdeshell KL, Zoeller RT, vom Saal FS. Evaluating the effects of endocrine disruptors on endocrine function during development. Environ Health Perspect 1999;107(suppl 4):613–8. Available at: http://ehpnet1.niehs.nih.gov/docs/1999/suppl-4/613-618bigsby/abstract.html. Accessed 10 December 2006.
25. Hess SY, Zimmermann MB, Arnold M, Langhans W, Hurrell RF. Iron deficiency anemia reduces thyroid peroxidase activity in rats. J Nutr 2002;132:1951–5.
26. Arthur JR, Nicol F, Beckett GJ. Selenium deficiency, thyroid hormone metabolism, and thyroid hormone deiodinases. Am J Clin Nutr 1993;57(2 suppl):236S–9S.
27. Arthur JR, Beckett GJ. Thyroid function. Br Med Bull 1999;55:658–68.
28. Petkovich M. Regulation of gene expression by vitamin A: The role of nuclear retinoic acid receptors. Annu Rev Nutr 1992;12:433–71.
29. Gaitan E, Cooksey RC, Lindsay RH. Factors other than iodine deficiency in endemic goiter: Goitrogens and protein-calorie malnutrition (PCM). In: Dunn JT, Pretell EA, Daza CH, Viteri FE, eds. Towards the eradication of endemic goiter, cretinism and iodine deficiency. Washington, DC: Pan American Health Organization/World Health Organization, 1986.

Sensitivity and specificity of a short questionnaire for food insecurity surveillance in Iran

Saeed Dastgiri, Helda Tutunchi, Alireza Ostadrahimi, and Soltanali Mahboob

Abstract

Background. *Food insecurity is frequent in both developed and developing countries, affecting from 5% to 25% of the general population. It has considerable health impacts on the physical, social, and psychological status of individuals in communities suffering from food insecurity.*

Objective. *The aim of this study was to document the epidemiologic features of food insecurity in the northwest region of Iran and to evaluate the sensitivity and specificity of a short-form (six items) questionnaire for screening of food insecurity in the region.*

Methods. *This cross-sectional study was conducted on 300 subjects (132 male and 168 female) selected randomly in the Asadabadi area of the northwest of Iran. Information on food consumption was obtained by a 24-hour food-recall questionnaire for 3 days in a week. This information was compared with the data from the Household Food Security Scale (six-item short questionnaire) to assess the applicability of this short scale for the surveillance of food insecurity. Hunger was defined as inadequate intake of energy. Hidden hunger was defined as adequate intake of energy and inadequate intake of one (or more) of four key nutrients (protein, calcium, vitamin A, and vitamin* B_2*).*

Results. *The prevalence of hunger and hidden hunger in the area according to the 24-hour food-recall questionnaire was 26% and 42%, respectively. Only 32% of the study population was secure in terms of having access to all key nutrients. The sensitivity, specificity, and accuracy of the short questionnaire for screening for hunger in the population were 98.7%, 85.5%, and 89%, respectively; and the corresponding values for hidden hunger were 23.5%, 96.9%, and 56.3%.*

Conclusions. *Our findings indicate that food insecurity is prevalent in the northwest of Iran. The short questionnaire (six items) may be used as a simple, low-cost, rapid, and useful tool for the screening of food insecurity and energy intake in similar areas.*

Key words: Food insecurity, Iran, nutritional epidemiology, short questionnaire

Introduction

The concept of household food insecurity includes problems with the quantity of available food, uncertainty about food supply, and experience of hunger in life [1, 2]. Food insecurity is frequent in both developed and developing countries, affecting from 5% to 25% of the general population in different research reports [3–9]. It has considerable health impacts on the physical, social, and psychological status of individuals in communities suffering from food insecurity. It may also affect the quality of life of households [10]. Various techniques and methods have been used to measure food insecurity in many countries [11–13].

The aim of this study was to document the epidemiologic features of food insecurity in the northwest region of Iran, and to evaluate the sensitivity and specificity of a short-form (six items) questionnaire for screening for food insecurity in the region.

Methods

This cross-sectional study was conducted on a total of 330 eligible individuals selected by simple random sampling from the study population using available

Saeed Dastgiri is affiliated with the Department of Community and Family Medicine and the National Public Health Management Center, Tabriz University of Medical Sciences, Tabriz; Helda Tutunchi and Alireza Ostadrahimi are affiliated with the School of Public Health and Nutrition, Tabriz University of Medical Sciences, Tabriz; Soltanali Mahboob is affiliated with the Centre for Nutritional Research, Tabriz University of Medical Sciences, Tabriz.

Please address queries to the corresponding author: Saeed Dastgiri, Department of Community and Family Medicine, School of Medicine, Tabriz University of Medical Sciences, Tabriz, Iran; e-mail: saeed.dastgiri@gmail.com.

records at the Asadabadi Medical Centre. Eligible individuals were followed and encouraged to come to the study setting for an interview, anthropometric measurements, and dietary assessment. The participation rate was 90%. Participants were required to sign an informed consent (showing the details of the study) to participate in the program. Approval for this study was obtained from the Research Committee of the Food and Nutrition Security Office of the Tabriz University of Medical Sciences.

Three hundred participants, including 132 males and 168 females, were eventually enrolled in the study. The setting of the study was the Asadabadi region of Tabriz, one of the major cities in the northwest of Iran. The area has previously been selected and described in detail as a reliable representative sample of the general population in terms of socioeconomic status, lifestyle, general health status, and population composition [14].

Information on food consumption was obtained with a validated 24-hour food-recall questionnaire over 3 non-consecutive days. These results were compared with data from the Household Food Security Scale [15, 16] to assess the applicability of this short scale for the surveillance of food insecurity. The short questionnaire had six questions (**table 1**). If the participants responded to two or more of the six items, they were considered food insecure. The questions were validated (in the local language) by a pilot study before starting the main research. To do this, the questionnaire was distributed to 20 individuals with the same characteristics and from the same area as the main study participants. The pilot study was carried out on the first 20 subjects of the whole study group. After assessment and evaluation of the results of this pilot, the next individuals (n = 330) were studied as the main part of the research. The results were then statistically analyzed to ensure that the short-scale questionnaire was valid for assessing household food insecurity in the main part of the research.

TABLE 1. Affirmative responses to individual items on the short questionnaire[a]

Question	No. (%)
1. Did you ever cut the size of meals or skip meals because of lack of money for food in the last 12 months? (Yes, No)	67 (22.3)
2. If yes, how often? (Almost every month, some months but not every month, only 1 or 2 months)	59 (19.6)
3. Did you ever eat less than you felt you should because there was not enough money to buy food in the last 12 months? (Yes, No)	62 (20.6)
4. Were you ever hungry but didn't eat because you couldn't afford enough food in the last 12 months? (Yes, No)	51 (17.0)
5. Food didn't last, and didn't have money to get more. (Was that often, sometimes, or never true for you in the last 12 months?)	56 (18.6)
6. Couldn't afford to eat balanced meals. (Was that often, sometimes, or never true for you in the last 12 months?)	178 (59.3)

a. "Yes" is the affirmative response to questions 1, 3, and 4; "almost every month" and "some months but not every month" are affirmative responses to question 2; "often" and "sometimes" are affirmative responses to questions 5 and 6. Two or more affirmative responses to the questions indicate food insecurity.

In this study, hunger was defined as inadequate intake of energy. Hidden hunger was defined as adequate intake of energy and inadequate intake of one (or more) of four key nutrients: protein, calcium, vitamin A, and vitamin B_2. Thus in hidden hunger, dietary intake provides adequate energy but does not provide sufficient amounts of protein and micronutrients. Food insecurity was defined as uncertain or limited availability of adequate supplies of nutritional and safe food in a socially acceptable way [17].

Height and weight were measured by nutritionists. Body weight was measured with a calibrated beam scale and was recorded to the nearest 0.5 kg. The subjects were measured barefoot wearing light clothing. Height was measured with a mounted tape with the subjects' arms hanging freely at their sides and recorded to the nearest 0.5 cm. Body-mass index (BMI) was then calculated as the weight in kilograms divided by the square of the height in meters. As recommended by the Iranian Ministry of Health for the whole country, underweight, overweight, and obesity were defined as BMI less than 18.5, 25 to 29.9, and 30 or more, respectively [18]. Data were analyzed and presented by descriptive statistics (mean and SD) and some epidemiologic indicators, including prevalence rate, sensitivity, specificity, error rate, predictive value, and likelihood ratio. Confidence intervals (95%) were calculated where applicable.

Results

Table 2 shows the distribution of food security and insecurity according to BMI in the study subjects. Food insecurity significantly increased the risk of underweight in the study subjects (RR = 13.2, CI: 6.9–25.5), while it decreased the risk of overweight and obesity (RR = 0.30, CI: 0.18–0.49 and RR = 0.32, CI: 0.16–0.62, respectively). The prevalence rates of hunger and hidden hunger according to the 24-hour food-recall questionnaire were 26% (CI: 21–31) and 42% (CI: 37–48), respectively. Only 32% (CI: 27–38) of the study population was secure in terms of having access to all key nutrients (including protein, calcium, vitamin A, and vitamin B_2).

The proportional distribution of the responses by

TABLE 2. Distribution of food security according to body-mass index (BMI)

Status	BMI category			
	< 18.5	18.5–24.9	25–29.9	≥ 30
Food secure—no. (%)	9 (4.7)	44 (23.0)	88 (46.0)	50 (26.1)
Food insecure—no. (%)	68 (62.3)	17 (15.5)	15 (13.9)	9 (8.2)
Total—no. (%)	77 (25.6)	61 (20.3)	103 (34.3)	59 (19.6)
Relative risk (95% CI)	13.2 (6.9–25.5)	0.68 (0.4–1.1)	0.30 (0.18–0.49)	0.32 (0.16–0.62)

CI, confidence interval

participants to the individual items on the Household Food Security Scale (six-item short questionnaire) is presented in **table 1.** Of the total respondents, 178 (59.3%) reported that they could not afford to eat balanced meals. At the same time, 109 (36.3%) of the respondents gave affirmative responses to two or more of the six items, indicating food insecurity.

Table 3 shows some epidemiologic indicators of the short questionnaire in two groups identified as having hunger or hidden hunger. The sensitivity, specificity, and accuracy of the short questionnaire for screening for hunger in the population were 98.7% (CI: 93–99), 85.5% (CI: 80–90), and 89%,(CI: 85–92), respectively; the corresponding values for hidden hunger were 23.5% (CI: 17–32), 96.9% (CI: 92–99), and 56.3% (CI: 50–63).

Discussion

This cross-sectional study was conducted to assess the applicability of a short questionnaire (six items) for screening for food insecurity in the northwest region of Iran. The findings showed that the epidemiologic indexes of the questionnaire, including sensitivity, specificity, and accuracy, were acceptably high for screening and surveillance of food insecurity and hunger in the area.

The prevalence of food insecurity according to this short questionnaire was about 36% in the study population. Gulliford and colleagues reported a similar figure of 25% from Trinidad and Tobago [7]. The percentages of affirmative responses to the six questions in our study were 22%, 20%, 21%, 17%, 19%, and 59%, respectively. In comparison with similar figures from the United States, the current population survey and Caribbean studies showed that the prevalence of food insecurity based on the affirmative responses to six items of the food security measurement was relatively high in Iran [6, 7, 19].

Our findings showed an association of food insecurity and BMI in the study population. Food insecurity increased the rate of underweight and decreased the rates of overweight and obesity. Some studies have reported an association between food insecurity and BMI. In a nationally representative sample of 6,506 individuals from Finland, Sarlio-Lahteenkorva and Lahelma showed that underweight subjects were at higher risk for food insecurity than obese or normal subjects [20]. Townsend et al. found an association between food insecurity and overweight in the female population, whereas there was no such association in male subjects [21]. A study from the United States showed that although mild or moderate food insecurity was associated with a higher risk of obesity, severe food insecurity was associated with a lower risk of obesity [22, 23]. Another research study reported an association of food insecurity with underweight but not with obesity [7].

These studies indicate that food insecurity may be associated either with thinness or with obesity in different populations, depending on the household's

TABLE 3. Indicators of the questionnaire according to hunger status

Indicator	Hunger—% (95% CI)	Hidden hunger—% (95% CI)
Sensitivity	98.7 (93.1–99.8)	23.5 (16.9–31.8)
Specificity	85.5 (80.4–89.6)	96.9 (91.5–99.0)
Accuracy	89 (85–92.1)	56.3 (49.7–62.7)
False positive error rate	14.4 (10.4–19.6)	3.03 (1.0–8.5)
False negative error rate	1.2 (0.2–6.9)	76.4 (68.2–83.1)
Positive predictive value (pV+)	70.6 (61.5–78.4)	90.6 (75.8–96.8)
Negative predictive value (pV–)	99.5 (97.1–99.8)	50.5 (43.5–57.6)
Likelihood ratio positive (LR+)	6.8 (4.9–9.4)	7.66 (2.4–24.8)
Likelihood ratio negative (LR–)	0.01 (0.002–0.105)	0.78 (0.71–0.88)

CI, confidence interval

economic status, its eating habits, and the availability and accessibility of food. More local studies are needed to identify the key factors affecting the association between food insecurity and BMI. This will help health authorities in planning and developing strategies to prevent obesity and overweight in the population, considering the food insecurity status in the same community households and its influencing factors.

We conclude that food insecurity as a prevalent health problem in the study region can be detected by this six-item questionnaire. It may then be used as a simple, low-cost, rapid and useful tool for the screening and surveillance of food insecurity and energy intake in similar areas. It might not, however, be an appropriate tool to measure sufficient nutrient intake. Twenty-four-hour food recall may alternatively be used to measure specific nutrient intake.

Acknowledgments

We thank Miss Nazila Farrin and the staff of the Asadabadi Medical Centre of the Tabriz University of Medical Sciences for their assistance in this project. We also thank the Food and Nutrition Security Office of the Tabriz University of Medical Sciences for funding this project.

References

1. Alaimo K, Briefel RR, Frongillo EA Jr, Olson CM. Food insufficiency exists in the United States: Results from the third National Health and Nutrition Examination Survey (NHANES III). Am J Public Health 1998;88:419–26.
2. Kendall A, Olson CM, Frongillo EA Jr. Relationship of hunger and food insecurity to food availability and consumption. J Am Diet Assoc 1996;96:1019–24.
3. Carlson SJ, Andrews MS, Bickel GW. Measuring food insecurity and hunger in the United States: Development of a national benchmark measure and prevalence estimates. J Nutr 1999;129(2S suppl):510S–6S.
4. Qasemi H. Food security programme in Islamic Republic of Iran. Tehran: Institute of Food and Nutrition Research, 1998 (in Persian).
5. Zarafati Shoa N. Validity of Radimer-Cornell questionnaire in Tehran. MSc Thesis, Tehran Shahid Beheshti University of Medical Sciences, 2003 (in Persian).
6. Hamilton WL, Cook JT, Thompson WW, Buron LF, Frongillo EA Jr, Olson CM, Wehler CA. Household food security in the United States in 1995: Summary report of the Food Security Measurement Project. Alexandria, VA, USA: US Department of Agriculture, Food and Consumer Service, September 1997.
7. Gulliford MC, Mahabir D, Rocke B. Food insecurity, food choices, and body mass index in adults: Nutrition transition in Trinidad and Tobago. Int J Epidemiol 2003;32:508–16.
8. Kaiser LL, Townsend MS, Melgar-Quinonez HR, Fujii ML, Crawford PB. Choice of instrument influences relations between food insecurity and obesity in Latino women. Am J Clin Nutr 2004;80:1372–8.
9. Bickel G, Nord M, Price C, Hamilton W, Cook J. Measuring food security in the United States. Guide to measuring food security. Revised 2000. Washington, DC: US Department of Agriculture, Food and Nutrition Service, 2000.
10. Campbell CC. Food insecurity: A nutritional outcome or a predictor variable? J Nutr 1991;121:408–15.
11. Coates J, Frongillo EA, Rogers BL, Webb P, Wilde PE, Houser R. Commonalities in the experience of household food insecurity across cultures: What are measures missing? J Nutr 2006;136:1438S–48S.
12. Webb P, Coates J, Frongillo EA, Rogers BL, Swindale A, Bilinsky A. Measuring household food insecurity: Why it's so important and yet so difficult to do. J Nutr 2006;136:1404S–8S.
13. Swindale A, Bilinsky P. Development of a universally applicable household food insecurity measurement tool: Process, current status, and outstanding issues. J Nutr 2006;136:1449S–52S.
14. Asadabadi Population and Health Survey. Tabriz Med J 2003;suppl 59:1s–42s, 115s–120s.
15. Blumberg SJ, Bialostosky K, Hamilton WL, Briefel RR. The effectiveness of a short form of the Household Food Security Scale. Am J Public Health 1999;89:12131–4.
16. Gulliford MC, Nunes C, Rocke B. The 18 Household Food Security Survey items provide valid food security classifications for adults and children in the Caribbean. BMC Public Health 2006;6:26.
17. Radimer KL, Olson CM, Campell CC. Development of indicators to assess hunger. J Nutr 1990;120(suppl 11):1544–8.
18. Laquatra I. Nutrition for weight management In: Mahan K, Escot-Stump S, eds. Krause's food, nutrition and diet therapy, 11th ed. Philadelphia, PA, USA: Saunders, 2004:565.
19. Gulliford MC, Mahabir D, Rocke B. Reliability and validity of a short form household food security scale in a Caribbean community. BMC Public Health 2004;4:22.
20. Sarlio-Lahteenkorva S, Lahelma E. Food insecurity is associated with past and present economic disadvantage and body mass index. J Nutr 2001;131:2880–4.
21. Townsend MS, Peerson J, Love B, Achterberg C, Murphy SP. Food insecurity is positively related to overweight in women. J Nutr 2001;131:1738–45.
22. Olson CM. Nutrition and health outcomes associated with food insecurity and hunger. J Nutr 1999;129(2S suppl):521S–4S.
23. Frongillo EA, Olson CM, Rauschenbach BA, Kendall A. Nutrition consequences of food insecurity in a rural New York State County. Institute for Research on Poverty Discussion Paper No. 1120-97. Madison, WI, USA: University of Wisconsin, 1997.

Economic inequality and undernutrition in women: Multilevel analysis of individual, household, and community levels in Cambodia

Rathavuth Hong and Rathmony Hong

Abstract

Background. *Many people in developing countries are still struggling to emerge from the realm of extreme poverty, where economic improvements tend to benefit a small, affluent group of the population and cause growing inequality in health and nutrition that affects the most vulnerable groups of the population, including women and children.*

Objective. *To examine how household and community economic inequality affects nutritional status in women using information on 6,922 nonpregnant women aged 15 to 49 years included in the 2000 Cambodia Demographic and Health Survey.*

Methods. *Nutritional status is defined with the use of the body-mass index (BMI). BMI less than 18.5 kg/m^2 is defined as undernourishment. The household wealth index was calculated from household ownership of durable assets and household characteristics. Community wealth is an average household wealth index at the community level. Household and community economic inequalities were measured by dividing the wealth index into quintiles. The effects of household and community economic inequality were estimated by multilevel analysis.*

Results. *Independently of community economic status and other risk factors, women in the poorest 20% of households are more likely to be undernourished than women in the richest 20% of households (RR = 1.63;* p *= .008). The results also show variation among communities in the nutritional status of women. Age, occupation, and access to safe sources of drinking water are significantly associated with women's nutritional status.*

Conclusions. *Improving household income and creating employment opportunities for women, in particular poor women, may be a key to improving the nutritional status of women in Cambodia.*

Key words: Cambodia, community level, inequality, undernourished women

Introduction

Many people in developing countries still live in extreme poverty [1]. In many countries, modest economic improvements are not distributed equally among the population, individually and across the communities within a country [2–4]. For these countries, economic growth tends to benefit only a small group of advantaged and affluent people and causes growing inequality in health and nutrition that affects particularly vulnerable groups of the population, such as women and children [5].

The economic inequality hypothesis posits that differences in individual health and nutrition status are observed among different economic strata [6, 7]. Moreover, the distribution of income in the society beyond the individual level also matters for population health and nutrition status. In another words, the health and nutritional status of individuals depends not only on individual and household economic status but also on the distribution of economic conditions within the community in which individuals live [8, 9]. In recent decades, several studies have used multilevel analysis to investigate the relationship between economic inequality and health, ranking from reported mental and physical health [10–13]. The results of these studies show negative effects of economic inequality on various health status indicators. However, to date no study, including those referred to earlier, has examined the effect of economic inequality on the nutritional status of women in a developing country using a multilevel analytical approach.

Rathavuth Hong is affiliated with the School of Public Health and Health Services, George Washington University, Washington, DC; Rathmony Hong is affiliated with the Department of Communicable Diseases Control, Ministry of Health, Cambodia.

Please direct queries to the corresponding author: Rathavuth Hong, Department of Global Health, School of Public Health and Health Services, George Washington University, 2175 K Street NW, Washington, DC 20037, USA; e-mail: rhong@gwu.edu.

This study used data from the 2000 Cambodia Demographic and Health Survey to examine two related but distinct hypotheses that capture the multilevel aspect of the economic–health/nutrition relationship. The first hypothesis is that the nutritional status of women improves when household economic status improves and that this relationship varies across communities. The second is that the nutritional status of women improves when the community they live in becomes richer, independently of other community characteristics.

Methods

This study is based on data from the 2000 Cambodia Demographic and Health Survey (CDHS). The survey included information from a nationally representative sample of 15,351 women aged 15 to 49 years nested in a sample of 471 clusters (communities). The sample design provides province-level estimates for the 12 large provinces and region-level estimates for 5 regions representing 13 smaller provinces and cities in Cambodia. The master sampling frame for the survey was based on the 1999 General Population Census, which consisted of 600 villages selected with probability proportional to the number of households in the village. The sampling design was a three-stage stratified process with a household response rate of 98.1% and an individual woman response rate of 98.7%. Details of the sample design are available in the main CDHS report [14]. Fifty percent of women interviewed in the survey were randomly selected for anthropometric measurements of weight and height. This study is limited to 6,922 nonpregnant women aged 15 to 49 years with valid anthropometric measurements included in the survey. The nutritional status of women is estimated from the body-mass index (BMI), calculated as the body weight in kilograms divided by the square of the height in meters. Undernourished women are defined as those with BMI less than 18.5 kg/m^2 [15].

The CDHS did not collect direct information on household income and expenditures. This study assesses the economic status of women using household wealth index as a proxy indicator. The analysis uses principal component analysis (PCA) to estimate a household wealth index, which quantifies differences in household economic status, from several asset variables [16]. The asset variables are household ownership of electricity, radio, television, refrigerator, bicycle, motorcycle/scooter, car/truck, telephone, shared toilet, wardrobe, sewing machine/loom, boat with/without motor, ox/horse cart, and farm land; and household sources of drinking water in the rainy season and the dry season, type of toilet facility, type of floor in dwelling, type of cooking fuel, and type of roofing material. The household wealth index is a composite measure of the cumulative living standard of a household, which places individual households on a continuous scale of relative wealth or economic status. The household wealth index is an individual household score that ranks households from the lowest to the highest economic status. Household economic status represents the economic status of the women who live in the household. Communities are represented by villages or clusters of households. Community economic status is an average household economic status or wealth index at the community level. Economic inequality is measured by dividing household wealth index and community wealth index into quintiles. The lowest household quintile represents the poorest 20% of households, and the lowest community quintile represents the poorest 20% of communities; the highest household quintile represents the richest 20% of households, and the highest community quintile represents the richest 20% of communities.

The effects of household economic inequality and community economic inequality on the nutritional status of women were estimated after adjusting for the effects of selected individual-, household-, and community-level covariates relative to intrahousehold food distribution and food consumption behavior and habits that could have affected nutritional status [17–19]. The individual- and household-level covariates include women's age (15–19, 20–24, 25–29, 30–34, 35–39, 40–44, and 45–49 years), total number of childbirths (none, one to four, five or more), marital status (never married, currently married, formerly married), age at first childbirth (no children, < 19, 19–20, 21+ years), educational level (no education, primary 1–4 years, primary 5+ years, secondary or higher), occupation (not working, professional/sales/service, agriculture/domestic, manual), household's source of drinking water (unsafe source, safe source), type of toilet facility (no toilet, nonhygienic toilet, hygienic toilet). The community-level covariates include residence (urban, rural), and geographic region (Phnom Penh, Plain, Great Lake, Coastal, Plateau/Mountain). See the footnotes of **table 1** for further details on variable definitions.

The effects of household economic inequality, community economic inequality, and other covariates on women's nutritional status were estimated by a multilevel logistic regression procedure (xtlogit) in STATA version 8.1. The multilevel statistical procedure provides a robust analytical framework when the exposure (economic inequality) affects the outcome at more than one level simultaneously. We conducted a multilevel analysis of the 6,922 women at level 1, nested within 471 communities at level 2. Since the outcome is binary (undernourished: no, yes), we used a multilevel logistic procedure based on a logit-link function. This function fits a fixed effect and a random effect for cross-sectional data. A fixed effect represents the effect of exposure at the level of analytical units (woman or community) independently of the group effects at the community level, and a random effect represents

TABLE 1. Sample distribution and nutritional status of nonpregnant women aged 15 to 49 according to household economic status and selected characteristics, Cambodia 2000

Characteristic	No.	% of total	% under-nutrition
Total sample	6,922	100	20.6
Individual/household covariates			
Household economic status			
Highest	1,572	22.7	16.7
Higher	1,370	19.8	20.5
Middle	1,335	19.3	20.6
Poorer	1,295	18.7	21.5
Poorest	1,349	19.5	24.2
Age group (yr)			
15–19	1,640	23.7	25.4
20–24	854	12.3	17.5
25–29	863	12.5	17.5
30–34	1,007	14.5	18.6
35–39	1,006	14.5	19.6
40–44	871	12.6	20.4
45–49	681	9.8	21.3
Total no. of childbirths			
0–1	3,196	46.2	22.0
2–4	2,021	29.2	18.7
≥ 5	1,705	24.6	20.2
Marital status			
Never married	2,287	33.1	23.1
Currently married	3,988	57.6	19.2
Formerly married	646	9.3	20.2
Age at 1st childbirth (yr)			
No children	2,580	37.3	22.5
< 19	1,082	15.6	16.9
19–20	1,146	16.6	18.6
≥ 21	2,113	30.5	21.1
Education			
None	1,899	27.4	22.1
1–4 yr primary	2,054	29.7	20.6
≥ 5 yr primary	1,836	26.5	18.5
Secondary or higher	1,133	16.4	21.3
Occupation			
Not working	1,196	17.3	24.2
Professional/sales/services	1,291	18.7	13.9
Agriculture/domestic	3,950	57.1	21.8
Manual	453	6.6	20.4
Source of drinking water[a]			
Unsafe	4,671	67.5	21.7
Safe	2,228	32.2	18.1
Toilet facilities[b]			
None	5,208	75.2	21.6
Nonhygienic toilet	1,058	15.3	17.5
Hygienic toilet	655	9.5	17.3
Community-level covariates			
Community economic status			
Highest	1,468	21.2	18.1
Higher	1,593	23.0	18.1
Middle	1,392	20.1	23.1
Poorer	1,410	20.4	20.9
Poorest	1,058	15.3	24.0
Residence			
Urban	1,180	17.1	16.1
Rural	5,741	83.0	21.5
Region			
Phnom Penh	716	10.3	20.3
Plain	2,913	42.1	22.8
Great Lake	2,015	29.1	18.1
Coastal	537	7.8	15.8
Plateau/Mountain	741	10.7	22.0

a. Safe sources are piped water from the public water supply, tube wells, protected dug wells, and bottled water; unsafe sources are unprotected dug wells, rivers, streams, ponds, lakes, rainwater, tanker trucks, and other unknown sources.
b. Safe toilets are flush toilets or latrines connected to a sewage system; unsafe toilets are flush toilets or latrines unconnected to a sewage system.

the group effect (variations across communities) that happens simultaneously with, and independently of, the fixed effect. For example, the effect of community economic inequality on the nutritional status of women may be independent of the variation of the nutritional status of women across communities, because other community characteristics could affect the nutritional status of women independently. In the survey, certain categories of respondents were oversampled, and nonresponse rates varied from one geographic area to another. In our analysis, weights were used to restore the representativeness of the sample [14]. Results are presented as odds ratios (OR) with level of statistic significance (*p* values).

This study is based on secondary analysis of existing survey data with all identifying information removed. The survey required and obtained institutional review board (IRB) approval from ORC Macro IRB #1. It also required and acquired informed consent from women included in this study before asking any questions and before obtaining anthropometric measurements [14]; a comprehensive verbal consent form was read to each respondent by the interviewer before conducting the interview.

Results

Table 1 shows the sample distribution of the nonpregnant women aged 15 to 49 years according to household and community economic status and selected individual/household and community covariates. The women were more or less evenly distributed by household economic quintiles. About 19% to 20% of the women were in the lowest, lower, middle, and higher economic quintiles, and 23% were in the highest economic quintile. Almost one-quarter of the women (24%) were 15 to 19 years of age, and only 10% were 45 to 49 years of age. Forty-six percent of the women had no child or one child, 29% had two to four children, and 25% had five or more children at the time of the survey. One-third of the women in the sample had never been married, 58% were currently married, and 9% were formerly married (widowed, divorced, or separated). Thirty-seven percent of the women did not have any children, 16% had their first child before the age of 19, 17% at 19 or 20, and 31% at 21 or older. The large majority of the women (84%) had only primary education or less. About 17% of the women were not working; 19% worked in the professional, sales, and service sectors; 57% worked in the agricultural and domestic sectors; and 7% worked in the manual labor sector. Only about one-third of the women (32%) lived in households with access to safe drinking water, and 10% lived in households with a hygienic toilet facility. About 21%, 23%, 20%, and 29% of women lived in communities in the highest, higher, middle, and lower economic quintiles, respectively, and 15% lived in communities in the lowest economic quintile. The majority of women (83%) lived in rural areas. Forty-two percent of the women lived in the Plain region, 29% in the Great Lake region, 11% in the Plateau/Mountain region, 8% in the Coastal region, and 10% in the capital city of Phnom Penh,

About one in every five nonpregnant women (21%) aged 15 to 49 was undernourished. The distribution of undernutrition by household economic status shows a reverse relationship: as household economic status decreases, the proportion of undernourished women increases. The proportion of undernourished women was 17% in the highest household economic quintile and 24% in the lowest household economic quintile. There was a U-shaped relationship between undernutrition and age, with younger and older women having higher proportions of undernutrition than women aged 20 to 34 years. The proportions of undernourished women among those with a total number of zero to one, two to four, and five or more childbirths were 22%, 19%, and 20%, respectively. Single women had a higher prevalence of undernutrition (23%) than women who were currently married (19%) and women who were formerly married (20%). Distribution according to age at first childbirth shows that the prevalence of undernutrition among women with no children was 23%, and as the age at first childbirth increased, the prevalence of undernutrition increased from 17% among those who first gave birth before the age of 19 to 21% among those who first gave birth at the age of 21 or older. The relationship between undernutrition and women's educational levels was inconsistent. Women who did not work had the highest proportion of undernutrition (24%) , and those who had a professional, sales, or service job had the lowest (14%). The prevalence of undernutrition was 22% among women who worked in the agricultural or domestic sectors and 20% among those who did manual labor. Living in a household with a safe source of drinking water and with a hygienic toilet facility was associated with a lower prevalence of undernutrition. Women in the highest and the higher quintiles of community wealth status had a lower prevalence of undernutrition (18%) than those in the lowest quintile of community wealth status (24%). The prevalence rates of undernutrition among women in the middle and the poorer community wealth quintiles were 23% and 21%, respectively. The prevalence of undernutrition was lower in urban areas than in rural areas (16% vs. 22%), and was much lower in the Coastal region (16%) than in other regions in Cambodia (18%–23%).

Effect of household wealth status and community wealth status on undernutrition

The unadjusted odds ratio of undernutrition among women in the lowest household wealth quintile was 1.61 in comparison with women in the highest household wealth quintile (OR = 1.61; 95% CI, 1.32 to 1.96; $p = .000$) (**fig. 1**).

