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## Caterpillar Cereal as a Potential Complementary Feeding Product for Infants and Young Children: Nutritional Content and Acceptability

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## Abstract

Micronutrient deficiency is an important cause of growth stunting. To avoid micronutrient deficiency, the World Health Organization recommends complementary feeding with animalsource foods. However, animal-source foods are not readily available in many parts of the Democratic Republic of Congo (DRC). In such areas, caterpillars are a staple in adult diets and may be suitable for complementary feeding for infants and young children. We developed a cereal made from dried caterpillars and other locally available ingredients (ground corn, palm oil, sugar and salt), measured its macro-and micronutrient content and evaluated for microbiologic contamination. Maternal and infant acceptability was evaluated among 20 mothers and their 8-10 month-old infants. Mothers were instructed in the preparation of the cereal and asked to evaluate the cereal in five domains using a Likert scale. Mothers fed their infants a 30-gram portion daily for one week. Infant acceptability was based on cereal consumption and the occurrence of adverse events. The caterpillar cereal contained 132 kcal, 6.9-g protein, 3.8-mg iron and 3.8-mg zinc per 30 grams and was free from microbiologic contamination. Mothers' median ratings for cereal characteristics were (5=like very much): overall impression=4, taste=5, smell=4, texture=4, color=5, and consistency=4. All infants consumed more than 75% of the daily portions, with five infants consuming 100%. No serious adverse events were reported. We conclude that a cereal made from locally available caterpillars has appropriate macro-and micronutrient content for complementary feeding, and is acceptable to mothers and infants in the DRC.

CONFLICT OF INTEREST STATEMENT: All authors report no conflicts of interest.

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## INTRODUCTION

In children under five years of age, malnutrition is responsible for 2.1 million deaths annually and 91 million disability adjusted life years (Black et al. 2008). Growth stunting, defined as height-for-age Z score two standard deviations or more below appropriate World Health Organization (WHO) standards, may be a consequence of long-term malnutrition and has been associated with multiple negative health outcomes, including increased mortality, poor cognitive and school performance, delayed motor development, impaired physical performance, reduced income in adulthood and lower birth weight in offspring (Phuka et al. 2009, Barker et al. 2005). If stunting from malnutrition is not reversed by the age of two years, the adverse effects are likely to be permanent (Dewey & Adu-Afarwuah 2008). The prevalence of stunting varies around the globe, but low income countries (LIC) are disproportionately affected. In the Democratic Republic of Congo (DRC), the stunting prevalence in children under five years of age is estimated to be greater than 40% (Black et al. 2008).

Children from three months to 36 months are particularly vulnerable to insults affecting linear growth, especially after the period of exclusive breast-feeding when complementary foods are introduced into the diet (Frongillo et al. 1999). Malnutrition from inadequate complementary feeding is a serious problem in many LIC where complementary foods consist of starch-based cereals or gruel that may provide sufficient energy but inadequate protein and micronutrients (Dewey 2003). Micronutrient deficiencies, most notably zinc deficiency, are associated with stunting of growth and other serious health consequences including anemia and a greater susceptibility to infection (Bhutta et al. 2008).

Authoritative guidelines on ideal complementary feeding recommend the daily consumption of animal-source foods in order to achieve adequate intakes of deficient nutrients, specifically iron and zinc, which are not achievable with plant-based diets alone (Dewey 2003, WHO 1998, Hambidge & Krebs 2007). Unfortunately, animal-source foods are not affordable in many areas of the DRC. However, insects have played a critical role in the diet of many people in Central Africa, with 70% of the adult population of Kinshasa, the capital of the DRC, consuming insects (Balinga et al. 2004, Latham 2005). Dried caterpillars have a protein and micronutrient content similar to beef (Latham 2005, Kodondi et al. 1987). Therefore, we speculated that dried caterpillars may be an alternative to meat as a source of protein and micronutrients. The overall goal of this project is to reduce growth stunting using a micronutrient-rich cereal made from caterpillars and other locally available ingredients that is an acceptable complementary food for infants in the DRC.