Adjusting for individual- and household-level covariates of age of woman, total number of childbirths,

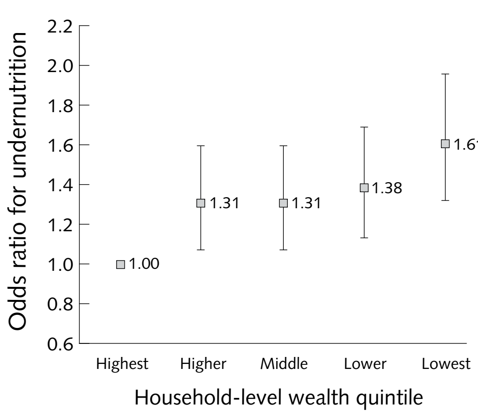

FIG. 1. Unadjusted effect of household economic status on undernutrition among nonpregnant women 15 to 49 years of age expressed as odds ratios with 95% confidence intervals, Cambodia 2000

marital status, age at first childbirth, educational level, occupation, source of drinking water, and type of toilet facility does not change the effect of household wealth status on women's nutritional status. After adjustment for individual- and household-level covariates, women in the poorest 20% of households were 1.52 times as likely to be undernourished as women in the richest 20% of households (OR = 1.52; 95% CI, 1.09 to 2.22; *p* = .013) (model 1, **table 2**). The random parameter result in model 1 (**table 2**) suggests that variation across communities is statistically significant.

Community wealth also has a smaller but significant unadjusted effect on women's nutritional status. Women who lived in the poorest communities were 1.47 times as likely to be undernourished as women who lived in the richest communities (OR = 1.47; 95% CI, 1.17 to 1.83; *p* = .001) (**fig. 2**).

Adjusting for community covariates of urban/rural residence and region slightly reduces the effect of community wealth (OR = 1.41; 95% CI, 1.05 to 1.89;

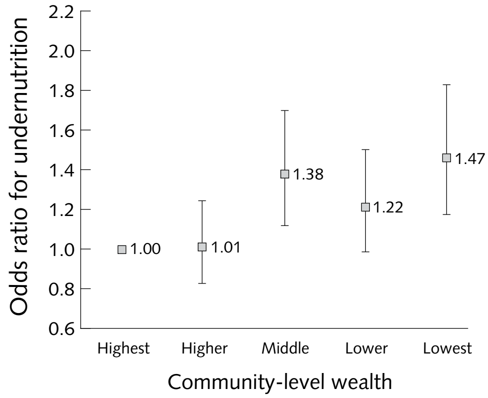

FIG. 2. Unadjusted effect of community economic status on undernutrition among nonpregnant women 15 to 49 years of age expressed as odds ratios with 95% confidence intervals, Cambodia 2000

TABLE 2. Multilevel logistic regression estimates of adjusted effects of household and community economic status and selected factors on undernutrition among nonpregnant women aged 15 to 49, Cambodia 2000

	Model 1		Model 2		Model 3	
Characteristic	OR	*p*	OR	*p*	OR	*p*
Fixed parameter						
Individual covariates						
Household economic status						
Highest	1.00	—			1.00	—
Higher	1.27	0.096			1.40	0.029
Middle	1.27	0.138			1.39	0.054
Poorer	1.32	0.088			1.45	0.038
Poorest	1.52	0.013			1.63	0.008
Age group (yr)						
15–19	1.00	—			1.00	—
20–24	0.64	0.000			0.62	0.000
25–29	0.61	0.000			0.60	0.000
30–34	0.67	0.003			0.65	0.001
35–39	0.73	0.029			0.71	0.019
40–44	0.74	0.051			0.72	0.033
45–49	0.81	0.173			0.77	0.102
Total no. of births						
0–1	1.00	—			1.00	—
2–4	0.97	0.815			0.97	0.815
5+	1.00	0.980			1.01	0.922
Marital status						
Never married	1.00	—			1.00	—
Currently married	0.84	0.303			0.84	0.291
Formerly married	0.87	0.457			0.87	0.461
Age at first childbirth (yr)						
No children	1.00	—			1.00	—

continued

TABLE 2. Multilevel logistic regression estimates of adjusted effects of household and community economic status and selected factors on undernutrition among nonpregnant women aged 15 to 49, Cambodia 2000 *(continued)*

	Model 1		Model 2		Model 3	
Characteristic	OR	*p*	OR	*p*	OR	*p*
< 19	0.96	0.835			0.96	0.848
19–20	1.13	0.541			1.14	0.519
≥ 21	1.33	0.136			1.35	0.124
Education						
None	1.00	—			1.00	—
1–4 yr primary	0.99	0.859			0.97	0.731
≥ 5 yr primary	0.87	0.140			0.86	0.095
Secondary or higher	1.27	0.040			1.24	0.060
Occupation						
Not working	1.00	—			1.00	—
Professional/sales/ services	0.55	0.000			0.56	0.000
Agriculture/ domestic	0.82	0.025			0.82	0.027
Manual	0.80	0.124			0.80	0.109
Source of drinking water[a]						
Unsafe	1.00	—			1.00	—
Safe	0.83	0.012			0.79	0.001
Toilet facilities[b]						
None	1.00	—			1.00	—
Nonhygienic toilet	0.92	0.538			0.94	0.632
Hygienic toilet	1.05	0.753			0.99	0.957
Community-level covariates						
Community economic status						
Highest			1.00	—	1.00	—
Higher			0.94	0.658	0.77	0.082
Middle			1.25	0.119	0.95	0.763
Poorer			1.11	0.465	0.84	0.292
Poorest			1.41	0.021	0.97	0.867
Residence						
Urban			1.00	—	1.00	—
Rural			1.29	0.024	1.18	0.155
Region						
Phnom Penh			1.00	—	1.00	—
Plain			0.94	0.682	0.82	0.252
Great Lake			0.69	0.019	0.58	0.001
Coastal			0.65	0.023	0.54	0.002
Plateau/Mountain			0.85	0.371	0.71	0.074
Random parameter						
Community level (ρ)	0.03	0.000	0.02	0.002	0.02	0.014
No. of women	6,859		6,911		6,859	

OR, odds ratio

a. Safe sources are piped water from the public water supply, tube wells, protected dug wells, and bottled water; unsafe sources are unprotected dug wells, rivers, streams, ponds, lakes, rainwater, tanker trucks, and other unknown sources.

b. Safe toilets are flush toilets or latrines connected to a sewage system; unsafe toilets are flush toilets or latrines unconnected to a sewage system.

p = .021) (model 2, **table 2**). The random parameter result in model 2 suggests that variation across communities is also statistically significant in this model.

In the full model (model 3, **table 2**), the effect of household wealth on nutritional status is further sharpened when all individual, household, and community covariates and community wealth status are adjusted for (OR = 1.63; 95% CI, 1.13 to 2.33; *p* = .008). The effect of community wealth status disappears in the model, even though the variation across communities remains statistically significant.

Effect of other covariates on undernutrition

After household wealth status, community wealth status, and other covariates were controlled for in model 3, women's age and occupation were significantly associated with undernutrition. Women aged 20 to 39 and women who worked in the professional, sales, or service sectors were at a lower risk for undernutrition than older women and women who did not work. Women who lived in households with a safe source of drinking water were at much lower risk for undernutrition than women who lived in households with an unsafe source of drinking water (OR = 0.79; 95% CI, 0.68 to 0.91; *p* = .001), and women who lived in the Great Lake region and the Coastal region were less likely to be undernourished than women living in Phnom Penh and other regions (OR = 0.58; 95% CI, 0.42 to 0.80; *p* = .001, and OR = 0.54; 95% CI, 0.37 to 0.81; *p* = .001, respectively) (model 3, **table 2**). No other covariates had a significant relationship with undernutrition among women in Cambodia.

Discussion

In this study, which is based on data from a national representative cross-sectional survey, we found evidence that although the distribution of household economic status significantly affects the nutritional status of women in Cambodia, the effects of community economics are weak and unstable. First, the results show that the association between the prevalence of undernutrition in women and household economic status is negative. The odds of undernutrition increase as household economic level decreases. However, this association is mostly confined to the poorest household economic quintile. The relationship is not linear; the highest probability of undernutrition is observed among the poorest household economic quintile, and the curve is relatively flat at the second, third, and fourth household economic levels (**fig. 1**).

Second, we found that community economic status has a significant unadjusted effect (**fig. 2**) and community-level (urban/rural residence and region) adjusted effects (model 2, **table 2**) on women's nutritional status. Nonetheless, the strength of the effect of community economic status is weak and its direction is irregular compared with the effect of household economic status.

Third, the results show that after adjusting for community economic status and individual, household, and community covariates, the effect of household economic status on women's nutritional status remains statistically significant and independent of community economic status and other covariates. After adjusting for household economic status and other factors, the effect of community economic status on the nutritional status of women disappears. This finding suggests that in Cambodia, community economic status depends on household economic status. In other words, the poorest and the poorer communities may be clusters of the majority of the poorest and the poorer households, respectively; and the richer and the richest communities may be clusters of the majority of the richer and the richest households, respectively. Moreover, factors that are more closely related to the individual, such as household economic status, are expected to have a stronger relationship with individual health status than factors that are more distantly related.

Fourth, this does not mean that other community characteristics are not important determinants of individual health. For example, a public health condition that exists in the community can imply a significant public health risk if exposure to the condition is widespread in the community. In Cambodia, even though community economic status may not be one of these conditions, such community conditions affecting the nutritional status of women do exist, as evidenced by the fact that there are statistically significant community variations (random effects) in women's undernutrition that cannot be explained by household economic status, community economic status, and the individual, household, and community covariates used in the analysis.

Study limitations

This study has a number of limitations. First, the study can be criticized for using an indirect measure of household and community wealth status. However, in developing countries such as Cambodia, where it is hard to obtain reliable income and expenditure data, a household asset index generally provides a good proxy for household and community economic status [20]. Second, this study is based on data from a cross-sectional survey, and therefore we were not able to determine the direction of any causal association. Also, the direction of the relationship between household economic status and women's nutritional status could potentially be bidirectional. Third, community variation in the nutritional status of women may be a result of the effects of certain community characteristics on women's nutritional status that we were not able to include in our analysis, even though we found evidence of these effects.

In this study, using a multilevel analysis approach, we investigated the relationship between household and community economic status and the nutritional status of women in Cambodia. We found that although household economic status is positively associated with the nutritional status of women independently of other covariates, the effects of community economic status on the nutritional status of women in Cambodia are inconsistent and unstable. Nonetheless, we found that other characteristics of the community affect women's nutrition and that there is evidence of community variation in women's nutritional status. Our findings suggest that improving household income, reducing economic disparities, and creating employment opportunities for women, particularly for poorer women, may be key to improving the nutritional status of women in Cambodia.

References

1. United Nations System Standing Committee on Nutrition. Fifth report on the world nutrition situation: Nutrition for improved development outcomes. New York: United Nations, 2004.
2. Houweling TA, Kunst AE, Borsboom G, Mackenbach JP. Mortality inequalities in times of economic growth: Time trends in socioeconomic and regional inequalities in under 5 mortality in Indonesia, 1982–1997. J Epidemiol Community Health 2006;60:62–8.
3. Gwatkin DR, Bhuiya A, Victora CG. Making health systems more equitable. Lancet 2004;364:1273–80.
4. Sastry N. Trends in socioeconomic inequalities in mortality in developing countries: The case of child survival in Sao Paulo, Brazil. Demography 2004;41:443–64.
5. Sicchia SR, Maclean H. Globalization, poverty and women's health: Mapping the connections. Can J Public Health 2006;97:69–71.
6. Kennedy BP, Kawachi I, Prothrow-Stith D. Income distribution and mortality: Cross sectional ecological study of the Robin Hood index in the United States. BMJ 1996;312:1004–7.
7. Blakely TA, Lochner K, Kawachi I. Metropolitan area income inequality and self-rated health—A multi-level study. Soc Sci Med 2002;54:65–77.
8. Blakely TA, Kennedy BP, Kawachi I. Socioeconomic inequality in voting participation and self-rated health. Am J Public Health 2001;91:99–104.
9. Mahmud Khan M, Hotchkiss DR, Berruti AA, Hutchinson PL. Geographic aspects of poverty and health in Tanzania: Does living in a poor area matter? Health Policy Plan 2006;21:110–22.
10. Blakely TA, Kennedy BP, Glass R, Kawachi I. What is the lag time between income inequality and health status? J Epidemiol Community Health 2000;54:318–9.
11. Kahn RS, Wise PH, Kennedy BP, Kawachi I. State income inequality, household income, and maternal mental and physical health: Cross sectional national survey. BMJ 2000;321:1311–5.
12. Subramanian SV, Kawachi I, Kennedy BP. Does the state you live in make a difference? Multilevel analysis of self-rated health in the US. Soc Sci Med 2001;53:9–19.
13. Subramanian SV, Delgado I, Jadue L, Vega J, Kawachi I. Income inequality and health: Multilevel analysis of Chilean communities. J Epidemiol Community Health 2003;57:844–8.
14. National Institute of Statistics, Directorate General of Health [Cambodia], ORC Macro. Cambodia Demographic and Health Survey 2000. Phnom Penh Cambodia and Calverton, MD, USA: National Institute of Statistics, Directorate General of Health and, ORC Macro, 2001.
15. Centers for Disease Control and Prevention. BMI--Body Mass Index: About BMI for adults. Available at: http://www.cdc.gov/nccdphp/dnpa/bmi/adult_BMI/about_adult_BMI.htm. Accessed 12 December 2006.
16. Rutstein SO, Johnson K. The DHS Wealth Index. DHS Comparative Reports No. 6. Calverton, MD, USA: ORC Macro, 2003.
17. Sudo N, Sekiyama M, Maharjan M, Ohtsuka R. Gender differences in dietary intake among adults of Hindu communities in lowland Nepal: Assessment of portion sizes and food consumption frequencies. Eur J Clin Nutr 2006;60:469–77.
18. Sudo N, Sekiyama M, Watanabe C, Bokul AT, Ohtsuka R. Gender differences in food and energy intake among adult villagers in northwestern Bangladesh: A food frequency questionnaire survey. Int J Food Sci Nutr 2004;55:499–509.
19. Lemke S, Vorster HH, van Rensburg NS, Ziche J. Empowered women, social networks and the contribution of qualitative research: Broadening our understanding of underlying causes for food and nutrition insecurity. Public Health Nutr 2003;6:759–64.
21. Filmer D, Pritchett L. Estimating wealth effects without expenditure data or tears: An application to educational enrollment in States of India. Demography 2001;38: 115–32.

Maternal income-generating activities, child care, and child nutrition in Mali

June N. Pierre-Louis, Diva Sanjur*, Malden C. Nesheim, Dwight D. Bowman, and Hussni O. Mohammed

Abstract

Background. *Women in sub-Saharan Africa play a key role in household food security. The income-generating activities of mothers are postulated to be related to the nutritional status of children.*

Objective. *The objective of the study was to examine whether maternal income-generating activities, maternal food production, and child care were determinants of the nutritional status of children in rural West Africa. The study hypotheses were that maternal income-generating activities and maternal food production are positively associated with children's dietary intake and anthropometry, and that maternal income-generating activities are not associated with child care.*

Methods. *Data were collected from a cross-sectional sample of mother–child pairs on maternal time use, child anthropometry, maternal food production, dietary intake, parasitic infection, and household, maternal, and child determinants of child nutritional status. The children were 12 to 36 months of age and included breastfed and nonbreastfed children. Food intake was assessed by the 24-hour recall method. The data were analyzed by multivariate regression and controlled for confounding variables.*

Results. *Time spent by the mother in income-generating activities was negatively associated with children's animal protein intake and height-for-age* ($p < .05$)*. Maternal cash crop production was positively associated with children's weight-for-height, whereas maternal staple food production was negatively associated with energy intake from non-breastmilk foods* ($p < .05$)*. The negative relationships observed for children's animal protein intake and children's height-for-age were not mediated by any child-care variable. Maternal supervision of feeding was a positive predictor of children's animal protein intake.* Giardia *infection was negatively related to children's weight gain* ($p < .05$)*.*

Conclusions. *Own-account cash crop farming by mothers benefits children's nutrition. Maternal income-generating activities in the context of extended families, sibling caretaking, and prolonged breastfeeding do not adversely affect child care.*

Key words: Africa, care, caregiving, child anthropometry, dietary intake, food intake, household food security, income-generating activities, maternal work, parasitic infection

June N. Pierre-Louis and Diva Sanjur were affiliated with the Division of Nutritional Sciences, Cornell University, Ithaca, New York, USA, at the time the study was conducted; Malden C. Nesheim is affiliated with the Division of Nutritional Sciences, Cornell University, Ithaca, New York, USA; Dwight D. Bowman is affiliated with the Department of Microbiology and Immunology, Cornell University, Ithaca, New York, USA; Hussni O. Mohammed is affiliated with the Department of Population Medicine, Cornell University, Ithaca, New York, USA.

Please direct queries to the corresponding author: June N. Pierre-Louis, 29 Lawrence Street, Greenlawn, New York 11740, USA; e-mail: junepierrelouis@yahoo.com.

This article is based on June Pierre-Louis' doctoral dissertation (Cornell University, 1995). It was presented at the annual meeting of the Federation of American Societies for Experimental Biology (FASEB) in Washington, DC, in April 1999.

*Deceased.

Introduction

Women throughout the world have the major responsibility for their families' nutrition. Therefore, any efforts to prevent malnutrition and enhance health depend substantially on women's activities and women's empowerment [1]. Maternal income-generating activities are hypothesized to be related to child nutrition by a number of mechanisms. Maternal income-generating activities increase household food availability by increasing food production or by increasing household income and food expenditures; they may decrease the time spent by mothers caring for, feeding, and breastfeeding their children [2] and increase caregiving by other individuals beside the mother. Maternal income-generating activities also may increase maternal access to resources to procure food, decision-making power, and control of income [3]. These factors and maternal education, which is closely related to maternal work [1], are characteristic of the economic and cultural context [3] and are hypothesized to mediate the effect of maternal income-generating activities on child nutrition.

The effects of maternal work on caregiving and child nutrition are as yet unclear and conflicting [1]. A study of 3- to 5-year-olds in Panama [4] and a study of children in Sri Lanka [5] demonstrated that maternal employment in Panama and maternal income in Sri Lanka were positively associated with children's dietary intake. In a study in Nicaragua, 12- to-18-month-old children of employed mothers residing in low-income urban neighborhoods had higher weight-for-height than children whose mothers were not employed, but substitute child care in the context of nuclear families was associated with lower height-for-age [6]. In the Philippines, participation of mothers in the labor force in the formal and informal sectors and child caretaking by siblings were negatively associated with anthropometric measurements of children under six [7], whereas a study in Tanzania found no relationship between the farming workload of mothers and the weight-for-age and dietary intake of children under three [8]. A study of children 12 to 71 months of age in Chad [9] found that caregivers' workloads and income were not associated with children's height-for-age, but that involvement of caregivers in decision-making about household food expenditures and family support for domestic work tasks were associated with children's height-for-age. Maternal income from handicrafts was negatively associated with child growth; these households, however, had no income from crop production. It is suggested that mothers also had less time to devote to child care in these households.

Food security and care are two of the pillars of good nutrition, along with health [10]. As farmers and traders, women in sub-Saharan Africa play a key role in household food security with regard to both food availability and access to food [10]. The objective of the study reported here was to examine the relationships among the income-generating activities of mothers, child care, and the dietary intake and anthropometry of their children in rural Africa.

Figure 1 shows the conceptual framework of the study. The study hypotheses were that maternal income-generating activities and maternal food production are positively associated with children's dietary intake and anthropometry, and that maternal income-generating activities are not associated with child care. The income-generating activities of African women are characteristic of their ethnic group and social class [11]. In the rural savannah of Mali, Bambara women generate their own income primarily by farming and selling food [12]. In addition to farming the household fields (*foroba*) with men, married women practice own-account farming, as in many countries in West Africa [11, 13]. A married woman may be given a plot of land to cultivate (*musoforo*) by her spouse's family in addition to gathering foods to generate income [12]. Although household food production is controlled by men [12], the food produced by the woman in her field belongs to her. This food production is often overlooked because it is small, yet the woman decides whether her family consumes it or whether she sells it, and it gives women autonomy from their spouses [13]. With the income a woman may buy condiments, other foods, soap, and clothing. Women also gather wild fruits, leaves, and legumes from the bush [12], which they may process or trade, for example, shea nuts to make shea nut butter and baobab leaves. It is hypothesized that time spent by the mother farming her own field is positively related to maternal food production, which, in turn, is positively related to how much food there is in the household—not only the food consumed directly from the farm but also other foods that are bought with the cash earned from the food the mother sells. It has been noted that women spend a greater percentage of their own income on food than do their husbands [14].

Child care is the provision in the household and the community of time, attention, and support to meet the physical, mental, and social needs of the growing child [15]. Maternal income-generating activities were hypothesized to be not associated with child care, because Malian culture is favorable to breastfeeding and breastfeeding is typically prolonged [16], and because the cultural norm is that the mother is the primary child caregiver during infancy and that after weaning, the child spends most of his or her time with other children of the same age supervised by older siblings or other women in the family [15, 17]. This

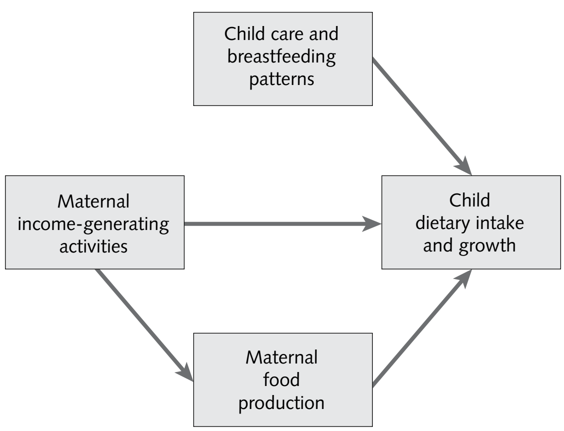

FIG. 1. Study conceptual framework

runs counter to earlier theoretical models that postulate that maternal work is negatively associated with child nutrition because the working mother has less time for child care [2] and because substitute child care is inadequate.

Methods

Study design

Data were collected on maternal time use, child care, and child anthropometry from a cross-sectional population sample of 65 mother–child pairs; data were collected from subsamples on maternal food production (53 mothers). dietary intake (45 children), parasitic infections (49 children), and household, maternal, and child determinants of children's nutritional status (53 households). Children were between 12 and 36 months of age; both breastfed and nonbreastfed children were studied. Data were collected during the rainy season in June 1993. This is the planting season, and it is called the hungry season because food intake and food stocks are low. Because food habits are related to ethnicity [1], one ethnic group, the Bambara, was studied. Ethnic group was determined by the mother, because some Bambara women in the study population were married to Fulani men, who were also farmers but traditionally are animal herders.

Study population

The study population was rural, residing in villages surrounding the town of Kolondiéba (population 5,000), the administrative center of the area. Kolondiéba is located in a remote area 200 km southeast of the capital city, near the border with Côte d'Ivoire. Some 50 km of unpaved road separates the town from the paved road. To the south, a river separates Kolondiéba from Côte d'Ivoire. The roads in Kolondiéba are frequently closed during the rainy season from June to October. There is no electricity and no land telephone communication in the villages. The majority of the population is Bambara, the largest ethnic group in Mali, who are mainly subsistence farmers. Agriculture is rainfed. Ninety percent of the population in the rural south of Mali is Moslem, and about half of marriages are polygamous [18]. The majority of Malian children do not go to school, and the literacy rate of women is 16% [19]. Since the data were collected in 1993, the living conditions of women and children under five have not changed dramatically. There has been a jump in school enrollment since 1999 [20].

Sampling methods

The eligibility criteria for the study were that the child was between 12 and 36 months of age, was Bambara, lived within 20 km of the town of Kolondiéba, and was not a twin. The sampling frame was a population register maintained by Save the Children. Except for eight children, the study child was the youngest child of the mother. The sample population was randomly selected and representative of the target population. A list of all eligible children was made from the population register. Because so few women in the study population had been to school, all mothers who had any schooling (as reported by the register) were included in the sample. The remaining women were sampled systematically by household. In households where there was more than one eligible child, one child was selected at random. The sample size was calculated based on finding a significant difference in energy intake (100 children). The weights and lengths of 88 children were measured at baseline. Some families had moved; a few children were deceased; some families did not want to participate in the study; a few children were part of the same family; and one mother was excluded because she had just given birth. The study sample size was further limited by practical considerations and difficult field conditions. The mean ages and weight-for-age z-scores of the children in the study sample, the subsample for dietary data, and the baseline sample were compared by *t*-tests; the differences were not statistically significant ($p < .05$).

Study variables

Data on maternal use of time were obtained to measure the productive workload or maternal income-generating activities and time spent in child-care activities, and to observe specific child-care practices. Maternal time use was observed by a trained local fieldworker from sunrise to sunset (6 a.m. to 6 p.m.). Maternal time use data on market days were excluded from the statistical analysis, because only 19 women went to the market on market day (one day a week) during this period during the planting season, although the majority of women were traders. Time spent in maternal income-generating activities, domestic work, child-care activities, social activities, and miscellaneous activities (personal care, walking, praying, etc.) was recorded to the nearest 5 minutes and expressed in hours in the statistical analysis. Maternal income-generating activities on nonmarket days were own-account farming, gathering shea nuts, and making shea butter.

Child-care activities were breastfeeding, feeding, bathing and dressing the child, playing with the child, and comforting the child. Other child-care variables that were observed, in addition to time spent breastfeeding and in child-care activities, were supervision of feeding by the mother, breastfeeding frequency or the number of times the child was nursed during the day, and the child's caregiver or the person who routinely watched the child.

Socioeconomic, demographic, and maternal food-production data were collected by a trained interviewer using a questionnaire. Household size and per capita livestock assets were proxies for socioeconomic status; household size is directly related to the number of economically productive adults in the household, whereas livestock assets primarily reflect the number of cattle, one of the most important household assets. Maternal food production was reported by the mother and was a categorical variable in the statistical analysis.

The food and nutrient intakes of 45 children were assessed by the 24-hour recall method. The 24-hour recall method was first explained to the mothers, and then the mothers were asked to observe what and how much their children ate during the day. The next day the mother was asked to recall the dietary intake of her child. The food intake of a subsample of eight children was measured by the food-weighing method; these data were used to calculate the median proportions of ingredients of mixed and prepared dishes measured by the 24-hour recall method. Energy intake was divided by the body weight in kilograms to control for the child's weight and size. The distribution of animal protein intake was skewed, and it was defined as a categorical variable for the statistical analysis (daily intakes < 10 g and ≥ 10 g).

The child's weight was measured to the nearest 10 g with a Seca balance by the first author. Stature was measured to the nearest centimeter by the first author and an assistant using a standard measuring board. The assistant placed the child's head against the headboard, eyes looking vertically, while the author extended the child's legs and held the child's feet perpendicular to the sliding footboard. For the statistical analysis, weight and length or stature measurements were expressed as height-for-age z-scores, weight-for-height z-scores, and changes in weight-for-age z-scores, using the WHO/NCHS international reference standards [21] (e.g., the height-for-age z-score is the height of the child compared with the median and standard deviation of the international reference height for the child's age). Z-scores were calculated by the US Centers for Disease Control Anthropometric Software Package (CASP). Height-for-age z-score was used as an indicator of stunting and weight-for-height z-score as an indicator of wasting. The cutoff value for wasting and stunting was –2 SD. Children's weights were measured at baseline and approximately 4 weeks later; weight gains were adjusted to 30 days and expressed as changes in weight-for-age z-score.

A 5-g stool sample was collected from each child, and 1 g was preserved in 10 mL of MIF (merthiolate-iodine-formaldehyde) solution [22]. Preserved samples were examined by the US Embassy Clinic laboratory in Bamako directly and concentrated by light microscope for *Ascaris lumbricoides, Trichuris trichura*, hookworm, *Giardia*, and *Entamoeba histolytica.*

Statistical analysis

The hypothesized associations between putative risk factors (maternal time use, maternal food production, household and maternal socioeconomic and demographic characteristics, child care, breastfeeding patterns, and children's parasitic infections) and the outcomes of interest (children's dietary intake and anthropometry) were evaluated by the bivariate approach. Factors found to be significant in the bivariate associations were further analyzed in multivariate models to assess the significance of association of each factor while simultaneously controlling for the effect of other factors. Multivariate analyses were performed using the least-squares regression for continuous outcomes and logistic regression analysis for dichotomous outcomes. The level of statistical significance was evaluated at a significance of $\alpha = 0.10$. Energy intake analyses controlled for breastfeeding, and anthropometric analyses controlled for age.

The model for the hypothesis that maternal income-generating activities are positively associated with children's dietary intake and anthropometry, and for the hypothesis that maternal food production is positively associated with children's dietary intake and anthropometry, is

$$Y = \beta_0 + \beta_1 X_1 + \Sigma\gamma_i Z_i + \Sigma\delta_j Z_j + \varepsilon$$

where Y is energy intake, animal protein intake, weight-for-height z-score, or height-for-age z-score; X_1 is time spent by the mother in income-generating activities or production of rice, sorghum, peanuts, or fonio; Z_i are child-care variables; and Z_j are potentially confounding variables.

The model for the hypothesis that maternal income-generating activities are not associated with child care is

$$Y = \beta_0 + \Sigma\beta_i X_i + \varepsilon$$

where Y is time spent by the mother in income-generating activities and X_i are child-care variables.

Ethical considerations

The National Public Health Research Institute of Mali gave permission to conduct the study. The research proposal was reviewed and approved by the Cornell University Committee on Human Subjects. The study objectives, procedures, and confidentiality were orally explained in the local language to individual study subjects by a Malian village health worker who accompanied the first author to the field. Study subjects gave oral consent to participate in the study.

Results

Study population

The study population resided in villages ranging in size from 200 to 2,000, with a high rate of out-migration to Bamako and Côte d'Ivoire. Household size and composition reflected the traditional patrilocal, extended family of agnates, each having one or more wives and children (**table 1**). The mean number of household members was 22.4, the mean number of nuclear families in a household was 2.7, the mean number of women was 6.5, and the mean number of children was 10.8. The mean number of children per mother was 3.2 (**table 2**). For 88% of mothers the study child was her youngest child. Approximately 50% of mothers were in monogamous marriages and 50% were in polygamous unions. There were no female household heads or divorced women. Sixty-two percent of households had a latrine, and 43% used a modern well or pump for drinking water. Cattle were the most important livestock asset, followed by sheep, goats, and horses. Eighty-five percent of households had cattle; the average number of cattle per household was 5.4.

The crops cultivated by women on their own plots of land were rice (79% of mothers), peanuts (53% of mothers), sorghum (34% of mothers), and fonio. The size of women's fields was less than 1 hectare and the only farming tool used was a hoe. The majority of women worked alone. On a typical nonmarket day, between sunrise and sunset, the mothers of young children spent a mean of 5.3 hours doing domestic work, including child-care activities, and 2.9 hours in income-generating activities (**table 3**). On nonmarket days, women generated income by farming their own plots of land, gathering shea nuts, and making shea butter. Less than 10% of women worked in the family fields, and, with one exception, these women did not work on their own plots of land on the same day. Domestic work consisted of preparing food (pounding cereal; lighting the fire; cooking porridge, *toh*—a thick gruel of sorghum or corn flour—rice, couscous, or sauce; and serving food), collecting wood, fetching water, sweeping, washing dishes and clothes, and child care including breastfeeding. The amount of time women spent preparing food depended on the number of women in the compound and whose turn it was to cook for the extended family. In small households women had to prepare food every day, whereas in large households women prepared *toh* once or twice a week. Time spent fetching water was relatively short because wells or pumps were located close to homes.

Mothers spent on average less than half an hour a

TABLE 1. Socioeconomic and demographic characteristics of Bambara households, Mali (n = 53)

Characteristic	Value
Composition (mean + SD no. per household)	
All members	22.4 + 14.8
Nuclear families	2.7 + 2.0
Men	4.6 + 3.5
Women	6.5 + 4.8
Children	10.8 + 7.4
Possessions (% of households)	
Latrine	62
Modern well or pump	43
Cattle	85
Sheep	70
Goats	60
Horse(s)	49
Parental occupation and education (% of parents)	
Father has off-farm income	40
Mother is a trader	81
Father went to school	21
Mother went to school	4

TABLE 2. Demographic characteristics and food production of Bambara mothers, Mali (n = 53)

Characteristic	Value
Demographic	
No. of children—mean + SD	3.2 + 1.4
Age (yr)—mean + SD	30.1 + 7.1
Monogamous marriage—%	47
Polygamous marriage—%	53
Food production—% of mothers	
Rice	79
Peanuts	53
Fonio	57
Sorghum	34

TABLE 3. Daily use of time by Bambara women on nonmarket days, Mali (n = 65)

Activity	% of women	Mean + SD hours
Income-generating activities		
Farming (women's field)	62	2.0 + 1.9
Gathering shea nuts and making shea butter	43	0.9 + 1.5
Domestic work		
Preparing food	100	2.9 + 1.5
Housework	94	0.8 + 0.6
Fetching water	77	0.5 + 0.5
Collecting wood	25	0.3 + 0.6
Child care		
Breastfeeding	74	0.5 + 0.3
Feeding, bathing, playing, comforting	86	0.3 + 0.3

day in child-care activities, excluding breastfeeding, and approximately half an hour a day breastfeeding (**table 3**). They were observed bathing, feeding, playing with, and comforting their children. Supervision of the child's feeding by the mother was observed for 43% of the 12- to 23-month-old children and 41% of the 24- to 35-month-old children. Caregiving activities and breastfeeding were highly integrated into maternal domestic work and income-generating activities. For example, mothers took their babies to the fields and breastfed them there. The caregiver of 45.7% of children 12 to 23 months of age was an older sibling, whereas for 45.7% of children the caregiver was the mother and for 8.6% the grandmother. The caregiver of 72.4% of children 24 to 35 months of age was an older sibling, whereas for 10.3% the caregiver was the mother and for 17.2% the grandmother.

The breastfeeding pattern according to age showed that all children under 21 months of age were breastfed and that all children 30 months of age or older were not breastfed. Between 21 and 29 months of age, there were both breastfed and nonbreastfed children. Nine foods were consumed by more than 20% of children: locust bean, shea butter, baobab leaves, dried fish, sorghum, corn, shea nuts, cow's milk, and mango (**table 4**). The mean number of different food items consumed by children the previous day was five. Cow's milk provided most of the children's animal protein intake. The same foods and frequencies were observed for breastfed children and nonbreastfed children. The major dishes and foods were *toh*; couscous or steamed fonio or corn cereal, sometimes cooked with milk; *serri* or porridge (a thin gruel of sorghum or corn meal), prepared sometimes with milk and shea butter; and *na* or sauce made with a variety of ingredients, including dried locust bean, fresh or dried baobab leaves, peanut paste, dried mushrooms, and dried catfish. The youngest children were breastfed and ate two meals a day, skipping breakfast or the evening meal. Older children, both breastfed and nonbreastfed, ate three meals a day, with or without a snack. The prevalence of stunting (chronic malnutrition or low height-for-age) was 29% and the prevalence of wasting (acute malnutrition or low weight-for-height) was 17%.

TABLE 4. Frequency of consumption of foods consumed by more than 20% of Bambara children 12 to 36 months of age, Mali ($n = 45$)

Food	% of children
Locust beans	100
Shea butter	87
Baobab leaves	80
Dried fish	60
Sorghum	53
Corn	47
Shea nuts	40
Cow's milk	27
Mango	22

The percentage of children's stools positive for *Giardia* was 17%, and the percentage of stools positive for *E. histolytica* was 5%. No helminths were found in the children's stools. Less than half of children infected with *Giardia* presented with diarrhea.

Determinants of children's dietary intake and anthropometry

Maternal income-generating activities and per capita livestock assets were negatively associated with children's height-for-age z-score (**table 5**). Household size was positively associated with children's height-for-age z-score. Maternal income-generating activities were

TABLE 5. Determinants of child nutrient intake and anthropometry—bivariate analyses, Bambara children 12 to 36 months of age, Mali ($p < .10$)

Outcome variable	Determinant	β	r^2
HAZ	Maternal income-generating activities	– .105	.048
	Household per capita livestock assets	– .017	.105
	Household size	.025	.091
Animal protein intake (< or > 10 g/d)	Maternal income-generating activities	– .535	.151
	Household per capita livestock assets	.042	.093
	Maternal supervision of child feeding	1.26	.065
WHZ	Maternal peanut production	.723	.094
	Time spent breastfeeding	– .845	.057
Energy intake (kcal/kg/d)	Maternal sorghum production	– 18.0	.067
	Maternal schooling	37.4	.100
Weight gain change in WAZ during 1 mo	*Giardia* infection	—[a]	

HAZ, height-for-age z-score; WHZ, weight-for-height z-score; WAZ, weight-for-age z-score

a. The difference in mean monthly weight gain between *Giardia*-infected and -noninfected children is significant ($p < .05$, *t*-test; $n = 49$).

also negatively associated with children's animal protein intake; per capita livestock assets were positively associated with animal protein intake. The regression analyses of maternal income-generating activities and child-care variables showed that none of the child-care variables were significantly related to maternal income-generating activities.

Of the caregiving variables, maternal supervision of feeding was positively associated with animal protein intake and time spent breastfeeding was negatively associated with children's weight-for-height z-score. Time spent by the mother taking care of her child and nonparental caregiving were not associated with any dietary or anthropometric indicator. Breastfeeding and breastfeeding frequency were not associated with any anthropometric indicator.

Of the maternal food-production variables, maternal production of peanuts, a cash crop, was positively associated with children's weight-for-height z-score. In contrast, maternal production of sorghum, a staple food crop, was negatively associated with children's energy intake after controlling for breastfeeding. Maternal schooling was also positively associated with energy intake. Maternal production of rice and fonio was not associated with any child dietary or anthropometric variable.

Giardia infection as well as animal protein intake was significantly associated with children's weight gain over a 4-week period.

Multivariate analysis

Maternal income-generating activities were negatively associated with children's height-for-age z-score after controlling for household size, per capita livestock assets, and child's age as confounding factors, and explained 34% of the variance in child height-for-age z-score (**table 6**). The estimated coefficient for the regression of child height-for-age on maternal income-generating activities predicts that an increase of 1 hour in time spent on maternal income-generating activities is associated with a decrease of 0.1 height-for-age z-score. Household size was positively associated with child height-for-age z-score and, as a proxy for socioeconomic status, did not change the negative association between maternal income-generating activities and height-for-age z-score.

Maternal income-generating activities were negatively associated with children's animal protein intake after controlling for per capita livestock assets and the presence of the mother during feeding, whereas maternal supervision of feeding was positively associated with children's animal protein intake. The logistic regression of children's animal protein intake on maternal income-generating activities indicates that an increase of 1 hour in time spent on maternal income-generating activities decreases by half the odds of the child's having an animal protein intake more than 10 g ($e^{-6.62}$ = 0.5 or odds ratio = 0.5). The logistic regression of children's animal protein intake on maternal supervision of feeding indicates that the odds of the child's having an animal protein intake more than 10 g was 3.5 times greater among children whose mothers were present during feeding ($e^{1.26}$ = 3.5 or odds ratio = 3.5) (**table 5**).

Maternal peanut production was positively associated with children's weight-for-height z-score after controlling for child's age and explained 32% of the variance in children's weight-for-height z-score; the estimated regression coefficient predicts that maternal peanut production is associated with an increase of 0.6 in weight-for-height z-score. On the other hand, maternal sorghum production was negatively associated with children's energy intake after controlling for breastfeeding; the estimated regression coefficient predicts that maternal sorghum production is associated with a decrease in energy intake of 20 kcal per kilogram per day. The households of women who cultivated sorghum consumed corn rather than sorghum, which is the preferred staple cereal ($p < .01$). Maternal sorghum production as well as time spent in own-account farming was associated with large household size and traditional household composition.

Discussion

This study is one of the few in sub-Saharan Africa to examine the interrelationships among maternal work, child care, and child nutrition. In spite of limited

TABLE 6. Determinants of child nutrient intake and anthropometry—multivariate analyses, Bambara children 12 to 36 months of age, Mali ($p < .05$)

Outcome variable	Determinant	β	r^2	Confounding variables
HAZ	Maternal income-generating activities	– .125	.340	Per capita livestock assets, household size, child's age
Animal protein intake (< or > 10 g/d)	Maternal income-generating activities	– .662	.360	Per capita livestock assets, maternal supervision of feeding
WHZ	Maternal peanut production	– .552	.321	Child's age
Energy intake (kcal/kg/d)	Maternal sorghum production	– 19.55	.178	Breastfeeding

HAZ, height-for-age z-score; WHZ, weight-for-height z-score access to land and assets by Bambara women in Mali, maternal control over income and the capacity to cultivate peanuts, a cash crop, had a beneficial effect on child nutrition. The strong positive association between own-account maternal cultivation of a cash crop and children's weight-for-height z-scores is an important finding in light of the high prevalence of wasting (17%) in this age group.

The negative associations between maternal income-generating activities and children's height-for-age z-score and animal protein intake and between maternal sorghum production and children's energy intake were not in the expected direction. These negative relationships were not mediated by the amount of time mothers devoted to breastfeeding or taking care of their children or child-care practices; they appear to be explained by sociocultural factors (traditional household size) and/or household economic factors (consumption of corn instead of the preferred staple, sorghum). The multivariate analysis nevertheless controlled for household socioeconomic status.

Maternal food production during the rainy season was related to the short-term nutritional status of the child (wasting, energy intake), whereas time spent by the mother in income-generating activities was weakly related to the long-term nutritional status of the child (stunting). This suggests that the latter represented overall work activity during the planting season and other seasons of the year, such as the harvest season and the dry season.

The lack of association between maternal income-generating activities and child-care activities is plausible because of the limited amount of time mothers devoted to child care and the cultural norms of caregiving by siblings and grandmothers and prolonged breastfeeding. Child care was characterized by breastfeeding until about 24 months of age, limited time devoted to child-care activities, and caregiving by siblings in the context of extended families.

In spite of the limited time spent on child-care activities overall, maternal supervision of feeding was strongly and positively associated with children's animal protein intake, supporting the hypothesis that the degree of parental control of feeding or responsiveness during feeding is a determinant of children's food intake and growth [23, 24]. The study findings suggest that maternal supervision of child feeding can more than offset the negative effect of maternal income-generating activities on children's animal protein intake. Depending on the cultural context, child-care patterns as well as household economic factors may enhance or limit the consumption of animal food sources [25].

The findings on women farmers from Mali show that some maternal income-generating activities are positively related to child nutrition, some are negatively related, and some have no association; that the relationships depend on mediating factors; and that negative findings do not necessarily imply that caregiving is less adequate because the mother is farming or trading. Measurement of the mix and kind of income-generating activities, information on maternal control over income, and an in-depth understanding of child-care practices as well as the cultural context are needed to elucidate the effects of maternal food production and trading on child care and child nutrition in Africa.

Acknowledgments

Intercooperation, Bern, Switzerland, and Save the Children, Westport, Connecticut, USA, provided support for the fieldwork. The United States Embassy clinic laboratory in Bamako assisted with the examinations for parasitic infections.

References

1. Sanjur D. Hispanic foodways, nutrition and health. Needham Heights, MA, USA: Allyn and Bacon, 1995.
2. Leslie J. Women's work and child nutrition in the Third World. Washington, DC: International Center for Research on Women, 1985.
3. Wandel M, Holmboe-Ottesen G. Women as nutrition mediators: A case study from Sri Lanka. Ecol Food Nutr 1988;21:117–30.
4. Tucker K, Sanjur D. Maternal employment and child nutrition in Panama. Soc Sci Med 1988;26:605–12.
5. Rathnayake IM, Weerahewa J. Maternal employment and income affect dietary caloric adequacy in households in Sri Lanka. Food Nutr Bull 2005;26:222–9.
6. Lamontagne JF, Engle PL, Zeitlin MF. Maternal employment, child care, and nutritional status of 12–18-month-old children in Managua, Nicaragua. Soc Sci Med 1998;46:403–14.
7. Popkin BM. Rural women, work, and child welfare in the Philippines. In: Buvinic M, Lycette MA, McGreevey WP, eds. Women and poverty in the Third World. Baltimore, MD, USA: Johns Hopkins University Press, 1983.
8. Wandel M, Holmboe-Ottesen G. Maternal work, child feeding, and nutrition in rural Tanzania. Food Nutr Bull 1992;14:49–54.
9. Bégin F, Frongillo EA Jr, Delisle H. Caregiver behaviors and resources influence child height-for-age in rural Chad. J Nutr 1999;129:680–6.
10. Oshaug A, Haddad L. Nutrition and agriculture. In: Nutrition: A foundation for development. Geneva: United Nations Standing Committee on Nutrition, 2002.
11. Savané MA. The effect of social and economic changes on the role and status of women in Sub-Saharan Africa. In: Moock JL, ed. Understanding Africa's rural households and farming systems. Boulder, CO, USA: West-

view Press, 1986.
12. Zuidberg L, Djire T. Les paysannes du Mali-Sud. Institut Royal des Tropiques Bulletin 1992:326.
13. Belloncle G. Femmes et développement en Afrique Sahélienne. Dakar, Senegal: Nouvelles Editions Africaines, 1980.
14. Dwyer D, Bruce J, eds. A home divided: Women and income in the Third World. Stanford, CA, USA: Stanford University Press, 1988.
15. Engle PL, Menon P, Haddad L. Care and nutrition concepts and measurement. Washington, DC: International Food Policy Research Institute, 1997.
16. Dettwyler KA. More than nutrition: Breastfeeding in urban Mali. Med Anthropol Q 1988;2:172–83.
17. Erny P. L'Enfant et son milieu en Afrique noire. Paris: L'Harmattan, 1987.
18. Cissé B, Katz K. Accroître l'accès aux services du planning familial au Mali rural. Raleigh, NC, USA: Family Health International, 1992.
19. UNICEF. State of the world's children 2005. New York: UNICEF, 2004.
20. Save the Children. Focus on Mali. School Health Nutr Newsl 2005;2:5–6.
21. World Health Organization. WHO Child Growth Standards and NCHS International Reference Data page. Available at: http://www.who.int/nutgrowthdb/reference/en/. Accessed 10 December 2006.
22. Beaver PC, Jung RC, Cupp EW. Clinical parasitology. Philadelphia, PA, USA: Lea and Febiger, 1984.
23. Pan American Health Organization/World Health Organization. Guiding principles for complementary feeding of the breastfed child. Washington, DC: Pan American Health Organization, 2003, p. 14.
24. Ruel MT, Arimond M. Measuring childcare practices. Approaches, indicators, and implications for programs. Food Policy Review No. 6. Washington, DC: International Food Policy Research Institute, 2003.
25. Gittelsohn J, Vastine A. Sociocultural and household factors impacting on the selection, allocation and consumption of animal source foods: Current knowledge and application. J Nutr 2003;133(11 suppl 2): 4036S–41S.

Risk factors for moderate to severe anemia among children in Benin and Mali: Insights from a multilevel analysis

Ismael Ngnie-Teta, Olivier Receveur, and Barthelemy Kuate-Defo

Abstract

Background. *Anemia currently affects 2 billion people throughout the world. Although the immediate causes of anemia among children are known (including malnutrition and infections), the importance of contextual determinants and their relationships with individual effects have rarely been explored.*

Objective. *To identify anemia risk factors at the individual, household, and community levels among Beninese and Malian children, using simple and multilevel regression methods.*

Methods. *An analysis was undertaken of nationally representative data collected in 2001 in Benin (*n *= 2,284) and Mali (*n *= 2,826) by the Demographic and Health Surveys. Sixteen potential risk factors for anemia were considered at the individual, household, and community levels. Comparative analyses were carried out using simple and multilevel logistic regression models.*

Results. *Simple and multilevel logistic regression analyses yielded broadly similar results. Risk factors for moderate to severe anemia included incomplete immunization, stunted growth, recent infection, absence of bednet, low household living standard, rural residency (Mali), low maternal education, and low community development index (Benin). In addition, multilevel analysis indicated a clustering level of anemia in communities (intraclass correlation) of 14% and 19% in Benin and Mali, respectively.*

Conclusions. *Risk factors for child anemia appeared at all three levels (individual, household and community). Community-level clustering seemed to be low. Therefore, interventions to address anemia need not be village- or region-specific. Identifying a successful and replicable program is now a priority in child survival endeavors. It is likely that such a program would include a focus on improving immunization coverage, increased bednet usage, and reduced protein–energy malnutrition.*

Key words: Anemia, Benin, children, demographic and health surveys, Mali, multilevel analysis

Introduction

Anemia represents a major public health problem among infants in poor countries. According to the most recent estimates of the World Health Organization (WHO), two billion people throughout the world are anemic, including 250 million children (12%) [1]. Moderate and severe anemia are associated with increased child morbidity and mortality [2] and a deterioration of the physical and cognitive development of children [3–7].

There are a variety of causes of anemia, related to both individual and environmental factors. Iron deficiency remains the main cause of anemia in poor countries and contributes to almost half of the current cases [8]. Other nutritional deficiencies (folate, vitamin B_{12}, vitamin A) are also risk factors for anemia [1, 9, 10], as are diets rich in phytates [11, 12] and poor in animal products [13]. In addition, diseases, and malaria in particular, are important risk factors: in endemic malarial settings typical of Africa, there is a much higher prevalence of anemia than there is in nonendemic areas [14].

Although the immediate causes of anemia among children are known (such as malnutrition and infections), the impact of household and community socioeconomic determinants is only now being explored, and the interrelationship between such contextual and

Ismael Ngnie-Teta is affiliated with PATH Canada, Ottawa; Olivier Receveur is affiliated with the Département de Nutrition, Faculté de Médecine, Université de Montréal, Montreal, Canada; Barthelemy Kuate-Defo is affiliated with the Département de Démographie, Université de Montréal, Montreal, Canada.

Please direct queries to the corresponding author: Ismael Ngnie-Teta, Nutrition Project Officer, PATH Canada, Suite 1105, One Nicholas Street, Ottawa, Ontario K1N 7B7, Canada; e-mail: iteta@healthbridge.ca.

These data were presented at the 2005 International Union of Nutritional Sciences congress in Durban, South Africa.

individual factors remains under-studied. Using multilevel analysis techniques, it is possible to identify and separate contextual-level effects from individual-level effects [15–20]; however, multilevel analysis has been rarely used by nutritional epidemiologists.

Using multivariate and multilevel approaches, this study attempted to separate individual and household factors from contextual factors associated with moderate and severe anemia in children in Benin and Mali. The choice of countries was based on anemia data availability in Demographic and Health Survey (DHS) data; in 2002 when the study started, data on anemia were available from only three sub-Saharan African countries: Benin, Mali, and Uganda. The Uganda database was temporarily withdrawn from the ORC Macro data bank (the organization in charge of DHS data administration) and consequently withdrawn from our study.

Methods

Data and study population

Data used in this study were gathered from recent DHS carried out in 2001 in Benin and Mali. These DHS are nationally representative surveys including 2,284 Beninese children and 2,826 Malian children aged 6 to 59 months. The sample design involved a probabilistic two-stage sampling. In the first stage, enumeration areas were randomly selected with equal probability. The second stage involved interviewing of eligible households and individual respondents. A detailed sampling methodology is presented in the DHS sampling manual [21] and DHS country reports [22, 23].

The surveys consisted of individual and household questionnaires that identified women of reproductive age (15 to 49 years). The household questionnaire collected comparable information on household composition, including relationship to head, age and sex of all the household members, and educational attainment for those aged 6 years and higher. The individual questionnaire included information on the background characteristics of survey respondents. Child health and nutrition information was collected for all children born to women during the 5-year period preceding the survey.

Ethical clearance and informed consent

Training for the EDSM-III and EDSB-III survey teams included special training for the field editors and the team leaders on how to present the informed consent statement. The informed consent was approved by the ORC Macro research ethics committee as well as the National Ethics Committee for Mali (Bamako) and for Benin (Cotonou). The questionnaire was translated into the six main Beninese languages (Adja, Dariba, Dendi, Fon, Yoruba, and Ditamari) and the three main Malian languages (Bambara, Sonrai, and Peuhl). Prior to the test, each woman was asked whether she agreed to participate in the study and, if so, to sign a form giving her permission for the collection of a blood droplet from herself and her children. The form explained the hemoglobin testing procedure and the causes and consequences of anemia. The women were assured that the results obtained from the testing would be kept confidential.

Study variables

Outcome: child anemia

Anemia was defined through blood hemoglobin levels in capillary blood. Hemoglobin measurement was undertaken in Benin and Mali by the HemoCue system, which consists of a portable photometer and a one-step blood collection device that is covered with dry hemoglobin conversion reagents. This is convenient for population-based surveys. The anemia cutoff points used in this study were those recommended by WHO for children [24]. Moderate to severe anemia was defined as hemoglobin lower than 9.9 g/dL and mild anemia as hemoglobin between 10.0 and 10.9 g/dL.

Explanatory variables

Potential risk factors for anemia are presented in **figure 1**, which reflects the rationale for the multilevel analysis carried out in this research. Variables were

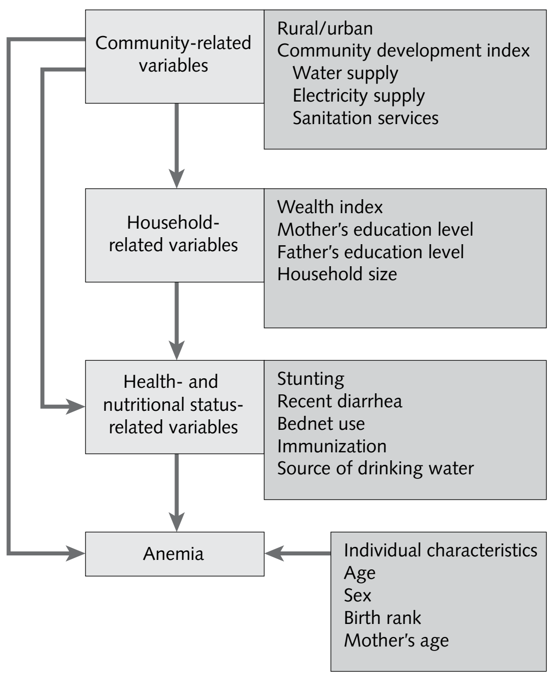

FIG. 1. Simplified conceptual framework for sociodemographic determinants of anemia among children

defined hierarchically, notably at the individual, household, and community levels.

The following child characteristics were studied: age (6–11 months, 12–35 months, 36–59 months); sex (male/female); birth rank (1, 2–3, 4–5, 6+); mother's age (15–29 years, 30–49 years); height-for-age z-score (HAZ < –2 / HAZ ≥ -2); recent diarrhea episodes (yes/no); and immunization (full/incomplete).

Studied household characteristics included: source of drinking water (safe/unsafe); mother's education (no education/any education); father's education (no education/any education); household size (< 5 persons, 5 to 7 persons, >7 persons), and household wealth index (low, medium, high). The wealth index is a composite measure of the cumulative living standard of a household. It is calculated using easy-to-collect data on a household's ownership of selected assets, such as televisions and bicycles, materials used for housing construction, and types of water access and sanitation facilities. The most important household-owned durable goods were identified and placed on a continuous scale by principal-component analysis [25]. We categorized in tertiles corresponding to low, medium, and high. The wealth index is particularly valuable in countries that lack reliable data on income and expenditures, the conventional indicators used to measure household economic status. These income and expenditure measures, when available, are often unreliable, and even in countries that collect these data, the DHS wealth index has outperformed the traditional indicators [25].

Communities were defined by grouping sampling clusters within administrative units. Community-related variables are the place of residence (urban/rural) and the community development index (low, medium, high). This community development index was based on the availability of three basic services in the community: water supply, electricity supply, and sanitation services. We considered communities with none of the three services as having a low development index, those with one or two services as having a medium development index, and those with all three services as having a high development index. This index could be constructed for Benin only because data were not collected for Mali.

Analytical procedures

For each country, we applied six simple (or single-level) logistic regression models and six multilevel logistic regression models to the data. Child, household, and community characteristics were entered progressively into a simple logistic regression model to examine their associations with moderate to severe anemia. These analyses were then repeated using multilevel logistic regression model techniques [17, 18, 20]. The single-level logistic regression model fitted a single-level regression to individual, household, and community characteristics and did not take account of the household- and community-level clustering of children. The multilevel logistic regression model allowed the incorporation of explanatory variables at different levels of the hierarchy. It also produced estimates of logistic regression coefficients, standard errors, confidence intervals, and significance tests that are generally more conservative than those obtained from simple logistic regression models [20, 26–29]. The simple logistic regression models were fitted using SPSS v13.0. Multilevel logistic regression models were fitted using MLwiN v2.0 [30].

Model specifications

We constructed six models for each country. The first model (basic model) contained biological risk factors such as child's age, child's sex, child's birth rank, and mother's age. The second model (health model) contained health-related variables, notably stunting, recent episodes of diarrhea, source of drinking water, bednet usage, and completion of immunization. The third model (socioeconomic model) examined household socioeconomic variables, namely, mother's and father's education levels, household size, and household wealth index. The fourth model (community model) contained community-level variables, including type of place of residence and community development index. The fifth model (socio-community model) combined the socioeconomic and community models. The sixth model (full model) included all of the studied variables.

Fixed effects

Fixed estimates presented in the results section are those of the full model. For the two analytical methods, estimates of association between moderate to severe anemia and explanatory variables are presented side by side in the same table to facilitate comparison. The β coefficients (standard errors) were converted into odds ratios (95% confidence intervals).

Random effects

In this study, multilevel logistic regression allowed us to examine the role of contextual (community) factors in the prevalence of child anemia. Possible contextual effects were measured by the variance partition coefficient (VPC); this is a variant of intraclass correlation (ICC) when the outcome is nonlinear [15, 20, 26, 30]. For a dichotomous variable such as presence or absence of moderate to severe anemia, VPC was calculated according to the formula used by Snijders and Bosker [26]:

$$VPC = V_n / (V_n + \pi^2/3)$$

$$V_n = \text{neighbourhood variance}$$

VPC represented the percentage of total variance in the risk of moderate to severe anemia attributable to the neighborhood level and was also used as a measure of clustering of anemia in communities. A high VPC would reflect a high clustering of anemia prevalence and a high neighborhood effect on individual risk of anemia. In model 4, we tested cross-level interaction by letting the level 1 explanatory variables slope be random at the neighborhood level. Multilevel logistic parameters were estimated using restricted iterative generalized least squares (RIGLS) and penalized quasi likelihood (PQL) (Goldstein, MLWIN manual) [30].

Results

Prevalence of anemia

The prevalence of anemia among children was high and comparable in the two countries: (82% and 83% in Benin and Mali, respectively). Moderate anemia (hemoglobin between 7.0 and 9.9 g/dL) affected 52% of Beninese children and 53% of Malian children. Severe anemia (hemoglobin < 7.0 g/dL) affected 9% of Beninese children and 12% of Malian children. The prevalence of moderate to severe anemia varied enormously from one region to another within countries. In Benin, the lowest prevalence was observed in the southern region of Atlantique (42%), while the highest prevalence was observed in the northern region of Atacora (74%). In Mali, the lowest prevalence of moderate to severe anemia was seen in Bamako, the capital (48%). Apart from Bamako and Segou (close to Bamako), the prevalence of moderate to severe anemia was more than 60% in the seven other regions of Mali. Bivariate analyses show that only a few explanatory variables are significantly associated with mild anemia. On other hand, almost the same variables are associated with moderate and severe anemia (**tables 1 and 2**). In this

TABLE 1. Bivariate association between potential risk factors and varying levels of anemia in Benin

Variable		*n*	Severe (%) 191	Moderate (%) 1,149	Mild (%) 495	Normal (%) 481
Control variables						
Child's age (mo)						
6–11		273	15.8	53.8	19.4	11.0
12–35		933	11.8	55.7	17.9	14.6
36–59		846	3.4	46.6	26.5	23.5
	p^a		< .001	< .001	.368	Ref
Child's sex						
Male		1,039	9.2	52.3	22.6	15.9
Female		1,013	8.5	51.1	20.6	19.7
	p		.096	.049	.029	Ref
Birth order						
1		414	6.5	49.0	21.7	22.7
2 or 3		654	8.0	50.0	22.2	19.9
4 or 5		481	11.0	52.6	20.6	15.8
6+		503	9.9	55.3	21.9	12.9
	p		.002	.002	.050	Ref
Mother's age (yr)						
15–29		1,116	9.3	52.6	21.1	17.0
30–49		936	8.3	50.6	22.3	18.7
	p		.832	.256	.129	Ref
Health-related variables						
HAZ						
Normal (≥ −2)		1,145	9.1	47.4	22.8	20.7
Stunted (< −2)		597	8.7	58.0	20.0	12.9
	p		.010	< .001	.070	Ref
Recent diarrhea						
No		1,719	8.3	50.0	22.5	19.1
Yes		325	12.0	60.6	16.9	10.5
	p		< .001	< .001	.166	Ref

continued

TABLE 1. Bivariate association between potential risk factors and varying levels of anemia in Benin *(continued)*

Variable		*n*	Severe (%) 191	Moderate (%) 1,149	Mild (%) 495	Normal (%) 481
Immunization						
No vaccine		168	11.3	60.1	15.5	13.1
1–4 vaccines		228	12.3	54.4	20.6	12.7
5–7 vaccines		282	12.4	54.6	16.3	16.7
Full immunization		1,292	7.3	49.4	23.8	19.5
	p		< .001	.010	.457	Ref
Bednet use						
No		1,153	10.1	55.7	21.1	13.1
Yes		892	7.3	46.6	22.3	23.8
	p		< .001	< .001	< .001	Ref
Source of drinking water						
Unprotected		1,134	7.6	49.7	22.7	20.0
Protected		857	10.6	54.8	20.1	14.5
	p		< .001	< .001	.006	Ref
Household-related variables						
Mother's education level						
No education		1,534	9.5	53.7	22.4	14.5
Any education		518	7.1	45.9	19.3	27.6
	p		< .001	< .001	< .001	Ref
Father's educational level						
No education		1,071	9.8	54.4	21.3	14.5
Any education		819	6.6	49.3	21.2	22.8
	p		< .001	< .001	.002	Ref
No. of household members						
< 5		454	8.8	44.3	24.2	25.5
5–7		765	8.4	53.2	20.9	20.3
> 7		833	9.4	54.4	20.4	18.6
	p		.1	< .001	.4	Ref
Wealth index						
Low		517	10.1	58.2	18.2	13.5
Medium		856	10.7	51.6	24.4	13.2
High		679	5.6	46.8	20.8	26.8
	p		< .001	< .001	< .001	Ref
Community-related variables						
Region						
Atacora		332	15.7	59.0	14.2	11.1
Atlantique		392	5.1	39.8	23.2	31.9
Borgou		355	12.1	56.1	19.4	12.4
Momo		289	3.8	55.4	21.1	19.7
Ouémé		326	9.5	50.9	25.8	13.8
Zou		358	7.0	51.4	25.7	15.9
	p		< .001	< .001	< .001	Ref
Place of residence						
Urban		632	6.8	49.1	19.5	24.7
Rural		1,420	9.8	52.9	22.6	14.7
	p		< .001	< .001	< .001	Ref

continued

TABLE 1. Bivariate association between potential risk factors and varying levels of anemia in Benin *(continued)*

Variable		*n*	Severe (%)	Moderate (%)	Mild (%)	Normal (%)
			191	1,149	495	481
Community development index						
Null		1,079	9.7	53.1	22.2	14.9
Low		309	12.0	52.8	23.0	12.3
Medium		282	4.3	51.4	18.8	25.5
High		187	4.8	38.5	23.5	33.2
	p		< .001	< .001	< .001	Ref

HAZ, height-for-age z-score

a. The chi-square test was used to compare the distribution of each variable for each level of anemia with that of the nonanemic group.