Problems with behavioral and social compliance to feeding interventions limit the efficacy of food supplementation (Mamiro et al. 2004, Bhandari et al. 2001, Lartey et al. 1999). Feeding programs have had limited success if little attention was paid to socio-cultural influences on infant feeding, including maternal attitude (Bentley et al. 1991). Both maternal and infant acceptability of food sources are important indicators of compliance with a feeding intervention, and lack of compliance has resulted in erroneous conclusions about the potential benefits of previous dietary interventions.<sup>17</sup> Therefore, one of the aims of this project was to test the acceptability of the caterpillar cereal. In this paper, we report the

results of the biochemical and microbiologic analyses of a cereal made from caterpillars and the investigation of maternal and infant acceptability of the cereal.

## MATERIALS AND METHODS

#### **Cereal Development**

We developed a cereal made from dried caterpillars, ground corn, sugar, salt and palm oil. We chose these ingredients based on the nutritional requirements of young children, the dietary habits of the population and the availability of these products in the local markets of the DRC. We chose corn because it constitutes a basic food source for many populations in the DRC, and its use in infant food is common throughout the country. We chose palm oil because it is a rich source of lipids, contributes  $\beta$ -carotene, a precursor of vitamin A, and produces a cereal of a more desirable color. We added small amounts of sugar and salt for palatability. We produced cereal in accordance with the international standards on the formulation of foods intended for infants and children up to two years of age outlined in the Codex Alimentarius (WHO 2006, WHO 1979).

To make the cereal, we processed each component separately and then mixed them together to create the final product.

- Caterpillar flour: The caterpillars were initially washed and soaked in water for 30 minutes then dried in the sun. Dried caterpillars were crushed in a grinding mill and filtered to a flour of fine granularity and uniform consistency.
- 2) Corn flour: Dried kernels were initially filtered in a sleeve with broad mesh to remove foreign material from the corn. Twice, the kernels were soaked in water at room temperature and rinsed. The kernels were dried in the sun, crushed in a grinding mill and filtered to a flour of fine granularity and uniform consistency.
- 3) Palm oil: Oil required no processing before mixing.
- 4) Sugar and salt: Each ingredient was crushed separately to obtain a fine powder.

We mixed caterpillar flour, corn flour, salt and sugar in a basin. The proportion of caterpillar flour to corn flour was 1:1. We chose this proportion such that the cereal would provide the recommended daily intake of zinc. We added palm oil to the mixture and dried the final mixture in an oven at 60° C for 24 hours. Single feeding portions (30 grams) of the cereal were sealed in plastic sachets. We assured hygienic production of cereal by cleansing all equipment prior to the cereal production, assuring hygiene of all personnel including hand washing, and requiring the use of masks and hair coverings during cereal production.

### **Chemical and Microbiologic Testing**

We performed chemical analyses on samples of the cereal. We conducted all analyses at the Research Institute in Sciences and Health in Kinshasa. To measure water content, we dried the cereal at a temperature between 100–105°C followed by cooling in a desiccating chamber. We periodically performed weights until a stable weight was achieved. We calculated water content from the difference in weights before and after dessication. We used the Kjeldahl method for analysis of protein content (Jones & Benton 1992), by

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digesting the cereal in sulfuric acid at a high temperature using potassium sulfate and cupric sulfate as catalysts. We added concentrated alkali (sodium hydroxide) to the digest to convert ammonium to free ammonia that was distilled, collected and titrated in the presence of an acidic solution. We calculated the percentage of nitrogen from milliequivalents of ammonia per grams of sample by multiplying using a standard conversion factor then converted to crude protein content by using a second standardized conversion factor. We used the Soxhlet method to determine lipid content in which lipids were extracted from the cereal using an organic solvent by backward flow under refrigeration. We placed the product in a drying oven to evaporate the organic solvent and then weighed it.