TABLE 2. Bivariate association between potential risk factors and varying levels of anemia in Mali

Variable		*n*	Severe (%)	Moderate (%)	Mild (%)	Normal (%)
			301	1,297	452	412
Control variables						
Child's age (mo)						
6–11		334	9.0	55.4	18.0	17.7
12–35		1,105	17.6	57.6	12.9	11.9
36–59		1,023	7.4	46.5	24.4	21.6
	p[a]		< .001	< .001	0.857	Ref
Child's sex						
Male		1,251	11.8	54.0	16.6	17.7
Female		1,211	12.7	51.4	20.1	15.8
	p		.225	.306	.029	Ref
Birth order						
1		356	11.0	52.2	18.8	18.0
2 or 3		750	10.7	52.0	18.1	19.2
4 or 5		585	14.9	53.7	17.6	13.8
6+		771	12.3	52.8	18.9	16.0
	p		.011	.126	.384	Ref
Mother's age (yr)						
15–29		1,321	11.9	53.7	17.4	17.0
30–49		1,141	12.6	51.5	19.5	16.4
	p		.283	.515	.152	Ref
Health-related variables						
HAZ						
Normal (≥ -2)		1,401	8.6	51.3	20.0	20.1
Stunted (< -2)		995	18.0	54.7	16.1	11.3
	p		< .001	< .001	.009	Ref
Recent diarrhea						
No		1,893	10.3	51.8	19.9	18.1
Yes		559	18.4	56.2	13.1	12.3
	p		< .001	< .001	.453	Ref
Immunization						
No vaccine		515	13.4	54.0	18.1	14.6
1–4 vaccines		466	17.4	50.6	13.9	18.0
5–7 vaccines		573	10.5	52.5	20.1	16.9
Full immunization		864	9.8	52.9	20.0	17.2
	p		.024	.481	.127	Ref

continued

TABLE 2. Bivariate association between potential risk factors and varying levels of anemia in Mali *(continued)*

Variable		*n*	Severe (%) 301	Moderate (%) 1,297	Mild (%) 452	Normal (%) 412
Bednet use						
No		1,100	13.1	53.0	17.1	16.8
Yes		1,334	11.7	52.5	19.2	16.6
	p		.520	.276	.199	Ref
Source of drinking water						
Protected		1,012	11.5	53.6	18.0	17.0
Unprotected		1,403	12.4	52.4	18.9	16.3
	p		.247	.459	.284	Ref
Household-related variables						
Mother's educational level						
No education		2,092	12.8	53.3	18.2	15.8
Any education		370	8.9	49.5	19.5	22.2
	p		.001	.003	.076	Ref
Father's educational level						
No education		1,892	12.8	53.2	18.9	15.1
Any education		505	9.9	50.1	17.0	23.0
	p		< .001	< .001	.001	Ref
No. of household members						
< 5		513	11.9	50.1	19.3	18.7
5–7		945	12.9	53.7	17.9	15.6
> 7		1,004	11.8	53.1	18.3	16.8
	p		.376	.248	.834	Ref
Wealth index						
Low		821	13.8	54.7	17.7	13.9
Medium		826	14.2	54.4	17.9	13.6
High		815	8.7	49	19.5	22.8
	p		< .001	< .001	.011	Ref
Community-related variables						
Region						
Kayes		404	17.8	48.3	11.4	22.5
Koulikoro		404	19.6	53.0	18.6	8.9
Sikasso		532	10.9	57.7	19.2	12.2
Segou		371	5.4	50.1	22.9	21.6
Mopti		282	15.2	56.4	19.5	8.9
Tombouctou		109	11.0	57.8	15.6	15.6
Gao		91	6.6	57.1	13.2	23.1
Kidal		13	15.4	53.8	23.1	7.7
Bamako		256	3.5	44.5	22.3	29.7
	p		< .001	< .001	< .001	Ref
Place of residence						
Urban		478	6.7	48.1	20.3	24.9
Rural		1,984	13.6	53.8	17.9	14.8
	p		< .001	< .001	.007	Ref

HAZ, height-for-age z-score
a. The chi-square test was used to compare the distribution of each variable for each level of anemia with that of the nonanemic group.

paper, multivariate and multilevel analysis are based on moderate to severe anemia as outcome (hemoglobin < 9.9 g/dL).

Multivariate vs. multilevel estimates

Multivariate and multilevel estimates (odds ratio and confidence intervals) are presented in **tables 3 and 4** for Benin and Mali, respectively. To facilitate comparisons between single-level logistic regression and multilevel logistic regression, both estimates are presented side by side in each table; **figure 2** compares these estimates in Benin and Mali. The variables are presented following the hierarchical rationale of anemia determinants,

TABLE 3. Multivariate and multilevel modeling of child anemia in Benin

		Multilevel estimates		Multivariate estimates	
Variable	*n*	OR	95% CI	OR	95 % CI
Control variables					
Child's age (mo)					
6–11	67	4.05	2.40–7.69	3.85	2.17–6.82
12–35	212	2.81	1.99–4.52	2.78	1.94–3.99
36–59	285	1.00		1.00	
Child's sex					
Male	279	1.13	0.83–1.60	1.15	0.82–1.67
Female	285	1.00		1.00	
Birth order					
1	130	1.00		1.00	
2 or 3	199	0.99	0.61–1.66	1.01	0.66–1.49
4 or 5	120	1.17	0.50–1.95	1.04	0.58–2.03
6+	115	2.05	1.02–3.97	1.96	1.04–3.71
Mother's age (yr)					
15–29	306	1.41	0.87–2.12	1.33	0.85–2.07
30–49	258	1.00		1.00	
Health-related variables					
HAZ					
Normal (≥ −2)	402	1.00		1.00	
Stunted (< −2)	162	1.81	1.21–2.89	1.89	1.29–2.72
Recent diarrhea					
No	507			1.00	
Yes	57	2.34	1.44–4.23	2.43	1.38–4.28
Immunization					
Incomplete	136	1.49	1.11–2.33	1.61	1.09–2.38
Full	428	1.00		1.00	
Bednet use					
No	261	1.75	1.32–2.60	1.82	1.30–2.71
Yes	303	1.00		1.00	
Source of drinking water					
Protected	374	1.00		1.00	
Unprotected	190	1.24	0.90–2.07	1.37	0.92–2.08
Household-related variables					
Mother's educational level					
No education	381	1.77	1.28–2.99	1.84	1.20–2.81
Any education	183	1.00		1.00	
Father's educational level					
No education	275	0.82	0.46–1.07	0.68	0.45–1.03
Any education	289	1.00		1.00	

continued

TABLE 3. Multivariate and multilevel modeling of child anemia in Benin *(continued)*

Variable	*n*	Multilevel estimates OR	Multilevel estimates 95% CI	Multivariate estimates OR	Multivariate estimates 95 % CI
No. of household members					
< 5	153	1.00		1.00	
5–7	205	1.72	0.96–2.54	1.52	0.96–2.41
> 7	206	1.20	0.65–1.75	1.04	0.64–1.70
Wealth index					
Low	105	1.29	0.76–2.24	1.22	0.75–1.99
Medium	227	1.26	0.75–1.99	1.16	0.72–1.87
High	232	1.00		1.00	
Community-related variables					
Place of residence					
Urban	202	1.00		1.00	
Rural	362	1.01	0.34–1.35	0.49	0.10–2.38
Community development index					
Null	293	1.33	0.76–2.86	1.50	0.70–3.24
Low	82	2.71	1.02–4.73	2.41	1.12–5.47
Medium	106	1.13	0.73–2.32	1.37	0.72–2.63
High	83	1.00		1.00	
R^2					0.34
VPC (%)		7.82			

OR, odds ratio; CI, confidence interval; HAZ, height-for-age z-score; VPC, variance partition coefficient

TABLE 4. Multivariate and multilevel modeling of child anemia in Mali

Variable	*n*	Multilevel estimates OR	Multilevel estimates 95% CI	Multivariate estimates OR	Multivariate estimates 95% CI
Control variables					
Child's age (mo)					
6–11	106	1.73	1.32–2.92	1.87	1.25–2.80
12–35	244	2.90	2.24–3.92	2.86	2.13–3.84
36–59	424	1.00		1.00	
Child's sex					
Male	382	0.92	0.76–1.32	0.96	0.73–1.24
Female	392	1.00		1.00	
Birth order					
1	118	1.00		1.00	
2 or 3	247	0.92	0.64–1.42	0.91	0.60–1.39
4 or 5	165	1.37	0.94–2.53	1.36	0.81–2.27
6+	244	1.19	0.69–2.31	1.12	0.63–2.00
Mother's age (yr)					
15–29	408	1.25	0.86–2.02	1.25	0.84–1.85
30–49	366	1.00		1.00	
Health-related variables					
HAZ					
Normal (≥ -2)	250	1.00		1.00	
Stunted (< -2)	524	1.80	1.37–2.38	1.67	1.25–2.23

continued

TABLE 4. Multivariate and multilevel modeling of child anemia in Mali *(continued)*

Variable	*n*	Multilevel estimates		Multivariate estimates	
		OR	95% CI	OR	95% CI
Recent diarrhea					
No	653	1.00		1.00	
Yes	121	1.46	1.04–2.32	1.53	1.07–2.19
Immunization					
Incomplete	469	1.21	0.88–1.62	1.23	0.91–1.67
Full	305	1.00		1.00	
Bednet use					
No	345	0.94	0.66–1.76	0.96	0.70–1.30
Yes	429	1.00		1.00	
Source of drinking water					
Protected	325	1.00		1.00	
Unprotected	449	0.97	0.57–1.09	0.81	0.60–1.12
Household-related variables					
Mother's educational level					
No education	638	1.08	0.78–1.59	1.09	0.74–1.61
Any education	136	1.00		1.00	
Father's educational level					
No education	586	1.20	0.85–1.72	1.18	0.84–1.67
Any education	188	1.00		1.00	
No. of household members					
< 5	175	1.00		1.00	
5–7	294	1.51	0.98–2.29	1.46	0.99–2.14
> 7	305	1.40	0.92–2.33	1.45	0.95–2.21
Wealth index					
Low	236	1.34	0.90–2.30	1.49	0.98–2.27
Medium	231	1.73	1.06–2.35	1.85	1.24–2.76
High	307	1.00		1.00	
Community-related variables					
Place of residence					
Urban	198	1.00		1.00	
Rural	576	2.04	1.38–3.44	1.79	1.02–3.13
R^2				0.28	
VPC (%)		15.35			

OR, odds ratio; CI, confidence interval; HAZ, height-for-age z-score; VPC, variance partition coefficient

starting with the individual-level variables, followed by household- and community-level variables. The outcome is moderate to severe anemia (hemoglobin ≤ 9.9 g/dL).

In this study, single-level logistic regression yielded broadly similar results to multilevel logistic regression. The same risk factors for moderate to severe anemia were identified using the two methods, and estimates (odds ratios) were quite similar for all of the variables. The innovation of the multilevel analysis is that it made possible the quantification of the contribution of community-level effects to the prevalence of moderate to severe anemia.

Contribution of contextual effects to the prevalence of moderate to severe anemia

The contribution of community effects was derived from the null model, which is the model containing only the outcome and a constant (no explanatory variable). **Figure 3** shows that 14% of the variability in the risk of anemia in Benin and 19% in Mali is attributable to the difference between communities. This means

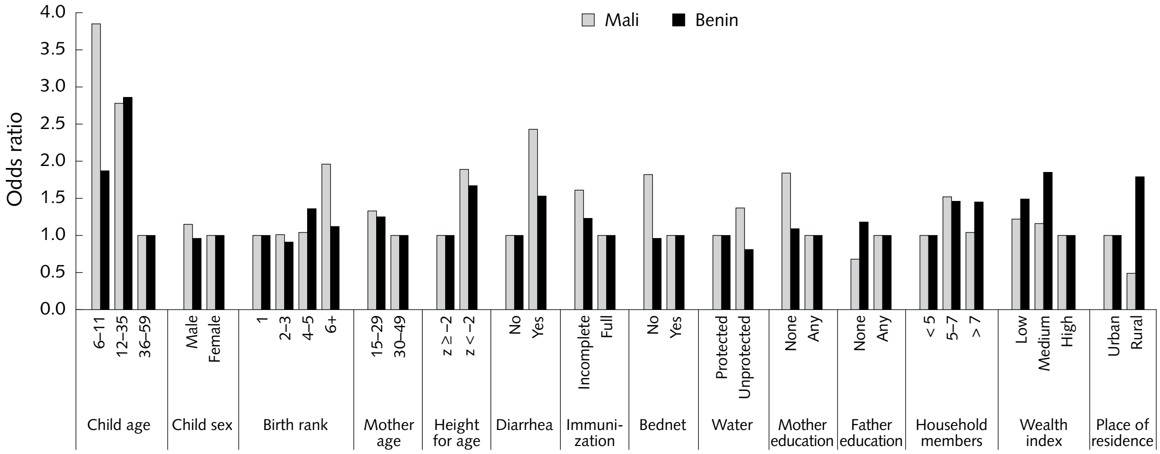

FIG. 2. Comparison of odds ratios of moderate-to-severe anemia among women in Benin and in Mali

that the prevalence of anemia is mostly attributable to differences between individuals (more than 80%) rather than to differences between communities.

Individual- and household-level factors

Age was strongly associated with the prevalence of moderate to severe anemia. The risk of anemia was 3 to 4 times higher among children under 3 years of age than among those between 4 and 5 years of age. In Benin, children with birth rank greater than five were twice as likely to have moderate to severe anemia as their older counterparts (OR = 2.05; 95%CI = 1.02–3.97). **Tables 2 and 3** also show that health-related variables were significantly associated with moderate to severe anemia in Benin and Mali. The risk of anemia was higher among stunted children (Benin: OR = 1.81; 95%CI = 1.21-2.89; Mali: OR = 1.80; 95%CI = 1.37–2.38) and children who have had recent episodes of diarrhea (Benin: OR = 2.34; 95%CI = 1.44–4.23; Mali: OR = 1.46; 1.04-2.32). Beninese children with incomplete immunization (Benin: OR = 1.49; 95%CI = 1.11–2.33) and children who do not sleep under a bednet (Benin: OR = 1.75; 95%CI = 1.32–2.60) are also at higher risk for moderate to severe anemia.

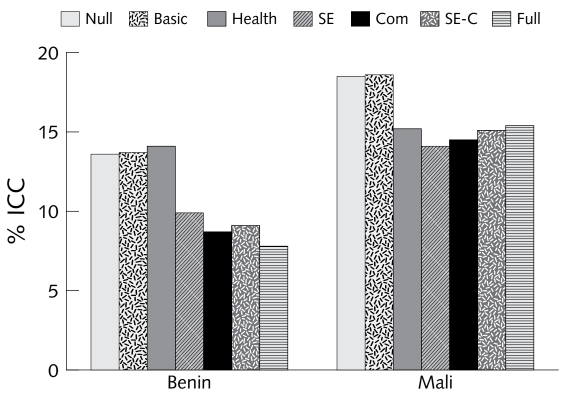

FIG. 3. Contribution of contextual effects to the prevalence of moderate to-severe anemia among Beninese and Malian children

ICC = Intra-class correlation ≈ Variance Partition Coefficient: Proportion of variability of risk of anemia that is attributable to community effects. Null = Null model. Basic = Basic model = age, sex, birth order, mother's age. Health = Health model = Basic + height-for-age + recent diarrhea + bednet use + immunization + source of drinking water. SE = Socioeconomic model = Basic + mother's education + father's education + household members + household living standard. Com = Community model = Basic + place of residence + community development index. SE-C = Socio-community model = Basic + SE + Com. Full = Full model = Basic + Health + SE + Com + SE-C

Mother's education was positively and significantly associated with moderate to severe anemia in Benin (OR = 1.77; 95%CI = 1.28–2.99) but not in Mali (OR = 1.08; 95%CI = 0.79–1.09). Father's education and household size were not associated with the prevalence of moderate to severe anemia. In Mali, children living in households with medium living standards had a lower risk of anemia than children living in households with either low living standards or high living standards.

Community-level factors

Living in a rural area was associated with a higher prevalence of anemia in Mali (OR = 2.04; 95%CI = 1.38–2.44) but not in Benin (OR = 1.01; 95%CI = 0.34–1.35). On the other hand, Beninese children living in communities with medium development levels were more at risk for moderate to severe anemia than their peers living in communities with either low or high development levels.

Discussion

This research, based on recent DHS undertaken in Benin and Mali, will undoubtedly contribute to a greater comprehension of the determinants of anemia at the individual, family, and community levels. The results of this study could be generalized to the population of children in the two countries and could help in planning anemia interventions in other African countries with comparable characteristics.

Single-level (simple) logistic regression model vs. multilevel logistic regression model

The aim of this study was to identify risk factors for moderate to severe anemia among children, using two analytical techniques: multilevel and single-level logistic regression. The results showed that the effects of individual and community characteristics on moderate to severe anemia were not affected by alternative specifications of the estimation methods. It is important to note that this was the first study comparing multilevel and simple regression for anemia risk factors using these DHS in the African context; more studies comparing the two methods are therefore needed to shed further light on the usefulness of multilevel analysis in nutritional epidemiology.

Risk factors for child anemia

Apart from this important methodologic result, the study reinforced current knowledge about infections and malnutrition as key determinants of anemia. Infections and health-seeking behaviors (characterized here by recent episodes of diarrhea, incomplete immunization, and absence of a bednet) were strongly associated with a high prevalence of moderate to severe anemia. Even after controlling for source of drinking water, diarrhea remains positively associated with anemia. Depending of the severity of diarrhea, it could lead to mild, moderate, or severe anemia [31]. Diarrheal illness is associated with loss of iron and decreased absorption of nutrients needed to maintain normal hemoglobin status [32]. It is also likely that, as demonstrated for other nutrient deficiencies, diarrhea shares common cause with anemia [33, 34].

Bednet use is well documented as an effective anemia prevention strategy [14, 35–42]. A recent study in Mali estimated that 30% of anemia is due to malaria [43]. An exhaustive review of the impact of malaria control on the risk of anemia among children [14] estimates the protective efficacy of bednet usage on severe anemia to be 60% (hemoglobin < 8 g/dL).

Stunting was associated with higher risk of anemia in the two countries. Previous studies in India and Kenya have shown that stunted children suffer from more severe anemia than nonstunted children [44, 45]. Even though it was not possible to determine in this study if stunting existed prior to anemia, the consistent association between these two nutritional conditions emphasizes the need for long-term efforts to optimize child nutritional status.

In addition to these immediate risk factors, Malian children living in households with intermediate socioeconomic levels had a higher prevalence of anemia than those living in poor households. The same phenomenon was observed in Benin at the community level, with children from communities with a medium level of development at less risk for anemia than their peers from low and high development level communities. It is likely that these communities with a medium development index represent communities undergoing developmental and nutritional transition, which may carry higher risks of malnutrition and infections [46].

Individual and contextual contribution to the prevalence of anemia

Risk factors for moderate to severe anemia vary throughout Benin and Mali, but the variability occurs at the individual level; there is little community-level clustering (ICC of 14% to 19% according to the results of the multilevel analyses). Therefore, anemia interventions need not be community-specific. If, through successfully addressing risk factors, anemia is decreased in one part of Benin or Mali, addressing the same risk factor could successfully reduce anemia throughout the country. Identifying a successful program to replicate is a priority; it is likely that such a program would include a focus on overall child health indicators as identified in this study (reducing infections, especially reducing malaria through bednet use, reducing protein–energy malnutrition, and improving immunization).

Our results could not be directly compared with previous studies on vitamin A or anemia clustering. Most previous studies have interpreted high prevalences of anemia or vitamin A deficiency in an area as “high levels of clustering” [47–50]. These studies did not separate the contribution of individual characteristics to the risk of anemia (or vitamin A deficiency) from the contribution of the cluster (which is the real clustering effect). Other studies have shown evidence of clustering at the household level [51, 52]. With the newly available hierarchical regression techniques, it is possible to separate these individual effects from contextual effects and therefore to give accurate measures of clustering. Agarwal et al. [53], the only study available that estimates clustering of vitamin A deficiency based on hierarchical analysis, reports an intraclass correlation of 13% (95% CI, 10%–17%) in villages. These results are comparable to our findings (ICC of 14%–19%). More multilevel studies are needed to determine whether the relatively low clustering effect found in this study applies to other countries.

Limitations

The primary limitation of this study was its cross-sectional design. Despite the robustness of the analyses, control for the principal confounders, and the consistency of our main results with those of other studies on anemia, no causal inference can be made. However, the correlations observed can help us guide the development and implementation of anemia intervention policies. Another limit of the study was the absence of data on children's dietary iron intake and supplementation in the DHS surveys. However, it is already well documented that iron-deficiency anemia contributes to approximately 50% of total anemia in West Africa [1, 54].

Acknowledgments

This work was partially funded by the Mellon Foundation through AFSSA scholarships.

References

1. Stoltzfus RJ. Iron-deficiency anemia: Reexamining the nature and magnitude of the public health problem. Summary: Implications for research and programs. J Nutr 2001;131(2S-2):697S–700S; discussion 700S–1S.
2. Brabin BJ, Premji Z, Verhoeff F. An analysis of anemia and child mortality. J Nutr 2001;131(2S-2):636S–45S; discussion 646S–8S.
3. Pollitt E. Iron deficiency and educational deficiency. Nutr Rev 1997;55:133–41.
4. Lozoff B. Perinatal iron deficiency and the developing brain. Pediatr Res 2000;48:137–9.
5. Beard JL, Connor JR, Jones BC. Iron in the brain. Nutr Rev 1993;51:157–70.
6. Dommergues JP. [Does iron deficiency in young children have an impact on psychomotor development?]. Arch Fr Pediatr 1989;46:237–9 [in French].
7. Peirano P, Algarin C, Garrido M, Pizarro F, Roncagliolo M, Lozoff B. Interaction of iron deficiency anemia and neurofunctions in cognitive development. Nestle Nutr Workshop Ser Clin Perform Programme 2001;5:19–35; discussion 35–9.
8. Berger J, Dillon JC. [Control of iron deficiency in developing countries]. Sante 2002;12:22–30 [in French].
9. Stoltzfus R. Defining iron-deficiency anemia in public health terms: A time for reflection. J Nutr 2001;131 (2S-2):565S–7S.
10. Baker SJ, DeMaeyer EM. Nutritional anemia: Its understanding and control with special reference to the work of the World Health Organization. Am J Clin Nutr 1979;32:368–417.
11. Hurrell RF. Phytic acid degradation as a means of improving iron absorption. Int J Vitam Nutr Res 2004; 74:445–52.
12. Zijp IM, Korver O, Tijburg LB. Effect of tea and other dietary factors on iron absorption. Crit Rev Food Sci Nutr 2000;40:371–98.
13. Bothwell TH, Baynes RD, MacFarlane BJ, MacPhail AP. Nutritional iron requirements and food iron absorption. J Intern Med 1989;226:357–65.
14. Korenromp EL, Armstrong-Schellenberg JR, Williams BG, Nahlen BL, Snow RW. Impact of malaria control on childhood anaemia in Africa—A quantitative review. Trop Med Int Health 2004;9:1050–65.
15. Merlo J, Chaix B, Yang M, Lynch J, Rastam L. A brief conceptual tutorial of multilevel analysis in social epidemiology: Linking the statistical concept of clustering to the idea of contextual phenomenon. J Epidemiol Community Health 2005;59:443–9.
16. Leyland AH, Groenewegen PP. Multilevel modelling and public health policy. Scand J Public Health 2003;31:267–74.
17. Diez Roux AV. A glossary for multilevel analysis. J Epidemiol Community Health 2002;56:588–94.
18. Diez-Roux AV. Multilevel analysis in public health research. Annu Rev Public Health 2000;21:171–92.
19. Rice N, Leyland A. Multilevel models: Applications to health data. J Health Serv Res Policy 1996;1:154–64.
20. Goldstein H, Browne W, Rasbash J. Multilevel modelling of medical data. Stat Med 2002;21:3291–315.
21. Macro International I. DHS sampling manual. Calverton, Md, USA: Macro International, 1996.
22. Institut National de la Statistique et de l'Analyse Economique (INSAE) et ORC Macro. Enquête demographique et de sante au Bénin 2001. Calverton, MD, USA: INSAE and ORC Macro, 2002.
23. Cellule de la Planification et de la Statistique du Ministère de la Santé (CPS/MS) et Direction Nationale de la Statistique et de l'Informatique (DNSI) et ORC Macro. Enquête demographique et de sante au Mali, 2001. Calverton, MD, USA: CPS/MS, DNSI, and ORC Macro, 2002.
24. World Health Organization. Nutritional anemia: Report of a WHO Scientific Group. Technical Report Series-405. Geneva: WHO, 1968.
25. Rutstein SO, Kiersten J. The DHS Wealth Index. Calverton, MD, USA: ORC Macro, 2004.
26. Snjjders T, Bosker RJ. Multilevel analysis: An introduction to basic and advanced multilevel modeling. London: Sage Publishers, 1999.
27. Greenland S. Principles of multilevel modelling. Int J Epidemiol 2000;29:158–67.
28. Zunzunegui MV, Garcia de Yebenes MJ, Forster M, Aguilar Conesa MD, Rodriguez Laso A, Otero A. [Multilevel model applications to the analysis of longitudinal data.] Rev Esp Salud Publica 2004;78:177–88 [in Spanish].
29. Moerbeek M, van Breukelen GJ, Berger MP. A comparison between traditional methods and multilevel regression for the analysis of multicenter intervention studies. J Clin Epidemiol 2003;56:341–50.
30. Rasbash J, Steele F, Browne F, Prosser W. A user's guide to MLwiN version 2.0. London: London Institute of Education, 2004.
31. Desai MR, Terlouw DJ, Kwena AM, Phillips-Howard PA, Kariuki SK, Wannemuehler KA, Odhacha A, Hawley WA, Shi YP, Nahlen BL, Ter Kuile FO. Factors associated with hemoglobin concentrations in pre-school children in Western Kenya: Cross-sectional studies. Am J Trop

Med Hyg 2005;72:47–59.
32. Hassan K, Sullivan KM, Yip R, Woodruff BA. Factors associated with anemia in refugee children. J Nutr 1997;127:2194–8.
33. Fotso JC, Kuate-Defo B. Socioeconomic inequalities in early childhood malnutrition and morbidity: Modification of the household-level effects by the community SES. Health Place 2005;11:205–25.
34. Tomkins AM. Protein-energy malnutrition and risk of infection. Proc Nutr Soc 1986;45:289–304.
35. Marchant T, Schellenberg JA, Edgar T, Nathan R, Abdulla S, Mukasa O, Mponda H, Lengeler C. Socially marketed insecticide-treated nets improve malaria and anaemia in pregnancy in southern Tanzania. Trop Med Int Health 2002;7:149–58.
36. Shulman CE, Dorman EK, Talisuna AO, Lowe BS, Nevill C, Snow RW, Jilo H, Peshu N, Bulmer JN, Graham S, Marsh K. A community randomized controlled trial of insecticide-treated bednets for the prevention of malaria and anaemia among primigravid women on the Kenyan coast. Trop Med Int Health 1998;3:197–204.
37. ter Kuile FO, Terlouw DJ, Kariuki SK, Phillips-Howard PA, Mirel LB, Hawley WA, Friedman JF, Shi YP, Kolczak MS, Lal AA, Vulule JM, Nahlen BL. Impact of permethrin-treated bed nets on malaria, anemia, and growth in infants in an area of intense perennial malaria transmission in western Kenya. Am J Trop Med Hyg 2003;68(4 suppl):68–77.
38. van den Broek N. Anaemia in pregnancy in sub-Saharan countries. Eur J Obstet Gynecol Reprod Biol 2001;96:4–6.
39. Fraser-Hurt N, Felger I, Edoh D, Steiger S, Mashaka M, Masanja H, Smith T, Mbena F, Beck HP. Effect of insecticide-treated bed nets on haemoglobin values, prevalence and multiplicity of infection with Plasmodium falciparum in a randomized controlled trial in Tanzania. Trans R Soc Trop Med Hyg 1999;93(suppl 1): 47–51.
40. D'Alessandro U, Langerock P, Bennett S, Francis N, Cham K, Greenwood BM. The impact of a national impregnated bed net programme on the outcome of pregnancy in primigravidae in The Gambia. Trans R Soc Trop Med Hyg 1996;90:487–92.
41. Browne EN, Maude GH, Binka FN. The impact of insecticide-treated bednets on malaria and anaemia in pregnancy in Kassena-Nankana district, Ghana: A randomized controlled trial. Trop Med Int Health 2001; 6:667–76.
42. Abdulla S, Gemperli A, Mukasa O, Armstrong Schellenberg JR, Lengeler C, Vounatsou P, Smith T. Spatial effects of the social marketing of insecticide-treated nets on malaria morbidity. Trop Med Int Health 2005;10:11–8.
43. Ayoya MA, Spiekermann-Brouwer GM, Traore AK, Stoltzfus RJ, Garza C. Determinants of anemia among pregnant women in Mali. Food Nutr Bull 2006;27:3–11.
44. Warrier RP, Lokeshwar MR. Growth of pediatric hematology/oncology in India. Am J Pediatr Hematol Oncol 1990;12:92–4.
45. Verhoef H, West CE, Veenemans J, Beguin Y, Kok FJ. Stunting may determine the severity of malaria-associated anemia in African children. Pediatrics 2002;110:e48.
46. Popkin BM. The nutrition transition: An overview of world patterns of change. Nutr Rev 2004;62(7 pt 2): S140–3.
47. Tarwotjo I, Tilden R, Pettiss S, Sommer A, Soedibjo S, Hussaini G, Sulistijo JS, Nendrawati H. Interactions of community nutritional status and xerophthalmia in Indonesia. Am J Clin Nutr 1983;37:645–51.
48. Katz J, Zeger SL, Tielsch JM. Village and household clustering of xerophthalmia and trachoma. Int J Epidemiol 1988;17:865–9.
49. Katz J, Zeger SL, West KP Jr, Tielsch JM, Sommer A. Clustering of xerophthalmia within households and villages. Int J Epidemiol 1993;22:709–15.
50. Katz J, Yoon SS, Brendel K, West KP Jr. Sampling designs for xerophthalmia prevalence surveys. Int J Epidemiol 1997;26:1041–8.
51. Polack SR, Solomon AW, Alexander ND, Massae PA, Safari S, Shao JF, Foster A, Mabey DC. The household distribution of trachoma in a Tanzanian village: An application of GIS to the study of trachoma. Trans R Soc Trop Med Hyg 2005;99:218–25.
52. Bailey R, Osmond C, Mabey DC, Whittle HC, Ward ME. Analysis of the household distribution of trachoma in a Gambian village using a Monte Carlo simulation procedure. Int J Epidemiol 1989;18:944–51.
53. Agarwal GG, Awasthi S, Walter SD. Intra-class correlation estimates for assessment of vitamin A intake in children. J Health Popul Nutr 2005;23:66–73.
54. Asobayire FS, Adou P, Davidsson L, Cook JD, Hurrell RF. Prevalence of iron deficiency with and without concurrent anemia in population groups with high prevalences of malaria and other infections: A study in Cote d'Ivoire. Am J Clin Nutr 2001;74:776–82.

Breastfeeding and mixed feeding practices in Malawi: Timing, reasons, decision makers, and child health consequences

Rachel Bezner Kerr, Peter R. Berti, and Marko Chirwa*

Abstract

Background. *In order to effectively promote exclusive breastfeeding, it is important to first understand who makes child-care and child-feeding decisions, and why those decisions are made; as in most parts of the world, exclusive breastfeeding until 6 months of age is uncommon in Malawi.*

Objective. *To characterize early infant foods in rural northern Malawi, who the decision-makers are, their motivation, and the consequences for child growth, in order to design a more effective program for improved child health and nutrition.*

Methods. *In a rural area of northern Malawi, 160 caregivers of children 6 to 48 months of age were asked to recall the child's age at introduction of 19 common early infant foods, who decided to introduce the food, and why. The heights and weights of the 160 children were measured.*

Results. *Sixty-five percent of the children were given food in their first month, and only 4% of the children were exclusively breastfed for 6 months.* Mzuwula *and* dawale *(two herbal infusions), water, and porridge were common early foods. Grandmothers introduced mzuwula to protect the children from illness; other foods were usually introduced by mothers or grandmothers in response to perceived hunger. The early introduction of porridge and dawale, but not mzuwula, was associated with worse anthropometric status. Mzuwula, which is not associated with poor growth, is usually made with boiled water and given in small amounts. Conversely, porridge, which is associated with poor child growth, is potentially contaminated and is served in larger amounts, which would displace breastmilk.*

Conclusions. *Promoters of exclusive breastfeeding should target their messages to appropriate decision makers and consider targeting foods that are most harmful to child growth.*

Key words: Child growth, exclusive breastfeeding, Malawi, mixed feeding

Introduction

The quality of the diet in the first years of life is a key factor in children's health and survival [1]. Mild to moderate child malnutrition has been estimated to account for 53% of all child deaths in developing countries [2]. Appropriate breastfeeding and complementary feeding methods could potentially halve African infant mortality rates. Current nutrition policy encourages mothers to exclusively breastfeed their children for the first 6 months of life [3], providing the infant with a nutritionally sufficient, clean, and safe diet. Despite the recommendations and widespread promotion of exclusive breastfeeding, it is often not practiced, even in developing countries where it would be most beneficial. Most recent estimates suggest that one-third of all infants in sub-Saharan Africa are exclusively breastfed, an increase from 15% in 1990 [4].

Malawi is a landlocked country in southeastern Africa with a population of approximately 11 million, 80% of whom live in rural areas. The infant mortality rate is 114 per 1,000 live births, and the under-five mortality rate is 183 per 1,000 live births [5]. High levels of child malnutrition have been observed in Malawi for over two decades [6, 7]. Growth faltering begins soon after birth, with rapid worsening beginning at approximately 5 months, continuing through the second year, when stunting prevalence peaks at 60%, and remain-

Rachel Bezner Kerr is affiliated with the Department of Geography, The University of Western Ontario, London, Ontario, Canada, Peter R. Berti is affiliated with HealthBridge, Ottawa; Marko Chirwa was affiliated with Soils, Food and Healthy Communities, Ekwendeni Hospital, Ekwendeni, Malawi.

Please direct queries to the corresponding author: Rachel Bezner Kerr, Department of Geography, SSC 2409, 1151 Richmond Street N, The University of Western Ontario, London, Ontario, Canada,N6A 5C2; e-mail: rbeznerkerr@uwo.ca.

These data were presented at the 2005 International Union of Nutritional Sciences congress in Durban, South Africa.

* Deceased.

ing above 50% up to the age of five [7]. About 25% of Malawian under-five children are underweight; peak levels occur between 12 and 24 months of age [7]. Food insecurity is a major problem, with between 70% and 85% of households experiencing food shortages on an annual basis [8].

Exclusive breastfeeding appears to be uncommon in Malawi, despite very high rates of breastfeeding (nonexclusive + exclusive breastfeeding = 95% at 17 months of age [9]) and a national effort to increase exclusive breastfeeding [9-11]. A confounding factor and a perceived disincentive to exclusive breastfeeding is the high prevalence rate of HIV-positive Malawian pregnant women, estimated at 14% to 20% [12]. One recent study suggested that HIV-infected Malawian women were concerned that exclusive breastfeeding would compromise their health status if they did not have better access to nutritious foods [13].

The objective of the research presented in this paper is to characterize early infant-feeding practices in a rural Malawi setting, and specifically, to document the foods that are introduced early to the children, who the decision makers are for these practices, their motivation for doing so, and the consequences for child growth. With this characterization, a more effective program for the promotion of exclusive breastfeeding and healthy feeding practices might be designed.

Description of study site

The research was conducted in a rural area of northern Malawi near the town of Ekwendeni in Mzimba District. Approximately 80% of the population are smallholder farmers. Maize is the primary staple crop and is harvested in May and June after the single annual rainy season. Other important crops include beans, squash, groundnuts, and sweet potatoes, as well as tobacco as the major cash crop.

This research was carried out within a larger community-based research and development project, the Soils, Food and Healthy Communities Project (SFHC), based at Ekwendeni Hospital, that began in 2000. SFHC tries to improve the health of resource-poor households through participatory research that introduces relay cropping and intercrops of legumes to improve soil fertility, food security, and nutrition of poor households, in particular by increasing legume consumption by young children.

Methods

The overall research approach was interdisciplinary and multimethod. The research team, composed of a sociologist, a nutritionist, hospital staff, and farmers, carried out a combination of qualitative and quantitative research to understand early infant-feeding practices. Early introduction of non-breastmilk foods to infants was identified as an issue of interest on the basis of the qualitative research, and specific questions about these practices were included in the survey. Twenty-one semistructured interviews were conducted in 2001 in which key informants (mothers, older women, and traditional medicine practitioners) identified by community members were asked about pregnancy, breastfeeding, early infant feeding, and general care practices and beliefs. The questions were developed on the basis of initial test interviews with informants. Four focus groups were held with groups of men and women in the villages, using similar questions as the semistructured interviews. In addition, free lists of foods eaten by young children were obtained from 28 informants [14]. The team worked in pairs to carry out the interviews, with one person interviewing and the other person translating or taking notes. Informed consent was obtained prior to all interviews. All interviews were recorded, and the tapes were transcribed and translated into English.

Using the qualitative research as a basis for design, an extensive survey on agriculture and child-care and child-feeding practices was conducted in February 2002 (during the "hungry season") with 264 households. Questions were included about the timing of the introduction of 19 foods (identified as the most common early infant foods in the qualitative research), the reason or reasons that each of these foods was introduced, and the individual(s) who made the decision to introduce the food.

The subjects of this study were children 6 to 48 months of age and their primary caregivers (99% of respondents were the children's mothers; the remaining respondents were one father, one grandmother, and one stepmother) from two sets of households: those in intervention villages and those in control villages. The original survey design was intended to compare intervention households with nonintervention households in terms of food security, soil fertility, child nutritional status, and child-feeding practices. Intervention-village households were participating in the SFHC Project; control villages were selected on the basis of similar socioeconomic and environmental conditions. Households were recruited to the Project at village-level meetings organized by the hospital. Survey participants from intervention-village households were randomly selected from this group. In order to reduce selection bias, since other studies have indicated that more food-secure households join participatory agricultural projects [15], and to control for growth differences based on age, control households were matched with intervention households on the basis of two criteria: age of the child and food-security status (operationalized as the month when self-grown maize stores are exhausted). The majority of respondents (87%) had a primary-level education, 6.5% had a secondary-level education, and 6% had no formal schooling. Most of the respondents (74%) were in monogamous marriages, 18% were in polygamous marriages, 6% were divorced, 1% were widowed, and 1% were single. Sixty-three percent of respondents had one under-five child in the household and 35% had two under-five children. The average number of people in the household was five.

In 160 households with a child under 4 years of age, the primary caregiver was interviewed in the home about the child's consumption of 19 specific early infant foods and was also asked if the child consumed other foods (**fig. 1**). In this sample, 112 households (57 control, 55 intervention) had children 6 to 24 months of age, and 48 (22 control, 26 intervention) had children 24 to 48 months of age. In the older group, 45 children were between 24 and 34 months of age and 3 were more than 34 months of age. For each of the 19 foods, the caregiver was asked if the food had been introduced to the child, and if so at what age (in months), the reasons for introducing the food (from a list of 10 choices, including "other"), and who was involved in deciding when the food should be introduced (a list was provided of 11 individuals, as well as "other," "don't know," and no answer). The surveys were pretested in a neighboring village and revised. In order to minimize biased responses due to extensive hospital education about exclusive breastfeeding, no direct questions were asked about how long the women had breastfed exclusively, The results suggest that asking women indirectly minimized response bias about breastfeeding practices. Although the time period for caregivers to recall what foods were given to their children was long (up to 48 months), exact amounts were not assessed, and initial qualitative interviews indicated that caregivers were able to easily recall what foods were initially given to their children. Thus, measurement bias is anticipated to be low.

Anthropometric data were collected from 405 children in a central village location (e.g., primary school) by trained research assistants following standardized procedures and using calibrated equipment. Nude weights were measured by an electronic TANITA "Baby and Mommy Scale" (model 1582; lb/kg version) and recorded to the nearest 0.01 kg. A 100-cm length board (Perspective Enterprises) was used to record the length of children from sampled intervention and control households.

The research protocol was reviewed and approved by the Malawi National Research Council and by the Cornell University Committee on Human Subjects. Informed consent was obtained orally from each adult caregiver prior to the survey and collection of anthropometric data.

Data management and analysis

Qualitative data were analyzed for trends, key concepts, and practices by data analysis techniques described by Miles and Huberman [16] and Patton [17]. A coding scheme was developed using different themes based

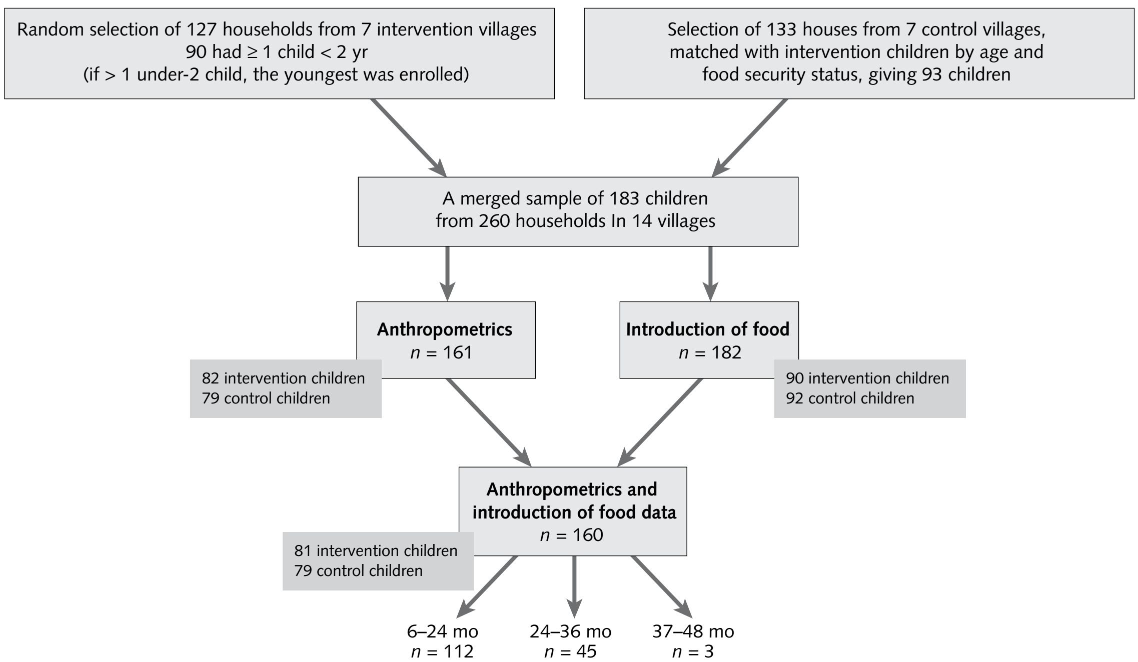

FIG. 1. Flow chart of sampling methods and number of participants, February 2002 survey

on the interview schedules. This code list was modified during the data analysis process as new themes emerged. A set of definitions for each code was also developed and modified throughout the analytical process. After coding, the data were read over and examined for emerging themes and trends. Inductive analysis was used to identify indigenous food concepts, feeding practices, and key social roles in child feeding. Follow-up interviews and focus group discussions were conducted to better understand these concepts, practices, and roles. Identification of sensitive topics in the qualitative research allowed the team to carefully word survey questions to avoid biased answers.

The quantitative data, i.e., the anthropometric data and the data from the survey regarding age at introduction of the food, reasons for introducing the food, and the decision makers, were entered in Excel and analyzed using SAS (version 8). For each month of age, the percentage of children given a food for the first time was calculated as (number of children first given the food at age X or younger) ÷ (number of children whose age was X or more). The effect of household food security and socioeconomic status (SES) on age at introduction of each food was tested by multinomial logistic regression (age at introduction vs. month household ran out of or expected to run out of maize, where ages at introduction are classified as < 1 month, 1-4 months, 4-6 months, and > 6 months or never [i.e., the child was > 6 months of age but had not been given the food]). The reasons for the introduction and the decision makers were tabulated, and cross-tabulations between "reasons" and "decision makers" were also generated.

The effect of early introduction of individual foods was tested by comparing growth status with age at introduction of the three key early infant foods. The children's heights and weights were converted to height-for-age, weight-for-age, and weight-for-height z-scores (HAZ, WAZ, and WHZ, respectively) by calculating the difference between the child's measure and the age-matched mean measure of a reference population, and then dividing by the standard deviation of a reference population [18]. In SAS, a general linear model was used where the dependent variable was HAZ, WAZ, or WHZ and the independent variable was age at introduction of a food (categorized as described above). The model included children from 6 to 48 months of age for whom we had both anthropometric data and data on the timing of introduction of early infant food. "Porridge" includes the earliest introduction of any type of porridge: porridge with unrefined maize flour (*mgaiwa*) and porridge with milk and maize flour (*chintuwe*) (**table 1**). "*Dawale*" includes dawale, "dawale water," and "dawale porridge," and the earliest age of introduction of any of the three types of dawale was used. The model tested for effects of the child's sex and age, the month at which the household

TABLE 1. Definitions of local food terms based on qualitative interviews

Term	Definition
Mzuwula	Herbal infusion made from the leaves of specific tree species found in the area. The infusion is made from crushed leaves mixed with boiled or cold water
Dawale	Herbal infusion made from the roots of a specific tree species found in the area. Sometimes other leaves are crushed and added to the infusion. Sometimes fed to infant as an infusion, sometimes added to porridge to make a very thin porridge.
Chinthipu	Very thin, watery, maize porridge
Mgaiwa	Unrefined maize flour
Chintuwe	Porridge with milk and maize flour

ran out of or expected to run out of maize (as an indicator of food security), and an SES index. The SES index consisted of a rating for housing, ownership of material goods, and maternal education. Housing materials were ranked from low to high for the following materials: sticks, thatch or grass, unfired brick, clay tiles, fired brick, and iron sheets. The material goods were oxcart, wheelbarrow, radio, plow, motorcycle, ridger (a soil-tilling implement), mosquito net, bicycle, sofa or armchair, table and chairs, and tobacco press. The index assigned one point for each item owned.

Qualitative interviews, participant observation, and focus group discussions suggested that housing materials and ownership of goods were good SES indicators. Other studies in Malawi have included ownership of goods and housing type as good proxy indicators of SES [19]. Maternal education was included because other studies in Malawi and elsewhere have found that maternal education can have a significant effect on children's nutritional status [11]. The independent variables are treated as fixed effects in this model. There was good concordance between individual indicators and SES points.

Results

In the results presented in this paper, there are no differences between intervention and control villages (data not shown), and therefore the data are presented for the villages combined.

Perceptions of breastfeeding

Informants felt that the first milk (colostrum) was good for babies, as well as breastmilk, but exclusive breastfeeding was not widely practiced. They said that breastmilk protects a baby from diseases, helps their bowels develop, and gives them energy. All women interviewed named breastmilk as the primary source of food for babies, but several other foods could be given to a baby if it cried. The crying of a baby was seen as a sign of hunger and an indication that the baby was not getting enough food from breastmilk. Many women said that breastmilk was given until the baby cried after breastfeeding, at which time a thin maize porridge was given. Some informants said that certain babies are "born hungry" and may have to be fed porridge at a very early age, or gripe water or different herbal infusions, discussed below, in some cases. The mothers said that the most important reason for introducing porridge was that the baby was crying.

Age at introduction

The age at introduction of the 19 foods is summarized in **figure 2** in a cumulative frequency plot. The percentage of children who have been given a food by the end of each month of age is shown. Sixty-five percent of the children are given some type of food in the first month. By 6 months, 96% of the children have been given some type of food (see the "Any food" line in **fig. 2**).

There are three notable patterns in the introduction of foods. First, mzuwula (an infusion made with the leaves of a local tree) is introduced to 50% of the children in the first month and then to only an additional 10% over the next 17 months. Mzuwula is an infusion of water and pounded leaves and roots from a particular tree species. Approximately 80% of the children who were given any food in the first month were given mzuwula. Second, plain water or water with dawale, another kind of root water, is introduced to 20% of the children in the first month and then to an additional 10% per month for the next 6 months. Third, porridge with chinthipu (thin porridge with white maize flour) or porridge with dawale is introduced to 10% of children in the second month and to 70% of children by the sixth month.

The exact amount given to children varied with the substance. In the qualitative interviews, informants were asked to estimate the amounts given to children. They indicated that mzuwula was typically given in small amounts (e.g., a teaspoon or about 5 mL), whereas porridge, water, or dawale could be given in amounts ranging from 25 to 500 mL. Specific amounts were not measured.

There was no relationship between age at introduction of each food, anthropometric measurements, and household food security or SES. There was a significant relationship between total SES points and the introduction of dawale. Households with a higher SES index were more likely to introduce dawale to children after the age of 6 months. The reasons for this difference may be linked to a local perception that food-insecure women produce insufficient breastmilk [20]. These perceptions and links to breastfeeding patterns are being further studied through qualitative research.

Reported reasons for introduction

The caregivers' reported reasons that the various foods were introduced to the children are summarized in **table 2**. A commonly reported reason for introducing

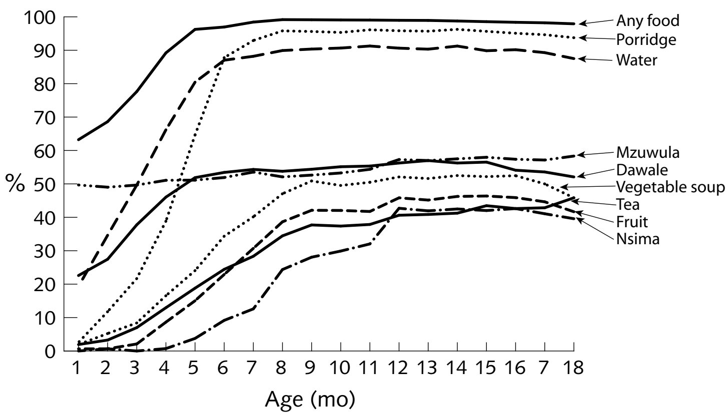

FIG. 2. Age at introduction of foods to Ekwendeni children: cumulative frequency.

Dawale includes "dawale," "dawale water," and "dawale porridge." Dawale is a root that is cut up and added to water, which is boiled and then strained and given as water or added to porridge. Porridge includes *chinthipu* (made from maize and water), *mgaiwa* (made from fermented, unrefined maize), and *chinthuwe* (made from milk and maize flour). Note that the percentage of children given the food fluctuates after the age of 6 to 9 months. This is a sampling artifact (fewer children are sampled at older ages); in reality there would be a plateau at the percentage of children who ever eat a food. There are other foods or food groups not shown that never exceed 10% cumulative frequency (gripe water, *chindongwa* [a groundnut-maize bread], soft drinks, sugar water, and milk) or 20% cumulative frequency (infant formula) many foods was that the child was hungry or crying. The most common reason reported as "other" was that the child was thirsty. However, mzuwula was given in 84% of cases to "protect" the child from illness believed to be caused by "promiscuity" of the mother or father (even within marriage) within 1 year of birth, or in some instances by promiscuity of anyone in the village [20].

Decision makers

The individuals who decided when a given food was to be introduced are summarized in **table 3**. Most often the caregiver (in 99% of cases the mother) was the decision maker, but the mother-in-law and occasionally the father-in-law were also important decision makers, particularly with regard to foods that were given to protect the child.

Cross-tabulations of "reasons × decision makers" revealed that water and porridge were given in 80% of cases because the child was perceived by the mother or mother-in-law to be hungry and crying. However, in 78% of cases when mzuwula was introduced, the mother-in-law made the decision and it was given to protect the child from the disease associated with promiscuity. Young women noted in focus groups that their mothers-in-law have tremendous influence over all child-care and feeding activities [20]. A crying baby is perceived as a sign of poor child care, and a grandmother can even remove a child from the home if she feels the child is not getting enough food. Vigorous discussions with older women indicated a very strong belief that breastmilk is not adequate for young babies, particularly during the "hungry season" from December to March, which is also the period reported by informants to have the highest level of illness from water-borne diseases.

Relationship between early infant-feeding practices and anthropometric measurements

We tested the relationship between anthropometric measurements of 160 children 6 to 48 months of age (of whom 112 were less than 24 months old) and the timing of introduction of four food types during infancy. The model tested the z-score vs. the age at introduction of water, mzuwula, porridge, or dawale, while controlling for the child's age and sex and the age at which the household ran out of or expected to run out of maize, as a general indicator of food security and SES. Age at introduction was classified as < 1 month, 1 to 3 months, 4 to 6 months, or > 6 months or never). The models

TABLE 2. Number of times various reasons were given by mothers for feeding particular foods to their children (n = 157)[a]

Reason	Water	Maize porridge	Mzuwula	Vegetable soup	Fermented, unrefined maize porridge (mgaiwa)	Fruit	Tea	Nsima	Dawale	Dawale water	Dawale porridge	Formula	Gripe water	Milk	Sweet potato	Maize bread	Soft drink	Milk and maize porridge	Sugar water
Advised to feed	4	2	1	1	4	1	1	3	1	1	1	0	0	1	0	1	0	0	0
Child ill	3	0	2	0	0	2	1	0	1	0	0	0	3	0	0	1	0	1	0
Child crying or hungry	102	103	9	26	40	21	29	28	19	16	15	14	4	5	6	1	6	8	1
Child's interest in food	1	1	0	4	2	23	19	16	0	0	0	5	0	0	5	6	2	0	1
Mother ill	2	1	0	2	2	2	1	2	0	0	0	0	0	0	0	0	0	0	0
Not enough breastmilk	9	15	0	0	5	1	4	3	0	3	1	0	0	4	0	0	1	3	0
To protect child	2	1	72	16	0	12	6	1	15	10	6	0	11	3	0	0	0	0	0
To strengthen child	2	17	6	27	30	19	5	16	7	11	6	6	0	4	2	1	3	0	0
Mother couldn't breastfeed	1	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
Other	22	0	0	0	1	2	0	1	0	2	1	0	0	0	0	2	1	0	2
No. of children given food	138	125	86	73	71	68	62	55	41	35	25	23	18	14	13	12	11	9	4

a. Up to three reasons were given per food.

TABLE 3. Individuals who decided when a food was to be introduced to the child's diet

Decision maker	Total no. of decisions	Water	Porridge with chinthipu	Mzuwula	Vegetable soup	Porridge with unrefined flour (mgaiwa)	Fruit	Tea	Nsima	Root water (dawale)	Dawale water	Dawale porridge	Formula	Gripe water	Milk	Sweet potato	Corn bread (chindongwa)	Soft drink	Porridge with chintuwe	Sugar water
Mother	627	100	96	15	69	63	56	56	50	16	12	12	22	7	11	11	11	10	7	3
Father-in-law or mother-in-law	206	31	28	63	3	7	5	1	4	23	21	12	1	1	1	0	1	2	2	0
Spouse	34	2	2	0	3	0	5	4	3	1	0	0	1	9	2	1	0	0	1	0
Parents	9	1	1	2	1	0	0	0	1	0	2	1	0	0	0	0	0	0	0	0
Grandparent	8	0	1	3	0	0	0	0	0	2	0	1	0	1	0	0	0	0	0	0
Sister or brother	5	0	0	0	1	0	3	0	0	0	0	0	1	0	0	0	0	0	0	0
Sister-in-law or brother-in-law	4	0	1	2	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
Child	3	0	0	0	0	0	1	0	2	0	0	0	0	0	0	0	0	0	0	0
Aunt or uncle	2	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Niece or nephew	1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
No answer	16	2	1	2	2	2	1	0	2	0	1	1	0	0	0	1	0	0	0	1
No. of children given food		138	125	86	73	71	68	62	55	41	35	25	23	18	14	13	12	11	9	4

for WHZ (p = .002, r^2 = 0.21) and WAZ (p = .009, r^2 = 0.19) were significant. The age at introduction of water, porridge, and dawale, but not mzuwula, was associated with WHZ and WAZ (p < .02 in all cases except WHZ–dawale, where p = .08). The results for porridge and dawale are depicted in **figure 3**; the early introduction of dawale or porridge is associated with a z-score disadvantage of 0.5 to 1.5. The relationship with age at introduction of water is ambiguous, with early (< 1 month) and late (> 6 months) introduction of water having an advantage of approximately 0.7 z-score over intermediate introduction.

Discussion

Giving an infant foods other than breastmilk during the first 6 months of life displaces the more nutritious breastmilk from the infant's diet and risks introducing diarrhea-causing pathogens to the child. It is acknowledged that the results of this study cannot be generalized beyond the study area, since the study population is in an area with an active primary health care program and was not randomly selected. In addition, the cultural reasons for specific feeding practices found in the Ekwendeni area are not applicable to the entire country. Nonetheless, some broader implications can be drawn from this research, because despite this active program, low rates of exclusive breastfeeding were found, and similar practices have been noted in other regions of southern Africa. The health consequences of not practicing exclusive breastfeeding have been demonstrated in numerous studies in numerous settings [21], and feeding anything other than breastmilk is discouraged by Ekwendeni Hospital. However, despite an ongoing exclusive breastfeeding promotion campaign and recognition of the hospital as a baby-friendly hospital since 2000, as in many settings throughout the world, exclusive breastfeeding is rarely practiced in the surveyed population. Sixty-five percent of infants were given some food in their first month, and only 4% of children were reported to be exclusively breastfed for 6 months.

Typically, promotion of exclusive breastfeeding is targeted to the mothers in health clinic education sessions. We found that

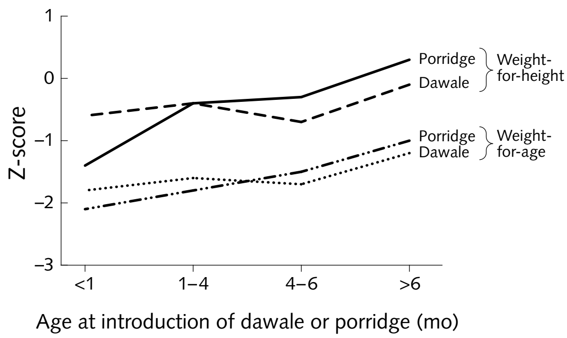

FIG. 3. Least-square means of weight-for-height and weight-for-age in children according to age at first introduction of porridge or dawale

such promotion may be inappropriate or inadequate, since the mother does not always decide what a child eats and often it is the mother-in-law who decides. Mzuwula is given to "protect" children against an illness believed to be caused by promiscuity [20]. This belief is firmly held by many villagers. Is it therefore necessary to promote the reduction of the practice of feeding mzuwula to infants? Conventional exclusive breastfeeding policy would certainly discourage giving infants mzuwula. However, mzuwula is often prepared with boiled water (and therefore is likely to be sterile), and it is given infrequently and in small amounts (and thus displaces little breastmilk). Perhaps not surprisingly, then, its early introduction is not related to child growth. Furthermore, it is tied in to a very strongly held belief system, the behavior is deeply engrained, and it would probably be very difficult to change. Project staff initiated a nutrition program with villagers that attempts to promote healthy ways of using mzuwula, such as always boiling the water prior to preparation, or bathing the child in mzuwula rather than feeding it to the child. Thus, the Project does not attack strongly held traditional beliefs while trying to improve child-feeding practices.

Porridge and dawale, on the other hand, are given to the child because the decision maker believes the child is hungry or thirsty. Implicitly tied to this idea is the notion that mothers produce insufficient breastmilk for babies, a belief found in other parts of southern Africa [22]. Porridge is prepared in the morning and then allowed to cool and is served throughout the day, and thus is a potential source of pathogens [23]. It has limited nutritional value but is filling, and it is given to the child regularly and in amounts that are likely to displace breastmilk. Further, its early introduction is related to poor growth in this population (**fig. 3**). Anthropometric status is not significantly associated with "age at introduction of any food" (or, to say it in a different way, months of exclusive breastfeeding), but is significantly associated only with the age at introduction of those specific foods that are of particularly low nutritional quality and are potentially contaminated, i.e., porridge and dawale.

There is no concordance between the timing of introduction of the four types of food discussed here. Many caregivers who introduce mzuwula early do not introduce dawale early, and vice versa, and therefore there can be differences in the relationship between age at introduction and anthropometric measurements for the different foods. Although the relationships may indicate causality, there is no evidence from this dataset to support a causal relationship. The early introduction of porridge and dawale may be associated with other harmful but unobserved behaviors, or it may be a general marker of food insecurity, maternal education, or some other macro-level variable. It also may be an example of reverse causality if children who are growing poorly are given infant foods earlier by concerned caregivers. If that is the case, the early introduction of non-breastmilk foods did not improve growth in these children, who up to 4 years later were still lagging behind their peers. Future research in the region, using longitudinal rather than cross-sectional sampling, will address this question.

A recent study that compared infant-feeding patterns and child growth in Ghana, Peru, and India found that there was no significant difference in the risk of death between exclusively breastfed and predominantly breastfed infants, and that nonbreastfed infants had a significantly greater risk of death than those who were exclusively or predominantly breastfed [24]. The authors of this study concluded that the risk of not breastfeeding needs to be taken into account when advising HIV-infected mothers about their infant-feeding options, and that rather than focusing on exclusive breastfeeding in areas where predominant breastfeeding is the norm, continued high rates of breastfeeding should be encouraged. Our findings support the idea of promoting predominant breastfeeding and the need to identify specific infant-feeding practices that may be particularly problematic, such as early introduction of porridge. Breastfeeding provides other important non-growth-related benefits to both mother and child, such as reduced child infections and delayed maternal fertility postpartum.

Whether or not the relationships are causal (or reverse-causal), the biological plausibility of the relationships and the importance of discouraging early introduction of foods, encouraging exclusive breastfeeding or predominant breastfeeding, and, in particular, discouraging early introduction of porridge and dawale should be priorities in nutrition programs in Ekwendeni. The next step in this project will be to use these findings to develop an appropriate exclusive breastfeeding campaign and to monitor the success of the campaign in terms of changing behavior and improving child health. An additional complicating factor in Malawi is the recommendation of exclusive breastfeeding in the face of HIV infection rates. The World Health Organization continues to recommend exclusive breastfeeding unless replacement feeding is "acceptable, feasible, affordable, sustainable and safe," which is not the case for most households in the Malawian context. Nonetheless, the current recommendation of the Ekwendeni Hospital staff for mothers who are HIV-positive is exclusive breastfeeding for 6 months followed by abrupt weaning. The recommendation is based on studies indicating that this method reduces the transmission of HIV [25]. Informants in follow-up qualitative research have expressed confusion about what breastfeeding method is the best for their babies in the face of conflicting messages. Abrupt weaning is against current common practice and suggests that breastmilk becomes unsafe. Given the prevalence of herbal remedies such as mzuwula that are used to protect infants, mothers may consider providing different herbal remedies to mitigate the dangers of infected breastmilk.

Because extensive promotion of exclusive breastfeeding appears to have had limited success in this region, we are launching a new phase of participatory research with households to encourage exclusive breastfeeding through small group discussions involving all household members who influence child nutrition, including grandmothers, mothers, and fathers. These small groups will try to use a problem-solving approach to encourage exclusive breastfeeding. If successful, the model (first, find the reasons why exclusive breastfeeding is not practiced; second, find out who has the decision-making power to change the practice; and third, consider which particular foods should be targeted for reduction) may be used elsewhere in Malawi and in other countries where conventional exclusive breastfeeding promotional methods have not been successful.

Acknowledgments

We could not have done this research without the support of many others: Tanya Trevors, Laifolo Dakishoni, Lizzie Shumba, Rodgers Msachi, Solomon Mkumbwa, Angela Shonga, Keston Ndlovu, the survey enumerators, and all members of the Farmer Research Team and the Nutrition Research Team. Ekwendeni Hospital staff provided helpful logistical support. David Ryan carried out very helpful data entry and analysis. David Pelletier provided useful feedback throughout the research. Finally, we thank the participating members of the Soils, Food and Healthy Communities Project in Ekwendeni, who made this work possible. This work was carried out with the aid of a grant from the International Development Research Centre, Ottawa, Canada, and Presbyterian World Service and Development, Toronto, Canada.

References

1. UNICEF. Strategy for improved nutrition of children and women in developing countries. New York: UNICEF, 1990.
2. Caulfield LE, de Onis M, Blossner M, Black RE. Undernutrition as an underlying cause of child deaths associated with diarrhea, pneumonia, malaria, and measles. Am J Clin Nutr 2004;80:193–8.
3. Fifty-fourth World Health Assembly. Infant and young child nutrition. World Health Assembly 54.2, Agenda item 13.1,18 May 2001.
4. UNICEF. Progress for children: A report card on nutrition. No. 4, May 2006. New York: UNICEF, 2006. Available at: http://www.unicef.org/progressforchildren/2006n4/. Accessed 10 December 2006.
5. UNICEF. State of the world's children 2005. New York: UNICEF, 2005.
6. NSO/Macro International. Malawi Demographic and Health Survey 1992. Zomba: Malawi National Statistical Office (NSO) and ORC Macro, 1994.
7. NSO/Macro International. Malawi Demographic and Health Survey 2000. Calverton, Md, USA, and Zomba, Malawi: National Statistical Office (NSO) and ORC Macro, 2001.
8. Bezner Kerr R. Informal labor and social relations in northern Malawi: The theoretical challenges and implications of ganyu labor for food security. Rural Sociol 2005;70:67–187.
9. Centre for Social Research. Malawi social indicators survey 1995: A survey into the state of health, nutrition, water and sanitation and education of children in Malawi. Zomba: Centre for Social Research, 1996.
10. Shrestha RM. Breast-feeding and weaning practices in urban areas of Malawi. Lilongwe: Ministry of Health/UNICEF, 1989.
11. Vaahtera M, Kulmala T, Hietanen A, Ndekha M, Cullinan T, Salin ML, Ashorn P. Breastfeeding and complementary feeding practices in rural Malawi. Acta Paediatr 2001;90:328–32.
12. UNAIDS. UNAIDS/WHO AIDS epidemic update. Available at: http://www.unaids.org/epi/2005/doc/report_pdf.asp. Accessed 10 December 2006.
13. Bentley ME, Corneli AL, Piwoz E, Moses A, Nkhoma J, Carlton Tohill BC, Ahmed Y, Adair L, Jamieson DJ, van der Horst C. Perceptions of the role of maternal nutrition in HIV-positive breast-feeding women in Malawi. J Nutr 2005;135:945–9.
14. Blum L, Pelto PJ, Pelto GH, Kuhnlein HV. Community assessment of natural food sources of vitamin A: Guidelines for an ethnographic protocol. Ottawa and Boston, MA, USA: International Nutrition Foundation for Developing Countries and International Development Research Centre, 1997.
15. Humphries S, Gonzales J, Jiminez J, Sierra F. Searching for sustainable land use practices in Honduras: Lessons

from a programme of participatory research with hillside farmers. Agricultural Research and Extension Network Paper 2000:104.
16. Miles MB, Huberman AM. Qualitative data analysis: An expanded sourcebook. Thousand Oaks, CA, USA: Sage Publications, 1994.
17. Patton MQ. Qualitative evaluation and research methods. Newbury Park, Calif, USA: Sage Publications, 1990.
18. Hamill PVV, Drizid TA, Johnson CL, Reed RB, Roche AF, Moore WM. NCHS growth curves for children birth–18 years. Vital and Health Statistics Series 11, No. 165. DHEW Publication No. (PHS) 78-1650. Hyattsville, MD, USA: US Department of Health, Education, and Welfare, Public Health Service, 1977.
19. Smale M, Phiri ADK. Institutional change and discontinuities in farmers' use of hybrid maize seed and fertilizer in Malawi: Findings from the 1996–97 CIMMYT MoALD survey. CIMMYT Economics Working Paper 98-01. Mexico City: CIMMYT, 1998.
20. Bezner Kerr R. Contested knowledge and disputed practice: Maize and groundnut seeds and child feeding in northern Malawi. PhD Dissertation, Department of Development Sociology, Cornell University, Ithaca, NY, USA, 2006.
21. WHO Collaborative Study Team on the Role of Breastfeeding on the Prevention of Infant Mortality. Effect of breastfeeding on infant and child mortality due to infectious diseases in less developed countries: A pooled analysis. Lancet 2000;355:451–5.
22. Sibeko L, Dhansay MA, Charlton KE, Johns T, Gray-Donald K. Beliefs, attitudes, and practices of breastfeeding mothers from a periurban community in South Africa. J Hum Lact 2005;21:31–8.
23. Simango C. Isolation of Escherichia coli in foods. Cent Afr J Med 1995;41:181–5.
24. Bahl R, Frost C, Kirkwood BR, Edmond K, Martines K, Martines J, Bhandari N, Arthur P. Infant feeding patterns and risks of death and hospitalization in the first half of infancy: Multicentre cohort study. Bull World Health Organ 2005;83:418–26.
25. Iliff PJ, Piwoz EG, Tavengwa NV, Zunguza CD, Marinda ET, Nathoo KJ, Moulton LH, Ward BJ, Humphrey JH; ZVITAMBO study group. Early exclusive breastfeeding reduces the risk of postnatal HIV-1 transmission and increases HIV-free survival. AIDS 2005;19:699–708.

A multicenter community study on the efficacy of double-fortified salt

Malavika Vinodkumar, S. Rajagopalan, I. P. Bhagwat, Sanjay Singh, Bipinkumar S. Parmar, Om Prakash Mishra, Shyam Sundar Upadhyay, N. B. Bhalia, and Shailesh R. Deshpande

Abstract

Background. *Iron and iodine deficiencies affect more than 30% of the world's population. Typical Indian diets contain adequate amounts of iron, but the bioavailability is poor. This serious limiting factor is caused by low intake of meat products rich in heme iron and intake of phytates in staple foods in the Indian diet, which inhibits iron absorption.*

Objective. *To test the stability of double-fortified salt (DFS) during storage and to assess its efficacy in improving the iron and iodine status of the communities.*

Methods. *The stability of both iodized salt and DFS during storage for a 2-year period was determined. The bioefficacy of DFS was assessed in communities covering three states of the country for a period of 1 year. This was a multicenter, single-blind trial covering seven clusters. The experimental group used DFS and the control group used iodized salt. The salts were used in all meals prepared for family members, but determination of hemoglobin by the cyanmethemoglobin method was performed in only two or three members per family, and not in children under 10 years of age (*n *= 393 and 436 in the experimental and control groups, respectively). The family size was usually four or five, with a male: female ratio of 1:1, consisting of two parents with two or three children. Hemoglobin was measured at baseline, 6 months (midpoint), and 12 months (endpoint). Urinary iodine was measured in only one cluster at baseline and endpoint. All the participants were dewormed at baseline, 6 months, and 12 months.*

Results. *The iron and iodine in the DFS were stable during storage for 2 years. Over a period of 1 year, there was an increase of 1.98 g/dL of hemoglobin in the experimental group and 0.77 g/dL of hemoglobin in the control group; the latter increase may have been due to deworming. The median urinary iodine changed from 200 µg/dL at baseline to 205 µg/dL at the end of the study in the experimental group and from 225 µg/dL to 220 µg/dL in the control group. There was a statistically significant (*$p < .05$*) improvement in the median urinary iodine status of subjects who were iodine deficient (urinary iodine < 100 µg/L) in both the experimental and the control groups, a result showing that DFS was as efficient as iodized salt in increasing urinary iodine from a deficient to sufficient status. There was a statistically significant increase (*$p < .05$*) in hemoglobin in all seven clusters in the experimental group compared with the control.*

Conclusions. *The iron and iodine in the DFS are stable in storage for 2 years. The DFS has proved beneficial in the delivery of bioavailable iron and iodine.*

Key words: Iron and iodine fortification, salt

Introduction

Iron is the second most abundant metal in the earth's crust, yet iron deficiency is a worldwide public health problem [1, 2]. Anemia leads to considerable morbidity, mortality, impaired immune status, and decreased productivity. Iron deficiency in infancy causes impairments in physical development and cognition that may be irreversible [3–5]. Among micronutrient deficiencies, iron and iodine deficiencies affect > 30 % of the global population [6].

Malavika Vinodkumar and S. Rajagopalan are affiliated with the Sundar Serendipity Foundation; I. P. Bhagwat was affiliated with BAIF Pune and is currently with UNICEF, New Delhi; Sanjay Singh is affiliated with the BAIF Institute of Rural Development, Allahabad, Uttar Pradesh; Bipinkumar S. Parmar is affiliated with the Gujarat Rural Institute for Socio-Economic Rehabilitation, Vadodara; Om Prakash Mishra is affiliated with the BAIF Institute of Rural Development, Chitrakoot, Uttar Pradesh; Shyam Sundar Upadhyay is affiliated with the BAIF Institute of Rural Development, Gonda, Uttar Pradesh; N. B. Bhalia is affiliated with the Gujarat Rural Institute for SocioEconomic Rehabilitation, Bharuch; Shailesh R. Deshpande is affiliated with BAIF Pune —all in India.

Please direct queries to the corresponding author: Malavika Vinodkumar, Sundar Serendipity Foundation, 6G Century Plaza, 560-562 Anna Salai, Teynampet, Chennai 600018, India; e-mail: vinodkumar_m_k@hotmail.com.

Typical Indian diets contain adequate amounts of iron, but the bioavailability of iron is poor because the intake of meat products rich in heme iron is low due to low socioeconomic status and also because of the presence of phytates that inhibit iron absorption in rice and wheat, the staple cereals of Indians. Thus, only 2% to 5% of the iron intake is absorbed, and it is not surprising that iron-deficiency anemia is widespread.

Among the public health approaches advocated for the control and prevention of iron deficiency are the distribution of supplements of medicinal iron and fortification of foods with a suitable iron compound. The former is recommended as a short-term measure to correct anemia, and the latter to improve the iron balance over a period of time and build up iron reserves. Since India already has a program to supply iodized salt, double fortification of salt with iron and iodine makes eminent sense.

Anemia is also a problem in the project areas of the BAIF Development Research Foundation (BAIF is the Bharatiya Agro Industries Foundation, one of India's oldest nongovernmental organizations, which has been involved in rural development for more than four decades). To address the problem on a larger scale, a field trial of double-fortified common salt was conducted in 2002–2004 by the BAIF Development Research Foundation. The Sundar Serendipity Foundation (SSF), Chennai, provided the double-fortified salt for the study.

Although there have been other studies on double-fortified salt in India [7] and by the Swiss Federal Institute Zurich [8] in Morocco and Côte d'Ivoire, it was felt that a multicenter study performed simultaneously in different parts of a vast country like India would throw light on the challenging task of combating anemia and iodine deficiency together.

The objective was to evaluate the use of double-fortified salt (DFS) as a public health measure in the control of anemia and iodine-deficiency disorders. The specific objectives were to test the stability of DFS in storage and provide DFS to selected families for 1 year in seven clusters, to determine the efficacy of salt in decreasing anemia and iodine deficiency disorders in this population.

Methods and materials

Production and supply of salt

Both DFS and iodized salt were obtained from SSF, Chennai. Twenty-two batches of iodized salt and DFS were prepared by SSF before the start of the study. These batches were transported to BAIF headquarters in Pune. They were then transported to the different study sites, where they were stored centrally and distributed on a monthly basis throughout the study. Control samples of the 22 batches prepared were used for the stability tests. The color of the DFS and iodized salt was tested using a reflectance meter, and there was no difference between the color of the DFS and iodized salt.

Salt was doubly fortified with ferrous sulfate monohydrate (Hiena Pharma, Mumbai) and potassium iodate (Calibre Chemicals, Mumbai). The ferrous sulfate was chelated in the laboratory of SSF with chelating agents, and the absorption promoter was added to enhance iron absorption. Ferrous sulfate was chelated with malic acid and sodium hexametaphosphate. The acidic pH was maintained by sodium dihydrogen phosphate, which served as an absorption promoter. The resulting chelated iron complex was white in color. The DFS contains 1,000 ppm of iron and 40 ppm of iodine, or 1 mg of iron and 40 µg of iodine per gram of salt. Our studies have shown that each person consumes about 10 g of salt per day. The DFS therefore provides about 10 mg of chelated iron and 400 µg of iodine per person per day. The iodine was encapsulated to prevent decomposition. Potassium iodate, the iodine compound, was coated in the SSF laboratory with an alkaline compound, food-grade sodium bicarbonate (P.D. Fine Chem, Bangalore), to keep it in an alkaline medium for stability. The potassium iodate was further encapsulated in cellulose acetate phthalate (GM Chemicals, Mumbai) and coated with a layer of silicone (Process Chemical Company, Mumbai) to provide heat resistance during cooking. The cellulose acetate phthalate coat protects the potassium iodate from the acidic environment of iron in the salt, thereby enhancing the shelf-life of iodine in the fortified salt as well as in the acidic medium of the stomach. Iodine is bioavailable and absorbed in the alkaline pH of the intestine, where the cellulose acetate phthalate coat disintegrates. Even though the iron is chelated, the environment of the salt is at an acidic pH, since the iron is not coated. The stability of iodine therefore is dependent on the cellulose acetate phthalate coat. In India it is mandatory for salt to contain iodine at a level of 30 ppm at the manufacturer's site.

The salt was packed in 1-kg bags and supplied to the families through self-help groups. Self-help groups are examples of community-based organizations at the lowest level. They are made up of 10 to 20 individuals, generally women. The idea is to meet regularly (at least every month), inculcate the habit of regular saving, and utilize the pooled money for individual needs as credit, with the repayment of loans contributing to increasing the pool. However, the self-help groups go beyond thrift-credit. They are platforms to discuss various issues relevant to the group and the village, including health and social issues. In fact, the groups play an important role in mobilizing the community, whether it is for village cleanliness, childhood immunization, de-addiction drives, or accessing and implementing government schemes. The enterprising self-help groups may start and manage income-generating activities, so it seemed prudent and consistent with their mission to entrust them with the distribution of the salt.

Storage stability studies

The stability of the iron and iodine contents in DFS and the iodine content in iodized salt were assessed for a period of 2 years. The control samples of the salts were stored at 30°C and 45% humidity in the SSF Laboratory, where the stability studies were performed. We chose 30°C and 45% humidity because they represent the average weather conditions of the sites where the study was conducted. Each batch of DFS produced was tested at 4-month intervals for iron and iodine, and each batch of iodized salt produced was tested at 4-month intervals for iodine, for a period of 2 years. The method of analysis of iron and iodine was according to the specifications of the Bureau of Indian Standards [9].

Bioefficacy study

The project areas were the seven clusters of the BAIF European Union Project. The multicenter single-blind study was carried out in three states of the country: Karnataka (three clusters—Tumkur, Uttar Kanada, and Dharwad), Gujarat (two clusters—Surat and Bharuch), and Uttar Pradesh (two clusters—Pratapgarh and Gonda). The clusters are administrative units for implementing the Transfer of Technologies for Sustainable Development project, which was funded by the European Union. Each cluster comprises a group of villages (an average of 20) covered under the project. For convenience, the clusters have been named after the districts where they are located. The members of the self-help groups were informed about the trial, and volunteer families were enrolled after informed oral consent had been obtained from the heads of the families. The study was approved by the Institutional Review Board of BAIF.

Climate of the seven clusters

Tumkur has a dry climate, with an annual rainfall of 550 mm, a maximum temperature of 35°C in the summer, and a minimum temperature of 18°–20°C in the winter. Uttar Kanada lies close to the western ghats. It is mountainous, with an annual rainfall of 2,500 mm during the monsoon season, which lasts from June to September. The maximum temperature is 30°C in the summer, and the minimum is 10°C in the winter. Dharwad is in the plains, with an annual rainfall of 1,000 mm. The summer temperature peaks to 40°C, and the winter temperature is around 18°–20°C. Gonda and Pratapgarh lie in the Gangetic plains. The summer temperature in Gonda peaks up to 45°C. Winters are very cold, with temperatures dropping to 1°–2°C. Winters are very foggy, making movement very difficult. Because of the cold conditions, finger-pricking for phlebotomy becomes very difficult. Therefore, the time of starting the study was chosen to avoid performing phlebotomy during the winter. Pratapgarh has scorching summers, with temperatures soaring to 45°C and winter temperatures dropping to 4°–5°C. Surat and Bharuch lie in the Narmada basin. The annual rainfall is 800–1,000 mm, summer temperatures are 40°–42°C, and winter temperatures are 18°–20°C.

Sample size and selection

Earlier studies had shown a rise in hemoglobin of about 0.5 to 1 g/dL after a year's consumption of DFS [10]. To calculate the sample size, we assumed an increase of hemoglobin of 0.5 g/dL, with SD of 1.0 and a 95% confidence interval, power of 80% using the formula $2^*(1.96 + 0.84)^2 * ((1)^2/(0.5)^2)$ and arrived at a sample size of 63 persons per group. Sample sizes of 63 persons per cluster in the experimental group and 63 persons per cluster in the control group were calculated for measurement of hemoglobin. Two to three members (preferably adults; children less than 10 years of age were excluded due to the difficulty of phlebotomy in young children and the reservations of parents of young children in consenting to the procedure) were included from each of the 30 families in the experimental group to constitute a sample size of 63 to 90 per cluster, and similarly 30 families with two or three members per family were included to constitute a sample size of 63 to 90 per cluster for the control. A total of seven clusters were selected.

The exact number of families in the cluster depended on the geographic distribution of the households. Some clusters had more than 60 families and some less. Surat and Bharuch, which were very close together, were considered as one unit. Since isolated villages were chosen for study, the sample size for hemoglobin estimation in Bharuch and Surat was around 30. Similarly, in Uttar Kanada, an isolated village was chosen, so the sample size for hemoglobin estimation was 28 in the experimental group. In the remaining clusters, the sample size was more than 60. The DFS and the iodized salt were supplied and consumed by all the members of the households, but the hemoglobin measurements were performed on only two or three members of the household.

Study design

Each cluster had an experimental area where DFS was supplied and a control area where plain iodized salt was supplied. In each cluster, two adjoining villages were selected randomly, and these formed the experimental and control groups. After the random selection process, it was determined that all clusters shared a similar economic background.

For a cluster, a village was a functional unit. In each village, self-help groups of women were identified, and volunteer families were listed. The salt was not provided free but was sold at the prevailing market price for common salt in the area. BAIF subsidized the remaining cost of the salt. Self-help groups were chosen to facilitate behavior change communication, the logistics of supply, and maintenance of records. Both the experimental and the control families were educated about anemia and the approach of using fortified salt in cooking all the meals of the family.

All members of the families in the experimental and control groups were dewormed with albendazole 400 mg at baseline, 6 months (midpoint), and 12 months (endpoint). This was essential because helminths compete for micronutrient absorption, and the intestinal tract had to be clear of worms for absorption of the nutrients.

The biochemical parameters assessed were hemoglobin and urinary iodine. Blood samples for hemoglobin analysis were collected at baseline, 6 months, and 12 months. Urinary iodine analysis was performed only in the Gujarat cluster at the beginning and the end of the trial in both the experimental and the control groups.

Blood collection, storage, and hemoglobin analysis

Fingerprick blood samples were obtained with accurately calibrated hemoglobin pipettes. Twenty microliters of blood from the fingerprick was collected and added to 5 mL of Drabkin's solution. The optical density was read with a portable field colorimeter within a few hours of collecting the blood in a central area in the village. Hemoglobin was determined by the cyanmethemoglobin method as described by Dacie and Lewis [11]. The same batch of hemoglobin standards was used to calibrate the colorimeter in all the clusters. Samples were analyzed by a study worker who was unaware of the previous results.

In all seven clusters, hemoglobin measurements were performed three times on 393 subjects in the experimental group and 436 in the control group. Twenty-six individuals in the experimental and 21 in the control group who were not present for all three rounds of hemoglobin analysis were excluded from statistical analysis.

Urinary iodine level was determined by the Pino modification of the Sandell Kolthroff reaction, using the portable colorimeter [12]. Random urine samples were collected by technicians from the households and analyzed within a few hours of collection.

Statistical analysis

Data were analyzed with SPSS version 11 and Microsoft Excel. ANOVA and *t*-tests were performed to determine the differences in hemoglobin over time and between groups. Median urinary iodine values were tested by the Wilcoxon signed-ranks test and the Mann-Whitney test.

Results

Benefits noted with the use of DFS were an increase in well-being and a decrease in menstrual problems in women. These changes were assessed by questionnaires administered to the heads of households and the women in each family. Overall, the people would prefer to consume DFS if it were available at an affordable price.

Characteristics and stability of iodized salt and DFS

Storage quality

No color changes in the salts were observed during transport or storage. Although the study lasted for only 1 year in each cluster, the entire study in all three states of the country took 2 years to complete because the study did not start simultaneously in all seven clusters. Hemoglobin analysis was performed with the same colorimeter in all seven clusters. Thus, the study started in each cluster after baseline hemoglobin measurements had been performed. The salt, however, was prepared before the start of the study. Since the entire study lasted for 2 years, analysis of stability for a period of 2 years was performed. The stability of iodine in iodized salt and of iron and iodine in DFS is given in **table 1.** Iron was found to be stable. ANOVA found no significant difference ($p > .05$) between the stability of iodine in DFS and in iodized salt.

Taste

There were no complaints regarding taste. People noted that the amount of salt to be added to food was slightly less than usual. The right amount was mastered over a period of time. People also observed that food turned slightly sour when kept for more than 6 hours. This may be due to the absorption promoters. The benefits of the salts were assessed by questionnaires addressed to the heads of the households and the women in the families.

Results of efficacy study

The mean (± SD) baseline hemoglobin was 10.34 ± 2.56 g/dL in the experimental group and 10.29 ± 2.62 g/dL in the control group, and there was no significant difference between the groups. After 6 months of

TABLE 1. Stability of iodine in iodized salt and of iron and iodine in double-fortified salt at 30°C and 45% relative humidity [a]

Measurement	Baseline	4 mo	8 mo	12 mo	16 mo	20 mo	24 mo
Iodine in iodized salt[b]	41.30 ± 3.08	39.80 ± 2.48	37.50 ± 1.86	38.15 ± 2.27	32.60 ± 3.14	31.10 ± 2.89	29.20 ± 2.96
Iodine in double-fortified salt[b]	40.90 ± 2.35	38.80 ± 3.16	37.10 ± 2.43	38.01 ± 2.43	31.90 ± 2.98	30.50 ± 2.57	27.70 ± 3.61
Iron in double-fortified salt[c]	1,026.70 ± 35.20	1,014.40 ± 27.18	1,013.60 ± 17.60	1,008.86 ± 33.80	1,008.13 ± 32.90	1,006.00 ± 23.33	996.20 ± 35.16

a. The values are means ± SD of 22 batches prepared for the study. Iron and iodine concentrations are given in parts per million. Differences are considered significant if $p < .05$.
b. No significant differences between the stability of iodine in double-fortified salt and iodized salt for the entire period.
c. No significant change in iron levels throughout the study.

intervention, the mean hemoglobin in the experimental group increased to 11.30 ± 2.21 g/dL, and after 1 year it further increased to 12.32 ± 1.93 g/dL. These increases at 6 months and 1 year were statistically significant ($p < .05$). The increase of 1.98 g/dL in 1 year could be due to the use of DFS and deworming. In the control group, the mean hemoglobin increased from 10.29 ± 2.62 g/dL at baseline to 10.61 ± 2.47 g/dL after 6 months and to 11.06 ± 2.59 g/dL after 1 year, and the increase from 6 months to one year was statistically significant. The increase of 0.77 g/dL in the control group in 1 year could be due to deworming, since the elimination of the helminths could have resulted in better absorption of the nutrients from the diets (**table 2**).

TABLE 2. Changes in hemoglobin concentration in groups receiving double-fortified salt (experimental group) and iodized salt (control group)[a]

Time	Experimental group (n = 393)	Control group (n = 436)
Baseline	10.34 ± 2.56[b]	10.29 ± 2.62
Midpoint (6 mo)	11.30 ± 2.21[b,c]	10.61 ± 2.47[c,d]
Endpoint (1 yr)	12.32 ± 1.93[b,c]	11.06 ± 2.59[c,d]

a. Hemoglobin concentrations are expressed as means ± SD (g/dL). A difference is considered significant if $p < .05$.
b. Significant improvement in the experimental group from baseline to midpoint and from midpoint to endpoint.
c. Significant improvement in the experimental group compared with control.
d. Significant improvement in the control group from midpoint to endpoint.

The increase in hemoglobin of 1.98 g/dL in the experimental group is significantly ($p < .05$) greater than the increase of 0.77 g/dL in the control group. The increase of 0.77 g/dL of hemoglobin in the control group could be attributed to deworming, and if this value is subtracted from 1.98 g/dL, the increase in hemoglobin in the experimental group (due to deworming and intake of fortified salt), then the increase of hemoglobin of 1.21 g/dL could be attributed to the bioavailability of iron from the fortified salt.

The increase in hemoglobin is significantly greater ($p < .05$) in the experimental group than the control group in all seven clusters. The increase in hemoglobin

TABLE 3. Changes in hemoglobin concentration in groups receiving double-fortified salt (experimental group) and iodized salt (control group) according to cluster[a]

Cluster	Experimental group				Control group			
	N	Baseline	Endpoint (1 yr)	Change	N	Baseline	Endpoint (1 yr)	Change
Uttar Kanada	28	9.23 ± 2.30[b]	10.08 ± 2.26[b]	0.85 ± 0.40[c]	47	9.34 ± 2.26	9.32 ± 2.46	−0.02 ± 0.60[c]
Dharwad	59	10.63 ± 2.96[b,d]	11.55 ± 2.65[b,e]	0.93 ± 0.43[c]	81	9.35 ± 3.10[d]	9.40 ± 3.13[e]	0.05 ± 0.47[c]
Tumkur	90	9.47 ± 2.74[b]	13.04 ± 1.21[b,e]	3.58 ± 2.73[c]	99	9.44 ± 2.31[f]	11.97 ± 2.08[e,f]	2.53 ± 1.86[c]
Pratapgarh	81	12.49 ± 1.40[b]	13.68 ± 1.03[b,e]	1.19 ± 0.67[c]	106	12.39 ± 1.38[f]	12.64 ± 1.33[e,f]	0.25 ± 0.62[c]
Gonda	88	9.77 ± 2.02[b,d]	11.91 ± 1.40[b]	2.13 ± 1.10[c]	69	11.06 ± 1.98[d,f]	11.57 ± 1.57[f]	0.51 ± 0.84[c]
Surat and Bharuch	47	9.31 ± 1.75[b,d]	11.13 ± 1.36[b,e]	1.82 ± 1.10[c]	34	7.94 ± 1.90[d,f]	8.34 ± 1.84[e,f]	0.40 ± 0.47[c]
Total	393				436			

a. Hemoglobin concentrations are expressed as means ± SD (g/dL). A difference is considered significant if $p < .05$.
b. Endpoint hemoglobin significantly higher than baseline hemoglobin in experimental group.
c. Change in hemoglobin at endpoint significantly more in experimental group than in control group.
d. Baseline hemoglobin significantly different in experimental and control groups.
e. Endpoint hemoglobin significantly higher in experimental group than in control group.
f. Endpoint hemoglobin significantly higher than baseline hemoglobin in control group.

in the experimental groups varies from cluster to cluster, from a high of 3.58 g/dL in the Tumkur cluster to a low of 0.85 g/dL in the Uttar Kanada cluster. In the control groups, the highest increase in hemoglobin was in the Tumkur cluster (2.53 g/dL) and the lowest was a decrease of 0.02 g/dL in the Uttar Kanada cluster (**table 3).** Although the experimental and control groups were selected from similar communities in each cluster, with the same dietary habits and belonging to the same socioeconomic groups, there were significant ($p < .05$) differences in baseline hemoglobin status between the experimental and control groups in Dharwad, Gonda, Surat, and Bharuch. In the other three clusters, Uttar Kanada, Tumkur, and Pratapgarh, there were no significant differences in mean baseline hemoglobin between the experimental and control groups (**table 3).** Baseline hemoglobin also varied from 9.23 g/dL in Uttar Kanada to 12.49 g/dL in Pratapgarh. These differences may be due to wide variations in food habits and environmental sanitation between different states of India. Despite these baseline differences in different states, in all age categories and in both males and females, there was a significantly higher increase in hemoglobin in the experimental group than in the control group ($p < .05$) (**table 4).**

TABLE 4. Changes in hemoglobin concentration from baseline to 1 year in groups receiving double-fortified salt (experimental group) and iodized salt (control group) according to sex and age[a]

Sex	Age (yr)	Experimental group		Control group	
		N	Change	*N*	Change
Female	10–18	30	2.09 ± 1.58	28	0.56 ± 1.00
	19–30	79	2.34 ± 1.74	70	0.35 ± 1.10
	31–45	78	1.88 ± 1.87	130	0.81 ± 1.53
	46–65	38	2.31 ± 2.14	50	1.43 ± 1.88
Male	10–18	26	1.94 ± 2.17	40	0.83 ± 1.33
	19–30	61	1.49 ± 1.32	30	0.75 ± 1.50
	31–45	45	1.82 ± 1.81	58	0.66 ± 1.26
	46–65	36	1.98 ± 2.10	30	0.94 ± 1.52
Total		393		436	

a. Hemoglobin concentrations are expressed as means ± SD (g/dL). There was a significantly ($p < .05$) higher increase in hemoglobin concentration in the experimental group than in the control group for both males and females of all age groups.

Results of urinary iodine studies

Urinary iodine studies were done only in Gujarat in the Bharuch and Surat clusters. This was because analysis of urinary iodine is much more difficult than analysis of hemoglobin, and we were able to train technicians to do the urinary iodine analysis in Gujarat only. Urinary iodine analysis was performed in 47 adults in the experimental group and 34 adults in the control group on whom hemoglobin tests were also performed. The median urinary iodine at baseline was 200 µg/L in the experimental group and 225 µg/L in the control group. There was no significant difference ($p > .05$) between the experimental and control groups at baseline.

At endpoint, urinary iodine increased to 205 µg/L in the experimental group and was reduced marginally to 220 µg/L in the control group. Thus, median iodine values were maintained at around 200 µg/L throughout the study in both the experimental and control groups. This could imply that the iodine from the DFS is absorbed in the same manner as the iodine from iodized salt. Thus, the bioavailability of iodine from DFS and iodized salt is similar (**table 5**).

TABLE 5. Urinary iodine excretion values in groups receiving double-fortified salt (experimental group) and iodized salt (control group) in Gujarat-Surat and Bharuch clusters according to baseline iodine status[a]

Iodine status	Experimental group			Control group		
	N	Baseline	Endpoint (1 yr)	*N*	Baseline	Endpoint (1 yr)
Deficient	7	75 (50–85)[d,h]	225 (120–600)[e,h]	14	50 (35–85)[d,h]	220 (95–340)[e,h]
Sufficient	40	200 (100–600)[f]	195 (70–500)[g]	20	325 (100–600)[f]	207.5 (60–600)[g]
Whole group	47	200 (50–600)[b]	205 (70–600)[c]	34	225 (35–600)[b]	220 (60–600)[c]

a. Urinary iodine excretion values are expressed as median (range) in micrograms per liter. Subjects with baseline urinary iodine values more than and less than 100 µg/L were considered iodine sufficient and deficient, respectively. Differences are considered significant if $p < .05$ according to the Mann-Whitney test or the Wilcoxon signed-ranks test.
b. No significant difference between experimental and control groups at baseline.
c. No significant difference between experimental and control groups at endpoint.
d. Significant difference in baseline iodine status between the experimental and control groups of subjects who were iodine deficient at baseline, with the control group having a lower urinary iodine concentration than the experimental group.
e. No significant difference in endpoint iodine status between the experimental and control groups of subjects who were iodine deficient at baseline.
f. Significant difference in baseline iodine status between the experimental and control groups of subjects who were iodine sufficient at baseline, with the control group having a higher urinary iodine concentration than the experimental group.
g. No significant difference in endpoint iodine status between the experimental and control groups of subjects who were iodine sufficient at baseline.
h. Significant increase from baseline to endpoint among subjects who were iodine deficient at baseline in both the experimental and the control groups.

In both the experimental and the control groups, the absorption of iodine, which is reflected by the increase in urinary iodine excretion, was significantly higher in the subgroup of those who were iodine deficient, i.e., with a median urinary iodine excretion of less than 100 μg/L at baseline. At the end of the study, in both the experimental and the control groups, median urinary iodine levels increased from the deficient level of less than 100 μg/L to the sufficient level of more than 100 μg/L (**table 5).** There was no significant difference ($p > .05$) in the endpoint status between the experimental and control groups in those subjects who were iodine deficient at baseline. Thus, iodized salt and DFS were equally effective in changing the iodine-deficient status to an iodine-sufficient status in both the experimental and the control groups.

There was a significant difference ($p < .05$) between the experimental and the control groups in the baseline status of those who were iodine sufficient at baseline, with the control group having a higher median urinary iodine than the experimental group. There was no significant difference ($p > .05$) between the experimental and control groups after 1 year of intervention in those subjects who were iodine sufficient at baseline. This means that iodized salt and DFS were equally effective in maintaining urinary iodine status in subjects who were iodine sufficient at baseline. At the endpoint, there was only one subject in the experimental group and one subject in the control group whose urinary iodine was less than the sufficient status of 100 μg/L. The reason for this is not known.

Discussion

The literature survey on DFS showed that maintaining the stability of iodine in the presence of iron is difficult [7]. The bioavailability of iron is also in question. In the study of DFS conducted by the Indian National Institute of Nutrition, there was a drop in hemoglobin of the children in both the experimental and the control groups [7]. Earlier studies used only inorganic iron compounds without biopromoters [7, 13]. Earlier studies also used ferrous sulfate encapsulated with hydrogenated soybean oils. This DFS developed a yellow coloration when the moisture content of the salt was 3% to 4%, even though the mean hemoglobin increased by 14 g/L [14]. In another study testing the stability of 16 forms of encapsulated iron in salts of north and west Africa, the authors found that encapsulated ferrous iron caused unacceptable color changes to salt. The authors felt that this might be because current encapsulating technology uses hydrogenated plant oils that do not sufficiently prevent moisture penetration, and iron solubility and capsule integrity are further compromised by mechanical abrasion during salt mixing. The capsules also melt at 45° to 50°C and may cause unwanted sensory changes during food preparation [8]. Other studies have used iron sources such as micronized ground ferric pyrophosphate (FePP) and iodine with no biopromoter added [15]. In our study we used ferrous sulfate as the iron source, but we chelated the iron and added an absorption promoter. Because the iron is well chelated, it does not react with the salt to produce discoloration and does not react with the food preparations to produce sensory changes. If a high-quality salt is used with low calcium and magnesium content, the chelated iron compound can be directly added to the salt and no microencapsulation is required, as was done in this study. However, if iron has to be added to salt with high magnesium, calcium, or moisture content, then microencapsulating the iron compound with compounds like glyceryl stearate or edible waxes is preferable. Thus, we feel that chelation of the ferrous sulfate not only increases its bioavailability but also prevents the interaction of ferrous sulfate with the salt and the food that produces yellow colorations in the salt during storage or color changes in food preparations.

The Micronutrient Initiative developed a DFS containing potassium iodide coated with maltodextrin and ferrous fumarate [16]. However, nonencapsulated ferrous fumarate added to low-grade salt from developing countries produced unacceptable dark-brown color changes [8]. The DFS developed by the Indian National Institute of Nutrition (NIN) uses ferrous sulfate and sodium hexametaphosphate without microencapsulation, and the stability of the iodine is dependent on the quality of salt. In the presence of magnesium chloride as an impurity, the salt lost a significant amount of iodine [17]. In field trials of the NIN DFS, one study found no overall benefit on hemoglobin concentrations, whereas in a second study, the hemoglobin concentrations decreased significantly in both the DFS and the iodized salt groups, but to a lesser degree in the DFS group [7]. This may be because the iron was not chelated and no absorption promoters were added.

We used chelated ferrous sulfate with biopromoters to ensure the maximum bioavailability of iron to enable bioabsorption of iron even in the presence of phytates, which are present in abundance in the Indian diet. We have shown in our earlier bioefficacy studies of DFS fortified with iron and iodine that there is an increase in hemoglobin among tea pickers using fortified salt, along with an improvement in their productivity [10]. We feel that chelated ferrous sulfate has a higher bioavailability than ferrous sulfate but without the problems caused by ferrous sulfate, such as coloration of the salt or food during cooking.

The quantity of salt consumed by a person per day is about 10 g. The fortified salt contains 10 mg of iron per 10 g of salt. Thus, the use of DFS in all the food preparations ensured a supply of small quantities of iron throughout the day. The types of food consumed in the three states are quite varied. In Uttar Pradesh and Gujarat, wheat is the predominant staple, whereas in Karnataka, the main staple is rice. However, there was a statistically significant improvement in hemoglobin in all the seven clusters. This may be because the iron was delivered in repeated small doses throughout the day, and fractional absorption of nonheme iron increases with decreasing dose [18]. Moreover, the salt has chelated ferrous sulfate with biopromoters that enhance the absorption of iron not only from the salt, but also from the food, as seen in other fortification studies that have used biopromoters or chelators [19–23]. Deworming alone may have caused an increase in hemoglobin by reducing blood loss and increasing iron absorption.

Malaria was not a major problem in any of the study areas, but infestation with helminths such as ascaris may be quite common, and this may be the reason for the increase in iron status in the control group where deworming alone caused an increase in hemoglobin status. The degree of infestation by helminths may be different in the seven clusters, but it might have been highest in the Tumkur cluster, where there was an increase in hemoglobin of 2.5 g/dL in the control group. This study also suggests the importance of deworming in studies, especially in rural areas, as pointed out in other studies [24, 25].

DFS is also an ideal vehicle for iodine. In our study we found no statistically significant differences between iodized salt and DFS in losses of iodine in storage. The potassium iodate in iodized salt was not microencapsulated. The potassium iodate in the DFS was microencapsulated to increase the shelf-life of iodine in the salt and to increase iodine bioavailability in the human system. The finding of no significant difference between iodized salt and DFS in loss of iodine during storage shows that the microencapsulation of iodine in DFS has done its job in protecting the potassium iodate from the harsh effects of iron, and that thus microencapsulation has prevented iodine losses. Similarly, the absorption of iodine from iodized salt and DFS was very similar, as seen from the urinary iodine changes in both the iodized salt group and the DFS group.

After use of the salt for 1 year, which included the rainy season, 98% to 100 % of the households rated the taste and color of DFS as acceptable. In fact, since the study was single blinded, the families did not know whether they were receiving iodized salt or DFS. There was no perceptible difference between iodized salt and DFS.

The cost of the chelated iron complex used in 1 kg of salt was 5 US cents. The cost of 1 kg of salt was 5 US cents. The microencapsulated potassium iodate used in 1 kg of salt cost 0.3 US cent. The total cost of the DFS per kilogram was 10.3 US cents. The cost of uncoated potassium iodate without microencapsulation in 1 kg of iodized salt is 0.1 US cent. The increase in cost of potassium iodate due to coating and microencapsulation is only 0.2 US cent.

In this study, we used salt of high purity and microencapsulated the iodine in DFS. However, the same iron complex can be used in salt of lower purity if the iron complex is also microencapsulated. We observed a significant improvement in hemoglobin status in the group using DFS among all age groups and among both males and females, which demonstrates iron availability. Iodine bioavailability is demonstrated by the improvement in urinary iodine. Thus, we feel that DFS with chelated iron compounds can be used as an effective strategy to combat micronutrient malnutrition in developing countries.

Acknowledgments

We thank Sundar Serendipity Foundation for providing the DFS used in this study and Mr. M. Sankarapandian of the Gandhigram Institute of Rural Health and Family Welfare Trust for statistical guidance.

References

1. World Health Organization(2000). Malnutrition. The global picture. Geneva: WHO.
2. United Nations Children's Fund.(2000). The state of the world's children 2000. New York: UNICEF.
3. Lozoff B, Jimenez E, Wolf AW. Long term developmental outcome of infants with iron deficiency. N Engl J Med 1991;325:6876–94.
4. Grantham-McGregor S, Ani C. A review of studies on the effect of iron deficiency on cognitive development in children. J Nutr 2000;131(2-S2):649S–66S; discussion 666S–68S.
5. Hautvast JLA, Tolbloom JJM, Kafwembe EM, Musonda RM, Mwanakasale V, van Staveren WA, van't Hof MA, Sauerwein RW, Willems JL, Monnens LAH. Severe linear growth retardation in rural Zambian children: The influence of biological variables. Am J Clin Nutr 2000;71:550–9.
6. WHO/UNICEF/UNU. IDA: Prevention, assessment and control. Report of a joint WHO/UNICEF/UNU consultation. Geneva: WHO, 1998:1–9.
7. Nair KM, Brahmam GN, Ranganathan S, Vijayaraghavan K, Sivakumar B, Krishnaswamy K. Impact evaluation of iron and iodine fortified salt. Indian J Med Res 1998; 108:203–11.
8 Wegmuller R, Zimmermann MB, Hurrell RF. Dual fortification of salt with iodine and encapsulated iron compounds: Stability and acceptability testing in Morocco and Côte d'Ivoire. J Food Sci 2003;68:2129–35.

9. Bureau of Indian Standards. Specification for common salt—Iron fortified. Specification IS 12981:1991. Specification for iodised salt, IS 7224:1985.
10. Rajagopalan S, Vinodkumar M. Effect of salt fortified with iron and iodine on the hemoglobin levels and productivity of tea pickers. Food Nutr Bull 2000;21:323–9.
11. Dacie JV, Lewis SM. Practical haematology, 8th ed. Edinburgh: Churchill Livingstone, 1995.
12. Pino S, Fang SL, Braverman LE. Ammonium persulfate: A safe alternative oxidizing reagent for measuring urinary iodine. Clin Chem 1996;42:239–43
13. Mannar MGV, Diosady LL. Double fortification of salt with iron and iodine. In: The Micronutrient Initiative,ed. Food fortification to end micronutrient malnutrition. Ottawa: The Micronutrient Initiative, 1998:89–94.
14. Zimmermann MB, Zeder C, Chaouki N, Saad A, Torresani T, Hurrell RF. Dual fortification of salt with iodine and microencapsulated iron: A randomized, double-blind, controlled trial in Moroccan school children. Am J Clin Nutr 2003;77:425–32.
15. Wegmuller R, Camara F, Zimmermann MB, Adou P, Hurrell RF. Salt dual-fortified with iodine and micronized ground ferric pyrophosphate affects iron status but not hemoglobin in children in Côte d'Ivoire. J Nutr 2006; 136:1814–20.
16. Sattarzadeh M, Zlotkin SH. Iron is well absorbed by healthy adults after ingestion of double-fortified (iron and dextran-coated iodine) table salt and urinary excretion is unaffected. J Nutr 1999;129:117–21.
17. Lofti M, Mannar MGV, Merx RJHM, van den Heuvel PN. Micronutrient fortification of foods. Ottawa: The Micronutrient Initiative, 1996:81–2.
18. Skikne B, Baynes RD. Iron absorption. In: Brock JH, Halliday JW, Pippard MJ, Powell LW, eds. Iron metabolism in health and disease. London: Saunders, 1993:152–87.
19. Walter T, Olivares M, Hertrampf E. Field trials of food fortification with iron: The experience of Chile. In: Lonnerdal B, ed. Iron metabolism in infants. Boca Raton, FL, USA: CRC Press, 1990:127–55.
20. Viteri FE, Alvarez E, Batres R, Torun B, Pineda O, Mejia LA, Sylvi J. Fortification of sugar with iron sodium ethylenediaminotetracetate (FeNaEDTA) improves iron status in semirural Guatemalan populations. Am J Clin Nutr 1995;61:1153–63.
21. Walter T, Pino P, Pizzaro F, Lozoff B. Prevention of iron-deficiency anemia: Comparison of high- and low-iron formulas in term healthy infants after six months of life. J Pediatr 1998;132:635–40.
22. Ballot DE, MacPhail AP, Bothwell TH, Gillooly M, Mayet FG. Fortification of curry powder with NaFe(111)EDTA in an iron-deficient population: Report of a controlled iron-fortification trial. Am J Clin Nutr 1989;49:162–9.
23. Thuy PV, Berger J, Davidsson L, Khan NC, Lam NT, Cook JD, Hurrell RF, Khoi HH. Regular consumption of NaFeEDTA-fortified fish sauce improves iron status and reduces the prevalence of anemia in anemic Vietnamese women. Am J Clin Nutr 2003;78:284–90.
24. Olsen A, Thiong'o FW, Ouma JH, Mwaniki D, Magnussen P, Michaelsen KF, Friis H, Geissler PW. Effects of multimicronutrient supplementation on helminth reinfection: A randomized, controlled trial in Kenyan school children. Trans R Soc Trop Med Hyg 2003;97:109–14.
25. Taylor M, Jinabhai CC, Couper I, Kleinschmidt I, Jogessar VB. The effect of different anthelmintic treatment regimens combined with iron supplementation on the nutritional status of schoolchildren in KwaZulu-Natal, South Africa: A randomized controlled trial. Trans R Soc Trop Med Hyg 2001;95:211–6.

Stability of iodine in salt fortified with iodine and iron

Srinivasaiyengar Ranganathan, Madhu G. Karmarkar, Muddepaka Krupadanam, Ginnela N. V. Brahmam, Mendhu Vishnuvardhana Rao, Kamasamudram Vijayaraghavan, and Bhattriprolu Sivakumar

Abstract

Background. *Determining the stability of iodine in fortified salt can be difficult under certain conditions. Current methods are sometimes unreliable in the presence of iron.*

Objective. *To test the new method to more accurately estimate iodine content in double-fortified salt (DFS) fortified with iodine and iron by using orthophosphoric acid instead of sulfuric acid in the titration procedure.*

Methods. *A double-blind, placebo-controlled study was carried out on DFS and iodized salt produced by the dry-mixing method. DFS and iodized salt were packed and sealed in color-coded, 0.5-kg, low-density polyethylene pouches, and 25 of these pouches were further packed and sealed in color-coded, double-lined, high-density polyethylene bags and transported by road in closed, light-protected containers to the International Council for the Control of Iodine Deficiency Disorders (ICCIDD), Delhi; the National Institute of Nutrition (NIN), Hyderabad; and the Orissa Unit of the National Nutrition Monitoring Bureau (NNMB), Bhubaneswar. The iodine content of DFS and iodized salt stored under normal room conditions in these places was measured by the modified method every month on the same prescribed dates during the first 6 months and also after 15 months. The iodine content of DFS and iodized salt stored under simulated household conditions was also measured in the first 3 months.*

Results. *After the color code was broken at the end of the study, it was found that the DFS and iodized salt stored at Bhubaneswar, Delhi, and Hyderabad retained more or less the same initial iodine content (30–40 ppm) during the first 6 months, and the stability was not affected after 15 months. The proportion of salt samples having more than 30 ppm iodine was 100% in DFS and iodized salt throughout the study period. Daily opening and closing of salt pouches under simulated household conditions did not result in any iodine loss.*

Conclusions. *The DFS and iodized salt prepared by the dry-mixing method and stored at normal room conditions had excellent iodine stability for more than 1 year.*

Key words: Double-fortified salt, iodine-deficiency disorders, iodized salt, iodine stability, iron-deficiency anemia, modified orthophosphoric acid method

Introduction

The control of micronutrient deficiencies by food fortification is one of the most significant developments in recent years. Probably no other technology available today offers such a wide scope to improve the health and nutritional status of people in the most cost-effective way [1, 2]. India has made rapid progress in this regard and has been successfully using low-cost technologies available for the production of iodized salt [3, 4] and iron-fortified salt [5, 6] in the country.

To tackle simultaneously the iodine-deficiency disorders and iron-deficiency anemia, iron- and iodine-fortified, double-fortified salt (DFS) was developed with refined common salt (100%); potassium iodate, KIO_3 (0.0067%); ferrous sulfate heptahydrate, $FeSO_4.7H_2O$ (0.508%); and sodium hexametaphosphate (1%) to provide simultaneously about 40 μg of iodine and 1,000 μg of iron per gram of DFS [7]. Sodium hexametaphosphate (SHMP) is a permitted food additive [8] and is extensively used in the food industry. Scientific evaluation of the large-scale production and sensory acceptability [9], ultrastructure [10], biosafety of long-term

S. Ranganathan, M. Krupadanam, G. N. V. Brahmam, M. Vishnuvardhana Rao, K. Vijayaraghavan, and B. Sivakumar are affiliated with the National Institute of Nutrition (Indian Council of Medical Research), Hyderabad, India; M. G. Karmarkar is affiliated with the International Council for Control of Iodine Deficiency Disorders, All India Institute of Medical Sciences, New Delhi, India.

Please direct queries to the corresponding author: S. Ranganathan, Head, Isotope Division, National Institute of Nutrition (Indian Council of Medical Research), Hyderabad 500 007, India; e-mail: drrangan@rediffmail.com.

consumption [11], impact, and efficacy [12-15] of DFS has been done.

When the iodine content of DFS is estimated by conventional iodometric titration [16] with the use of sulfuric acid, several problems may occur. There can be a wide variation in the iodine content of duplicate analyses of the same samples, and low iodine values have also been observed in freshly prepared DFS (8–31 ppm), although the expected iodine content was about 40 ppm. There was no consistency in the iodine content estimated over a period of time (10.1–22.6 ppm). In some batches, very high iodine content (50 –70 ppm) was also obtained for the same sample of DFS. Lack of consistency in the iodine content of DFS created a lot of confusion regarding the stability of iodine in the multicenter study of DFS [17].

However, more detailed studies were carried out to exclude any undesirable element in DFS so as to ensure the primary objective of providing enough iodine and iron through DFS to achieve the goal of controlling iodine-deficiency disorders and iron-deficiency anemia. During this pursuit, a modified method using orthophosphoric acid was developed to solve the problems [18], and this method was used to test the iodine stability of DFS at two different laboratories (Delhi and Hyderabad) simultaneously on the same predetermined dates, ensuring uniform conditions of storage, duration of storage, and analysis. Loss of iodine, if any, was also measured on a weekly basis in salt pouches stored under simulated domestic conditions.

Materials and methods

Production

One-half metric ton each of DFS, iodized salt from refined salt (IRS), and iodized salt from ordinary common salt (IOS) were produced in the salt factory of M/S Prince International at Bhubaneswar, according to the dry-mixing process prescribed for iodized salt [3] and DFS [9]. M/S. Sahayamatha Salterns (P) Ltd., Tuticorin, supplied the refined salt, and M/S. Mohapatra Scientific Supply Syndicate, Bhubaneswar, supplied ordinary common salt, SHMP, ferrous sulfate, potassium iodate, low-density polyethylene (LDPE) pouches, and high-density polyethylene (HDPE) bags. Food-grade chemicals were used for the production of DFS and iodized salt. The pH of a 5% aqueous solution of SHMP was 6.0, and its purity determined by its P_2O_5 content was 68%. Both of these values were well within the prescribed limits for SHMP for use in DFS. The refined salt (99.5% NaCl) was white with an average crystal diameter of 100 μm in and contained 0.01% magnesium, 0.01% calcium, 0.05% sulfate, 0.14% moisture, and 0.01% insoluble residue. The ordinary common salt (96.0% NaCl) was off-white with an average crystal diameter of 500 μm in and contained 0.10% magnesium, 0.18% calcium, 0.50% sulfate, 3.8% moisture, and 0.80% insoluble residue.

After estimation of the initial iron content of DFS and the initial iodine content of DFS, IRS, and IOS, the three salts were packed and sealed in color-coded 0.5-kg LDPE pouches (white, orange, or yellow) in the salt factory by the factory staff. Twenty-five LDPE pouches were further packed and sealed in double-lined HDPE bags of the same colors. The managing director of the salt factory gave the color code to ensure blinding. The key to the code was handed over to the director of the National Institute of Nutrition (NIN) in a sealed cover for safe custody and was broken at the end of the study. The investigators of the two laboratories were blinded to the type of salt until the color code was broken at the completion of the study.

Transportation

The LDPE salt pouches in HDPE bags, placed in closed containers protected from light, were transported from the factory by road and reached NIN, Hyderabad (17 HDPE bags per color × 3 = 51 HDPE bags containing 1,275 LDPE pouches), and the laboratory of the International Council for the Control of Iodine Deficiency Disorders (ICCIDD), New Delhi (17 HDPE bags per color × 3 = 51 HDPE bags containing 1,275 LDPE pouches), in a week. The remaining HDPE bags (6 HDPE bags per color × 3 = 18 HDPE bags containing 450 LDPE pouches) were sent to the Orissa Unit of the National Nutrition Monitoring Bureau (NNMB) at the Regional Medical Research Centre, Bhubaneswar, to assess the stability of iodine in a coastal environment.

Storage

The HDPE bags containing 25 LDPE salt pouches per HDPE bag were stored in rooms in Bhubaneswar, Hyderabad, and New Delhi. The average maximum and minimum temperatures were 40° and 23°C during February, March, April, and May (summer), 32° and 20°C during June, July, August, and September (rainy season), and 26° and 10°C during October, November, December, and January (winter). The average relative humidity was 18% in the summer, 90% in the rainy season, and 49% in the winter. The average rainfall was about 500 mm during the rainy season, 45 mm during the summer, and a trace in the winter.

Sampling and testing of fortified salts for iodine stability

Sampling of DFS and iodized salt and estimation of iodine were performed simultaneously at the NIN and ICCIDD laboratories on the same predetermined dates to ensure uniform conditions of storage, duration of storage, and analysis. The investigators of the two laboratories did not know the type of salt until the color code was broken at the end of the study and were measuring the iodine content based on the color code of the LDPE pouch. Every month during the first 6 months on the predetermined date, one LDPE pouch per HDPE bag (from 25 LDPE pouches) was randomly picked up and the iodine content was estimated in each LDPE pouch by taking duplicate aliquots of each sample. On the predetermined date every month, the staff of the NNMB Orissa Unit dispatched randomly picked salt samples (LDPE pouches) to the NIN and ICCIDD laboratories, and the salt samples were analyzed simultaneously in the two laboratories. Loss of iodine, if any, under simulated household conditions was measured in a sub-sample of each category of fortified salt. For this purpose, six LDPE pouches of each category of salt (white, orange, or yellow) were drawn randomly, and after estimation of the initial iodine content, the salt pouches were closed with rubber bands and stored. Subsequent estimation of iodine in these salt pouches was performed on the 7th, 14th, and 21st days of every month during the first 3 months. These salt pouches were opened and closed daily, as salt is generally handled in the households.

Standardization of iodine estimation method in DFS and iodized salt

Following the conventional iodometric titration used for the estimation of iodine in iodized salt, 10 g of iodized salt was dissolved in 50 mL of distilled water, and 1 mL of 2 N sulfuric acid (H_2SO_4) was added followed by the addition of 5 mL of 10% potassium iodide (KI). The reaction mixture was kept in the dark for 10 minutes, and the iodine liberated was estimated by titration with 0.005 M sodium thiosulfate ($Na_2S_2O_3$) using a starch indicator near the end point of titration (**table 1**).

Kolthoff and Belcher have recommended the use of orthophosphoric acid (H_3PO_4) instead of H_2SO_4 for iodine estimation in the presence of iron to overcome any interference from iron [19]. According to Kolthoff and Belcher, iodine should be liberated only after sufficient iodide (KI) is present in the solution to minimize the loss of iodine by volatilization [20]. This is accomplished by adding iodide before the addition of acid. The pH of a solution of 10 g of salt in 50 mL of distilled water was acidic (2.0) for DFS and alkaline (7 to 8) for iodized salt. However, iodine is not stable at the acidic pH of DFS. We therefore used H_3PO_4 in the procedure and modified the method by adding 0.50 mL of 1% KI first to 10 g of DFS or iodized salt followed by 50 mL of distilled water and 5 mL of 4N H_3PO_4, and titration was done with 0.005M $Na_2S_2O_3$ after keeping the solution in the dark for 10 minutes [18]. The modified method showed excellent agreement with the conventional titration method for the estimation of iodine (r^2 = 0.9998) both in the KIO_3 standard at different iodine levels and also in iodized salt from the factory or market [18]. Therefore, the NIN and ICCIDD laboratories employed the modified method for the estimation of iodine in DFS, IRS, and IOS throughout the study period, in order to ensure uniformity in methodology.

TABLE 1. Methods used in the estimation of iodine in double-fortified salt (DFS) and iodized salt (IS)[a]

Method	Acid used	Procedure
Conventional titration[b]	Sulfuric acid (H_2SO_4)	10 g DFS or IS + 50 mL distilled water + 1 mL 2N H_2SO_4 + 5 mL 10% KI. Keep in dark for 10 min and titrate with 0.005M $Na_2S_2O_3$.
Modified method[c]	Orthophosphoric acid (H_3PO_4)	10 g DFS or IS + 0.5 mL 1% KI + 50 mL distilled water + 5 mL 4N H_3PO_4 Keep in dark for 10 min and titrate with 0.005M $Na_2S_2O_3$.

a. Values based on reference salt & KIO_3 standard
b. Sullivan et al. [16].
c. Ranganathan et al. [18].

Quality control

In order to ensure the reliability of results, the NIN and ICCIDD laboratories strictly adhered to internal as well as external quality control measures for iodine estimation. For internal quality control, multiple analyses (20 times) of the iodine content of the KIO_3 standard (1 mg iodine/mL) in 10 g of plain noniodized salt were performed. The 95% confidence interval of mean iodine was calculated along with the operating control range (lower limit, mean – 2 SD; upper limit, mean + 2 SD) for preparing the quality control charts. Furthermore, the same procedure was adopted using a reference salt with a known level of iodine to which 1,000 ppm of iron and 100 mg of SHMP were added fresh before iodine estimation. Whenever the iodine content of DFS, IRS, or IOS was estimated on the predetermined dates in the two laboratories, the iodine content of reference salt (with known levels of iodine and iron) and the KIO_3 standard were also measured.

For external quality control, 10 samples each of DFS, IRS, and IOS drawn randomly at NIN were sent to the ICCIDD laboratory. The salt samples were measured in duplicate for iodine content simultaneously by the investigators from both of the centers using the same reagents as the ICCIDD laboratory.

Estimation of iron in DFS

Soon after production (before color coding in the factory) and at the end of the study (after the code was broken) the iron content of DFS was determined according to the method of Wong [21].

Statistical analysis

Analysis of variance was performed with the laboratories and duration of storage as independent variables and iodine values as the dependent variable. The percentage frequency distribution according to iodine content of the three categories of salt was also determined. Regression analysis and *t*-tests were performed wherever necessary.

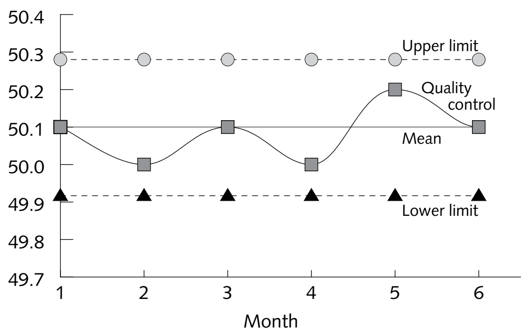

FIG. 1. Internal quality control chart of KIO_3 standard. Mean ± 2 SD = 50.1 ± 0.18 ppm; CV = 0.18%

Breaking of the code

Breaking the code for the fortified salts at the end of the study revealed that the orange pouches contained DFS, yellow pouches IRS, and white pouches IOS.

Results

Initial iodine content

The initial iodine content (mean ± SD) was 40.3 ± 3.8 ppm in DFS, 42.7 ± 3.5 ppm in IRS, and 30.0 ± 2.0 ppm in IOS.

Quality control

The result of the internal quality control of the KIO_3 standard showed that the operating range of iodine content for the KIO_3 standard was 49.92 to 50.28 ppm, with a coefficient of variation of 0.18% (**fig. 1**), and 45.16 to 45.64 ppm with a coefficient of variation of 0.26% for the reference salt (with known levels of iodine and iron). The day-to-day values of the reference salt (with known levels of iodine and iron) and the KIO_3 standard were well within these ranges throughout the study period at the two laboratories, indicating effective internal quality control.

The external quality control revealed good agreement between the duplicate values, within and between the laboratories, irrespective of DFS, IRS, or IOS. The iodine content (mean ± SD) of DFS was 41.5 ± 2.9 ppm

TABLE 2. Iodine content of fortified salts stored at Hyderabad, Delhi, and Bhubaneswar according to number of months of storage

Month	Mean ± SD iodine content (ppm)					
	Double-fortified salt		Iodized refined salt		Iodized ordinary salt	
	NIN	ICCIDD	NIN	ICCIDD	NIN	ICCIDD
Hyderabad and Delhi (*n* = 17 samples per salt per laboratory for each measurement)						
1	40.1 ± 4.3	40.1 ± 4.3	44.3 ± 4.0	44.5 ± 6.0	30.7 ± 2.7	33.6 ± 4.2
2	42.5 ± 4.7	42.0 ± 4.2	43.8 ± 4.9	45.5 ± 4.0	32.4 ± 1.9	33.9 ± 2.1
3	42.0 ± 3.6	39.3 ± 1.8	43.1 ± 3.3	43.0 ± 3.3	31.9 ± 1.2	31.2 ± 1.5
4	42.0 ± 3.6	39.3 ± 1.8*	42.9 ± 2.5	40.3 ± 2.1	32.4 ± 3.0	30.8 ± 1.8*
5	41.4 ± 3.3	40.4 ± 1.6	41.4 ± 3.0	38.2 ± 1.8*	30.2 ± 0.9	28.8 ± 1.6*
6	40.2 ± 2.1	39.6 ± 1.8	41.6 ± 2.3	37.6 ± 2.4*	30.0 ± 1.2	28.2 ± 1.4*
Bhubaneswar (*n* = 6 samples per salt per laboratory for each measurement)						
1	40.2 ± 1.4	43.5 ± 3.0	41.2 ± 1.1	42.3 ± 1.5	33.6 ± 1.0	34.0 ± 0.8
2	40.9 ± 1.3	41.6 ± 1.0	41.4 ± 2.1	41.3 ± 3.4	32.8 ± 1.4	31.9 ± 1.4
3	44.4 ± 3.3	40.2 ± 0.7	42.5 ± 5.6	38.5 ± 2.2*	30.8 ± 1.0	30.3 ± 0.2
4	44.4 ± 3.3	41.3 ± 1.1	43.2 ± 5.0	40.2 ± 1.0*	32.0 ± 0.3	31.4 ± 0.8
5	40.7 ± 0.6	39.8 ± 0.9	44.0 ± 0.6	40.0 ± 1.6*	28.7 ± 2.6	30.7 ± 0.9*
6	40.7 ± 0.9	40.4 ± 0.8	40.5 ± 1.1	40.0 ± 0.8	29.8 ± 0.8	29.5 ± 0.8

NIN, National Institute of Nutrition; ICCIDD, International Council for Control of Iodine Deficiency Disorders
*Difference between laboratories significant at $p < .05$ (*t*-test).

for NIN and 42.5 ± 3.0 ppm for ICCIDD; the iodine content of IRS was 46.1 ± 2.8 ppm for NIN and 46.0 ± 3.1 ppm for ICCIDD; and the iodine content of IOS was 35.1 ± 2.9 ppm for NIN and 35.9 ± 2.8 ppm for ICCIDD. The intraclass correlation was close to unity (ρ = 0.97).

Iodine content of DFS, IRS, and IOS stored at Hyderabad and New Delhi

Regular monthly analysis of the salt samples carried out at the two laboratories showed that the mean iodine content of DFS as well as IRS was about 40 ppm, while that of IOS was 30 ppm (**table 2**). The iodine levels of all three fortified salts during the study period were essentially the same as their initial levels. Analysis of variance showed that there was no difference in iodine content between the two laboratories and at different time points within the same laboratory. However, there were minor differences in iodine content between the laboratories at some points ($p < .05$), mainly in IOS and IRS (**table 2**). These differences could be attributed to batch-to-batch variations at the time of production. Since the frequency distributions of the iodine values were nearly identical, these differences are of no practical relevance. Furthermore, the percentage of salt samples having ≥30 ppm iodine was 100% in DFS, IRS, and IOS throughout the study period (mandatory level: 15 to 30 ppm).

Iodine content of DFS, IRS, and IOS stored at Bhubaneswar

The iodine contents of the three fortified salts stored at Bhubaneswar and tested at the NIN and ICCIDD laboratories are given in **table 2**. The mean iodine content of DFS and IRS was about 40 ppm, whereas that of IOS was 30 ppm throughout the 6-month period. No significant differences, in general, were observed in the iodine content.

TABLE 3. Iodine content of fortified salts after 15 months of storage

Salt	Mean ± SD iodine content (ppm) (n = 17 samples per salt per laboratory)	
	NIN	ICCIDD
Double-fortified	31.9 ± 1.8	31.7 ± 2.2
Iodized refined	41.2 ± 0.9	42.9 ± 3.1
Iodized ordinary	29.8 ± 0.7	30.0 ± 1.5

NIN, National Institute of Nutrition; ICCIDD, International Council for Control of Iodine Deficiency Disorders

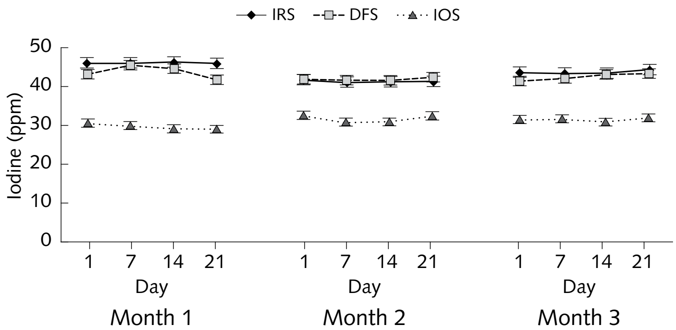

FIG. 2. Weekly variation of iodine content of fortified salts during the first 3 months. DFS, double-fortified salt; IRS, iodized refined salt; IOS, iodized ordinary salt

Iodine content of DFS, IRS, and IOS after 15 months

In view of the excellent iodine stability observed in the first 6 months, all of the salt samples were stored for a long time and the iodine content was measured in these samples after 15 months by the same protocol and sampling procedures followed during the first 6 months. The results revealed consistency in iodine stability even after 15 months (**table 3**); DFS and IOS had about 30 ppm iodine and IRS about 40 ppm iodine. The percentage of salt samples having ≥ 30 ppm iodine was 100% in DFS, IRS, and IOS even after 15 months (mandatory level: 15 to 30 ppm).

Variation of iodine content in the fortified salts stored under simulated household conditions

Weekly analysis of subsamples of DFS, IRS, and IOS stored under simulated household conditions during the first 3 months of the study period revealed no significant changes in iodine content; the mean iodine contents of DFS and IRS were about 40 ppm, and that of IOS was 30 ppm (**fig. 2**).

Overall iodine stability

The study revealed that the stability of iodine was consistently satisfactory in DFS, IRS, and IOS (**fig. 3**). All three types of fortified salt had ≥ 30 ppm iodine, even after 15 months.

Iron content of DFS

The mean iron content of DFS was 1,030 ± 62 ppm soon after production (n = 40) and 1,034 ± 58 ppm at the end of 6 months (n = 40), indicating satisfactory iron stability.

Discussion

This study has reconfirmed the large-scale production

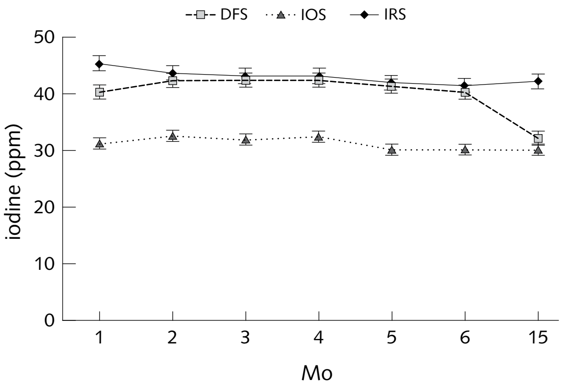

FIG. 3. Mean iodine content of fortified salts according to duration of storage. Mandatory iodine level, 15 to 30 ppm. DFS, double-fortified salt; IRS, iodized refined salt; IOS, iodized ordinary salt

of DFS and iodized salt by the dry-mixing method and transportation over long distances. The initial iodine content of IOS was less (30 ppm) than that of IRS and DFS (40 ppm), although the same amount of KIO_3 was used in all three salts. This difference in iodine levels could be attributed to the composition of common salts, especially the higher levels of moisture (3.8%) and magnesium (0.10%) in ordinary salt, as compared with the low levels of moisture (0.14%) and magnesium (0.01%) in refined salt. Earlier studies of iodized salt revealed a certain amount of iodine loss during the initial months, although KIO_3 was used as the source of iodine. This was attributed to the use of water in the spray-mixing process employed for the production of iodized salt [4, 22-25]. In contrast, no iodine loss was observed when the dry-mixing process was adopted, and excellent iodine stability was ensured during prolonged storage [3, 17, 22, 26]. The present study confirms these observations.

The monthly testing of DFS and iodized salt revealed that DFS and iodized salt stored at Bhubaneswar, Hyderabad, and New Delhi had excellent iodine stability and the iodine content was always ≥30 ppm in 100% of the samples tested throughout the study period; the salt pouches stored and handled under simulated household conditions did not show any iodine loss in DFS and iodized salt; the modified method using H_3PO_4 is suitable not only for DFS but also for iodized salt; and the coastal environmental conditions at Bhubaneswar did not affect the stability of iodine in DFS or iodized salt, indicating that the poor iodine stability observed earlier in DFS at Bhubaneswar [17] could be due to the inherent problems in the method of iodine estimation followed at that time.

It can therefore be concluded that DFS prepared according to the NIN formula had excellent iodine stability even after 15 months. This study confirmed the usefulness of the modified method for estimation of iodine in iodized salt, whether the salt is derived from ordinary common salt or from refined common salt.

Acknowledgments

The authors wish to thank Prof. N. K. Ganguly, Director General, Indian Council of Medical Research (ICMR), Prof. M. K. Bhan, Secretary, Department of Biotechnology, Government of India, and all the members of the Scientific Advisory Committee of NIN for their keen interest and constant encouragement throughout the study. The authors are thankful to Dr. S. K. Kar, Director, RMRC, Bhubaneswar, and the staff of the NNMB, Orissa Unit, for their excellent cooperation during the study. This work would not have been possible but for the generous financial support from ICMR, for which the authors are grateful to the Council.

References

1. World Bank. Enriching lives: Overcoming vitamin and mineral malnutrition in developing countries. Washington, DC: World Bank, Report no. 13800, November 1994.
2. Adamson P. Vitamin and mineral deficiency: A global progress report. Oxfordshire, UK: UNICEF/Micronutrient Initiative, 2004.
3. Ranganathan S, Sundaresan S, Raghavendra I, Kalyani S. Dry mixing technique for the large scale production of iodine fortified salt in India. Asia Pacific J Clin Nutr 1997;6:92–4.
4. Ranganathan S, Rao BSN, Reddy V, Prakash R, Sundaresan S, Ansari MA. A new process for stabilizing iodine in iodized salt. In: Geertman RM, ed. Salt 2000. 8th World Salt Symposium. Vol 2. The Hague: Elsevier, 2000:1051–6.
5. Ranganathan S. Fortification of salt with iron: Use of polyphosphate stabilizers. Food Chem 1992; 45: 263–7.
6. Ranganathan S, Dillikumar PK, Ramamoorthy P, Reddy V. Large-scale production of iron fortified salt. J Food Sci Technol 1993;30:166–8.
7. Narasinga Rao BS. Fortification of salt with iron and iodine to control anaemia and goiter: Development of a new formula with good stability and bioavailability of iron and iodine. Food Nutr Bull 1994;15:32–9.
8. JECFA (Joint FAO/WHO Expert Committee on Food Additives). Compendium of food additive specifications. Vol. 2. Combined specifications from the first to the thirty-seventh meeting (1956–1990). FAO Food and Nutrition Paper 52/2. Rome: Food and Agriculture Organization, 1992.
9. Ranganathan S, Reddy V, Ramamoorthy P. Large-scale production of salt fortified with iodine and iron. Food Nutr Bull 1996;17:73–8.

10. Ranganathan S, Singotamu L. Scanning electron microscope studies on iodine. Scanning 1998;20:271–3.
11. Nair KM, Sesikaran B, Ranganathan S, Sivakumar B. Bio-effect and safety of long term feeding of common salt fortified with iron and iodine (double-fortified salt) in rat. Nutr Res 1998;18:121–9.
12. Nair KM, Brahmam GNV, Ranganathan S, Vijayaraghavan K, Sivakumar B, Krishnaswamy K. Impact evaluation of iron and iodine fortified salt. Indian J Med Res 1998;108:203–11.
13. Brahmam GNV, Nair KM, Laxmaiah A, Gal Reddy C, Ranganathan S, Vishnuvardhana Rao M, Naidu AN, Vijayaraghavan K, Sivakumar B, Krishanaswamy K, Sastry JG, Ram MM, Rao NP, Reddy V. Community trials with iron and iodine fortified salt (double-fortified salt). In: Geertman RM, ed. Salt 2000. 8th World Salt Symposium. Vol. 2. The Hague: Elsevier, 2000: 955–60.
14. Sivakumar B, Brahmam GNV, Madhavan Nair K, Ranganathan S, Vishnuvardhan Rao M, Vijayaraghvan K, Krishnaswamy K. Prospects of fortification of salt with iron and iodine. Br J Nutr 2001;85(suppl 2):S167–73.
15. Sivakumar B, Nair KM. Double fortified salt at crossroads. Indian J Pediatr 2002;69:617–23
16. Titration methods for salt iodine analysis. In: Sullivan KM, Houston R, Gorstein J, Cervinskas J, eds. Monitoring universal salt iodization programmes. Atlanta, GA, USA: Programme against Micronutrient Malnutrition/Micronutrient Initiative/International Council for Control of Iodine Deficiency Disorders, 1995:86–9.
17. Operational evaluation of the stability of iodine in double-fortified salt: A multicentric study. Interim report, Hyderabad, India: National Institute of Nutrition (Indian Council of Medical Research), January 2003.
18. Ranganathan S, Karmarkar MG. Estimation of iodine in salt fortified with iodine and iron. Indian J Med Res 2006;123:531–40.
19. Kolthoff IM, Belcher R. Volumetric analysis. Vol III. Titration methods: Oxidation-reduction reactions. New York: Interscience Publishers, Inc., 1957:247.
20. Kolthoff IM, Belcher R. Volumetric analysis. Vol III. Titration methods: Oxidation-reduction reactions. New York: Interscience Publishers, Inc., 1957:235.
21. Wong SY. Hawk's physiological chemistry, 14th ed. New York: McGraw-Hill, 1965:1094.
22. Ranganathan S, Narasinga Rao BS. Stability of iodine in iodized salt. Indian Food Ind 1986;5:122–4.
23. Chauan, SA, Bhatt AM, Bhatt, MP, Majeethia KM. Stability of iodized salt with respect to iodine content. Res Ind 1982;37:38–41.
24. Pandav CS, Anand K, Karmarkar MG. Costing of a salt iodine monitoring laboratory in India. Natl Med J India 1994;7:13.
25. Kelly FC. Studies on the stability of iodine compounds in iodized salt. Bull World Health Organ 1953;9:217–30.
26. Holman JC, McCartney W. Iodized salt. In: WHO Monograph Series No. 44. Endemic goitre. Geneva: World Health Organization, 1960:424–6.

Book reviews

Nevin S. Scrimshaw

Advances in food mycology. Edited by A. D. Hocking, J. I. Pitt, R.A. Samson, and U. Thane. Springer, New York, 2006. (ISBN 0-387-28385-4) 378 pages, hardcover. US$139.00.

This book presents the proceedings of the Fifth International Workshop on Food Mycology held in Denmark in 2003. Particular emphasis was placed on the fungi that produce mycotoxins, their ecology, and potential control measures. Chapters are also devoted to yeasts and the inactivation of fungal spores by the use of heat and high pressure. The 22 chapters are grouped under the headings "Understanding the fungi producing important mycotoxins," "Media and method development in food mycology," "Physiology and ecology of mycotoxigenic fungi," and "Control of fungi and mycotoxins in food." A final chapter is devoted to "recommended methods for food mycology." There is also an appendix listing recommended media.

Although this volume will be of primary interest to food microbiologists and food technologists, others concerned with food safety may find it an updated and useful reference.

Impact of animal source foods on growth, morbidity and iron bioavailability in Kenyan school children. Monika Grillenberger. Thesis, Wageningen University, Netherlands, 2006. (ISBN 90-8504-387-5) 176 pages. Available at: http://library.wur.nl/wda/dissertations/dis3935.pdf.

From time to time, the *Bulletin* reviews a published Ph.D. thesis on a topic of special interest to its readers. The case for the significance of animal protein intake for the growth and development of children forced by poverty to consume a diet with limited animal protein is such a topic. The research presented is part of a randomized, controlled evaluation of the effect of increasing the consumption of foods of animal origin on growth, morbidity, and cognitive function of children of subsistence farmers in Kenya. Their habitual diets contained very little meat or dairy products and consisted primarily of cereal. Children receiving the animal food supplementation were compared with those receiving an isoenergetic food supplement or none at all.

The meat diet had a beneficial effect on gain in lean body mass. The intake of animal foods, with their higher content of iron, zinc, and other available micronutrients than the usual cereal-based diet, clearly benefited the growth of the children. A trend toward lower morbidity and severity of infectious disease was not statistically significant. The role of iron absorption inhibitors was explored. Addition of meat and a source of ascorbic acid improved iron status. Although these results are not unexpected, they confirm the potential developmental benefit of adding animal protein to predominantly cereal diets whenever this can be achieved.

Nutrient–drug interactions. Edited by Kelly Anne Meckling. Taylor & Francis/CRC Press, Boca Raton, Fla., USA, 2007. (ISBN 1-57444-915-X) 352 pages, hardcover. US$99.95.

Despite its title, this book does not attempt to review systematically the interactions of dietary components with the great range of commonly prescribed pharmacologic agents. Instead it focuses on "food, herbals and their constituents." Nevertheless, the two initial chapters on diabetes, obesity, and the metabolic syndrome and on hypolipidemic therapy do review a full range of glucose-lowering, antiobesity, and lipid-lowering agents as well as their interactions with diet components.

The next chapters focus entirely on dietary and herbal therapy of cancer, of neurodegenerative disorders (with the use of antioxidants), and of neurodegenerative disorders and unipolar and major depression. The chapter on supplements and anesthesiology deals only with vitamins, minerals, and herbals.

The chapter on nutrient- and drug-responsive genes as nutrigenic tools for the prevention of chronic vascular disease and cancer emphasizes relevant genetic polymorphisms. It makes a plea for thinking of nutrient, drug, and gene interactions when designing research studies. The final chapter on nutrigenomics and pharmacogenomics of human cancer has over 300 references, a 5-page tabulation of interactions between common metabolic gene variants and dietary factors in cancer risk modulation, and a 12-page tabular summary of polymorphic genes encoding factors affecting the outcome of cancer chemotherapy and radiotherapy.

Some will find the first and last two chapters of considerable interest. Individuals looking for information on interactions between diet and herbals in therapy for some common disorders will find this book useful. Those needing a systematic review of interactions between dietary constituents, genetic polymorphisms, and synthetic pharmacologic agents in current use will need to look elsewhere.

Nutrient–gene interactions in cancer. Edited by Sang-Woon Choi and Simonetta Friso. Taylor & Francis/CRC Press, Boca Raton, Fla., USA, 2006. (ISBN 0-8493-3229-X) 281 pages, hardcover. US$149.95.

The complete mapping of the human genome has accelerated research on interactions between nutrients and gene expression as a means by which nutrients modulate carcinogenesis. The first three chapters cover basic elements of the biology and pathology of gene–nutrient interactions. They focus on mechanisms and on signal transduction pathways in lung cancer prevention investigations.

The nine chapters that make up the body of the book describe in detail the specific gene–nutrient interactions that are the most striking examples of the interactions between genetics, epigenetics, and nutrition in many different types of cancer. These include interactions of folate metabolites with cancer genes and with S-adenosylmethionine and methionine adenosyltransferase genes; the effects of alcohol dehydrogenase on alcohol-associated carcinogenesis; the effects of carotenoid supplementation on lung cancer prevention; vitamin D receptors in gene regulatory actions; the role of alcohol dehydrogenase polymorphism in alcohol-associated carcinogenesis; polymorphisms of N-acetyltransferase genes as risk modifiers of colorectal cancer; ferritin, and serine hydroxyl methyltransferase; and the impact of *Brassica*–gene interactions on cancer risk.

The authors and editors have contributed much to this field, and the chapters are well referenced. This is a field in its early stages, and the book is a sampling of some of the most rewarding current research. It serves to describe initial progress in an interdisciplinary research area of growing importance.

Obesity: Dietary and developmental influences. Edited by Gail Woodward-Lopez, Lorrene D. Ritchie, Patricia B. Crawford, and Dana E. Gerstein. Taylor & Francis/CRC Press, Boca Raton, Fla., USA, 2006. (ISBN 0-8493-9245-4) 360 pages, hardcover. US$89.95.

Overweight and obesity and the chronic diseases associated with them are major health problems facing almost every country. As would be expected, there are many new books on various aspects of this topic, all seeking to be useful. This publication is funded by the Centers for Disease Control, the US Department of Agriculture, the University of California at Berkeley, and the American Dietetics Association.

Its preface states "It is particularly critical that nutrition experts, educators and decision makers in government, academia, clinical practice and public health provide clinical, actionable, and unequivocal dietary recommendations for the prevention of overweight." This book attempts to contribute to this by "reviewing all of the available literature" to arrive at nine dietary recommendations that are most strongly supported by currently available evidence. However, they are not identified until the last chapter and in a disappointingly brief and prosaic manner.

What the book does do is to review data on each of the critical periods throughout the life cycle, the multiple individual dietary influences on energy balance, parental influences on eating habits, breastfeeding, and food insecurity. This information is presented in 38 tables that summarize observational studies, 38 graphs depicting trends in dietary intake, and 9 tables that summarize prevention trials. On the basis of this mass of information, the nine recommendations are identified and will be acceptable to almost everyone. They merit more effective promotion.

WHO child growth standards. World Health Organization, Geneva, 2006. (ISBN 92-4-154693-X) 312 pages, softcover. US$45.00.

A comprehensive review of the uses and interpretation of anthropometric measures, including the National Center for Health Statistics/World Health Organization (NCHS/WHO) growth reference in current use, undertaken by WHO in the early 1990s, concluded "that new growth standards were needed to replace the existing international references." To develop new standards, a multicountry assessment was carried out to collect primary and related information from 8,440 breastfed infants and young children from diverse ethnic backgrounds and cultural settings in Brazil, Ghana, India, Norway, Oman, and the United States. The resulting growth standards depict "normal early childhood growth under optimal environmental conditions." They are considered the best available for the assessment of children everywhere, regardless of ethnicity, socioeconomic status, and type of feeding.

The methods and development of standards for length/height-for-age, weight-for-age, weight-for-length, weight-for-height, and body mass index-for-age are described in detail. The actual data analyses and summaries are presented in 136 figures and 97 tables. Although there is no reason to believe that additional ethnic, cultural, or geographic data were needed or could have affected the results, including an East Asian population would have enhanced credibility. This has been a monumental effort. It is the best set of anthropometric reference standards for infants and young children available and must now be the first choice of all nutrition and health workers concerned with young children.

News and notes

Food and Agriculture Organization (FAO)

Strengthening national food control systems: Guidelines to assess capacity building needs

This new tool, developed by FAO in collaboration with the World Health Organization (WHO), provides practical assistance to help governments identify their capacity-building needs in the core components of a national food control system. The *Guidelines* include five modules for food control management, legislation, inspection, laboratories, and information, education, and communication (IEC). Each module sets out a seven-step process to critically examine existing capacity, consider the improvements desired for the future, pinpoint capacity-building needs, and identify options to address them. Internationally accepted benchmarks are included, as well as useful resources to facilitate the process. Available at: ftp://ftp.fao.org/docrep/fao/009/a0601e/a0601e00.pdf.

Strengthening national food control systems: A quick guide to assess capacity building needs

FAO's *Quick guide* is targeted at users who want to carry out a general rapid assessment of the food control system as a whole. Complementing the more in-depth *Guidelines to assess capacity building needs*, the *Quick guide* provides support to profile the existing food control system, define the goals and objectives of the desired improved food control system, and generate a capacity-building action plan. Tips and suggestions are given at each step of the process, and a variety of questionnaires, templates, and other resources are included as annexes. Available in 2007 in English, French, and Spanish.

Food safety risk analysis: A guide for national authorities

FAO and WHO have developed this *Guide* to improve food-safety regulators' understanding and use of risk analysis. The *Guide* provides essential background information, guidance, and practical examples of ways to apply food-safety risk analysis. It presents internationally agreed-upon principles, a generic framework for application of the different components of risk analysis, and wide-ranging examples. Current information and knowledge, including materials developed by FAO and WHO, are incorporated or referenced throughout the document. Available in 2007 in English, French, and Spanish.

For more information regarding these publications, please contact us at:

Nutrition and Consumer Protection Division (AGN)
Food Quality and Standards Service (AGNS)
Agriculture, Biosecurity, Nutrition and Consumer Protection Department
Food and Agriculture Organization of the United Nations
Viale delle Terme di Caracalla
00153 Rome, Italy
http://www.fao.org/ag/agn/index_en.stm

7th International Food Data Conference (IFDC)

Food Composition and Biodiversity
São Paulo, Brazil, 22-24 October 2007
Pre-conference workshop on 21 October 2007

Preliminary Announcement and Call for Abstracts
http://www.fcf.usp.br/7ifdc/

ILSI (International Life Sciences Institute)

April 16–18, 2007. Micronutrient Forum. Consequences and Control of Micronutrient Deficiencies: Science, Policy, and Programs—Defining the Issues. Istanbul, Turkey.

For information contact:
Micronutrient Forum Secretariat
Tel: 202-659-9024
Fax: 202-659-3617
E-mail: mnforum@ilsi.org
Website: www.micronutrientforum.org

UNU Food and Nutrition Program Notice

It has been a feature of the *Food and Nutrition Bulletin* to publish the complete listing of the United Nations University Food and Nutrition Institutional Network with full contact information in its March issue. It does not appear in this issue because the entire UNU Food and Nutrition Program and its network of Associated and Cooperating Institutions is undergoing reorganization and renewal. The new network structure will be published in the *Bulletin* when the updated information is available.

Food and Nutrition Bulletin
Support for Subscriptions to Developing Countries

International agencies

The United Nations University (UNU)
The United Nations Children's Fund (UNICEF)

Bilateral agencies

United States Agency for International Development (USAID)

Nongovernmental organizations

International Life Sciences Institute (ILSI)

Corporations

Akzo Nobel Chemicals
DSM Nutritional Products
Kraft Foods
Procter & Gamble Co.
Unilever

Useful web sites and free materials

Access to Global Online Research in Agriculture (AGORA)	www.aginternetwork.org/en/about.php
Bioversity International	www.bioversityinternational.org
Food and Agriculture Organization (FAO)	www.fao.org
International Atomic Energy Agency (IAEA)	www.iaea.org
International Life Sciences Institute (ILSI)	www.ilsi.org
International Nutritional Anemia Consultative Group (INACG)	http://inacg.ilsi.org
International Nutrition Foundation (INF)	www.inffoundation.org
International Vitamin A Consultative Group (IVACG)	http://ivacg.ilsi.org
International Union of Nutritional Sciences (IUNS)	www.iuns.org
Iron Deficiency Project Advisory Service (IDPAS)	www.micronutrient.org/idpas
The Micronutrient Initiative	www.micronutrient.org
Pan American Health Organization (PAHO)	www.paho.org
Save the Children	www.savethechildren.org
Unilever	www.unilever.com
United Nations Children's Fund (UNICEF)	www.unicef.org
United Nations University (UNU)	www.unu.org
UN Standing Committee on Nutrition (SCN)	www.unsystem.org/scn
World Bank	www.worldbank.org
World Food Program	www.wfp.org
World Health Organization (WHO)	www.who.int/en

Information for Authors

The editors of the *Food and Nutrition Bulletin* welcome contributions of relevance to its concerns (see the statement of editorial policy). Submission of an article does not guarantee publication; acceptance depends on the judgment of the editors and reviewers as to its relevance and quality. All potentially acceptable manuscripts are peer-reviewed. Contributors should examine recent issues of the *Bulletin* for content and style.

Ethical approval of studies and informed consent. For investigations of human subjects, authors should state in the Methods section the manner in which informed consent was obtained from the study participants (i.e., oral or written), and describe how the study investigators protected the rights of participants as described in the Declaration of Helsinki.

Language. Contributions must be submitted in English.

Format. **Manuscripts should be prepared on a computer, and submitted electronically via e-mail directly to the Managing Editor.**

Abstract. An abstract of not more than 250 words should be included at the beginning of the manuscript, in the following format:

» *Background.* The context of the problem you are investigating, with relevant historical information.
» *Objective.* A one- or two-sentence description of the purpose of the study and what you expected to find.
» *Methods.* Outline of study design, subject selection, analytical methods, data analysis.
» *Results.* What you found based on your data. Give specific data and their statistical significance here if possible.
» *Conclusions.* One- or two-sentence description of what you conclude from your results.

Emphasize new and important aspects of the study or observations. Do not include any information that is not given in the body of the article. Do not cite references or use abbreviations or acronyms in the abstract.

Key words. Authors should provide a minimum of four key words for the article.

Tables and figures. Tables and figures should be placed on separate pages in the manuscript file. Footnotes should be keyed to the relevant data points by letters or symbols. Figures should be submitted electronically, as part of the manuscript file or as a separate electronic file. The original data files for figures that use bar graphs, scatterplots, or other graphic representations of data should be sent along with the manuscript. Please double-check your data for accuracy and consistency with the text.

Photographs. Photographs may be mailed or submitted electronically. Mailed photographs will not be returned unless specifically requested.

Units of measure. All measurements should be expressed in metric units. If other units are used, their metric equivalent should be indicated.

Abbreviations. Please spell out all abbreviations used on the first reference.

References. References should be listed at the end of the article. Unpublished papers should not be listed as references, nor should papers submitted for publication but not yet accepted. Please double-check that reference numbers correspond to the correct numbers in the text.

Number references consecutively in the order in which they are first mentioned in the text. Identify references in the text and tables and figure legends by arabic numerals enclosed in square brackets. References cited only in tables or figure legends should be numbered in accordance with the first mention of the relevant table or figure in the text. **Be sure references are complete and current.**

Reference citations should follow the format below.

Journal reference

—standard journal article (list all authors):

1. Alvarez MI, Mikasic D, Ottenberger A, Salazar ME. Caracteristicas de familias urbanas con lactante desnutrido: un análisis critico. Arch Latinoam Nutr 1979;29:220–30.

—corporate author:

2. Committee on Enzymes of the Scandinavian Society for Clinical Chemistry and Clinical Physiology. Recommended method for the determination of gammaglutamyltransferase in blood. Scand J Clin Lab Invest 1976;36:119–25.

Book or other monograph reference

—personal author(s):

3. Brozek J. Malnutrition and human behavior: experimental, clinical and community studies. New York: Van Nostrand Reinhold, 1985.

—corporate author:

4. American Medical Association, Department of Drugs. AMA drug evaluations, 3rd ed. Littleton, Mass, USA: Publishing Sciences Group, 1977.

—editor, compiler, chairman as author:

5. Medioni J, Boesinger E, eds. Mécanismes éthologiques de l'évolution. Paris: Masson, 1977.

—chapter in book:

6. Barnett HG. Compatibility and compartmentalization in cultural change. In: Desai AR, ed. Essays on modernization of underdeveloped societies. Bombay: Thacker, 1971: 20–35.

World Wide Web reference

7. WHO HIV infections page. WHO web site. Available at: http://www.who.int/topics/hiv_infections/en/. Accessed 12 October 2004.
8. Nielsen J, Palle V-B, Martins C, Cabral F, Aaby P. Malnourished children and supplementary feeding during the war emergency in Guinea-Bissau in 1998–1999 [serial online]. Am J Clin Nutr; 2004; 80:1036-42. Available at: http://www.ajcn.org/cgi/content/full/80/4/1036. Accessed 12 October 2004.

Identification. Please give the full name of each author, the name of departments and institutions to which the work should be attributed, the name, address, fax number, and e-mail address of the corresponding author, and sources of support for the work. If the material in the article has been previously presented or is planned to be published elsewhere—in the same or modified form—a note should be included giving the details.

Page charges. The *Bulletin* has a page charge of US$60 for unsolicited original research articles. One printed page in the *Bulletin* is equivalent to approximately 3 double-spaced manuscript pages. Authors contributing to special issues and supplements are not responsible for page charges.

Contributions should be addressed to:

Susan Karcz, Managing Editor
Food and Nutrition Bulletin
150 Harrison Ave.
Boston, MA 02111 USA
FNB@inffoundation.org

Subscribe to the *Food and Nutrition Bulletin*

Annual Subscriptions

The annual subscription cost of the *Bulletin* is US$56.00, which includes both the quarterly issues and supplements. To subscribe, write or email:

International Nutrition Foundation
150 Harrison Ave.
Boston, MA, 02111, USA
Tel: 617-636-3771
Fax: 617-636-3727
E-mail: infoperations@inffoundation.org

Subsidized Subscriptions Available

The International Nutrition Foundation (INF) is raising funds to increase the subsidized distribution of the *Food and Nutrition Bulletin* to nutrition scientists and institutions in developing countries. This effort has been supported by the United Nations University (UNU), the United Nations Children's Fund (UNICEF), the International Atomic Energy Agency (IAEA), the United States Agency for International Development (USAID), The Micronutrient Initiative (MI), and the International Life Sciences Institute (ILSI). Contributions have also been received from Akzo Nobel Chemicals, DSM Nutritional Products, Kraft Foods, Procter & Gamble Co., and Unilever.

If you (or your organization) are working in the field of nutrition, and are from a developing country, you may be eligible for a donated or subsidized subscription. The extent to which requests for free subscriptions can be met depends on available funds. The *Bulletin*'s goal of promoting a wide geographic and institutional distribution will be taken into consideration. Individuals and institutions working in developing countries and countries in transition may apply biannually for a subsidized subscription.

Preference for subsidized subscriptions will be given to libraries. If you are affiliated with a university in a developing country, we encourage you to make this information available to the library of your institution.

Normally, individuals holding international posts working with international and bilateral agencies, international nongovernmental organizations (NGOs), and private corporations are not eligible for free subscriptions.

Subscription exchanges with journals of similar purposes and interests are welcome.

To apply for a subsidized subscription, write or email:

International Nutrition Foundation
150 Harrison Ave.
Boston, MA, 02111, USA
Tel: 617-636-3771
Fax: 617-636-3727
E-mail: infoperations@inffoundation.org

Food and Nutrition Bulletin Subscription Form

☐ Please enter my subscription to the *Food and Nutrition Bulletin* (4 issues).

Regular rates: ☐ 1 year, US$56 ☐ 2 years, US$106 ☐ 3 years, US$150
All rates include delivery by surface mail.

Total payment enclosed: ____________

Individuals are requested to enclose payment with their orders. Prices subject to change without notice. Payment must be made in US dollars only. Checks should be made payable to: International Nutrition Foundation, Inc. Subscriptions will begin with the issue following placement of the order.

Name: ____________________

Address: ____________________

____

____

Send to: Subscriptions
International Nutrition Foundation, Inc.
150 Harrison Ave.
Boston, MA 02111 USA