We performed microbiologic analyses by measuring the total organism count, and testing for the presence of Enterobacteria, *Staphylococcus aureus, Salmonella, Shigella*, yeast and fungus. We plated a sample on each of the following culture mediums: MacConkey, cellulose with blood, Hektoen and Mannitol Salt culture and the media were incubated at 37°C for 24 hours. We suspended any recovered colonies in mediums for identification, including: citrate of Simmons, Kliger, and Mannitol. After incubation, we isolated fungus on Sabouraud cellulose agar. We identified all organisms based on colony morphology.

#### Maternal and Infant Acceptability

We recruited a convenience sample of five mother-infant dyads from those patients presenting to health centers in each of four communities in the rural Equateur province of the DRC. We enrolled healthy male and female infants between the ages of eight and ten months and their mothers. We excluded infants with inter-current illness that may have interfered with oral intake, infants of multiple gestation, infants with congenital anomalies, and infants who were receiving free or subsidized complementary foods.

We provided each mother with a sachet containing a 30-gram portion of caterpillar cereal. We instructed mothers to cook the cereal in 100 mL of boiling water to a puree consistency. To assess maternal acceptability, we asked mothers to rate five features of the prepared cereal: smell, taste, texture, color and consistency. Their responses were ranked on a five point Likert scale, from "dislike very much" (score of one) to "like very much" (score of five). We defined maternal acceptability as a median score for each feature of the cereal of three or greater and the upper limit of the lowest quartile of equal to or greater than two.

To assess infant acceptability, we supplied each mother with seven sachets of cereal containing 30 grams of dry cereal each, and instructed her to feed her infant one prepared portion daily. Study personnel visited the home three times during the week to reinforce preparation instructions, observe feedings and monitor for signs or symptoms of feeding intolerance. We advised mothers to save all unconsumed cereal. On the eighth day of the trial, study personnel collected all unconsumed cereal from the preceding four days and surveyed mothers about their infants' health and feeding status during the trial. Study personnel estimated the amount of cereal remaining from each daily portion. We based cereal consumption on the amount the infant consumed during the last four days of the trial. We defined infant acceptability as 100% of infants consuming greater than or equal to 75% of the cereal allotment during the last four days of the trial and all infants being free from adverse symptoms attributable to cereal consumption.

The Institutional Review Boards at the University of North Carolina at Chapel Hill and Kinshasa School of Public Health approved this study. The trial was registered through clinicaltrials.gov (NCT01258647).

## RESULTS

#### **Chemical and Microbiologic Testing**

A 30-gram portion of the cereal contained 6.9 grams of protein, 6.3 grams of fat, 12.0 grams of carbohydrate and yielded 132 kilocalories. A 30-gram portion also contained 3.8 mg of iron and 3.8 mg of zinc (Table 1). The cereal was free from *Salmonella, Shigella,* Enterobacteria, *Staphylococcus aureus*, yeast, or fungus (Table 2).

#### Maternal and Infant Acceptability

Twenty maternal-infant dyads were enrolled in the study to determine acceptability. One dyad voluntarily withdrew after enrollment. On a five-point Likert scale mothers' median scores for cereal characteristics were: overall impression 4 (range 4–5), taste 5 (3–5), smell 4 (3–5), texture 4 (3–5), color 5 (2–5), and consistency 4 (4–5) (Table 3).

All participating infants consumed more than 75% of the daily cereal portions during the last four days of the trial. Five infants (26%) consumed 100% of the cereal. One infant experienced vomiting during the first day of the study and continued the trial without further symptoms. No other adverse feeding events were reported.

## DISCUSSION

Lutter and Dewey have proposed an ideal composition for fortified complementary foods (Lutter & Dewey 2003). They recommend quantities of macronutrients in complementary foods for 6–11 month old infants that include 176 kilocalories, 3–4.5 grams of protein, and 4.8 grams of fat. A 30-gram portion of our caterpillar cereal provides their recommended daily requirements for protein and fat. If 30 grams of cereal were the sole source of complementary food, it would likely be deficient in calories. However, it appears to be a satisfactory supplement to breast milk and existing complementary foods that provide adequate energy in the form of carbohydrates.

Lutter and Dewey recommend a daily intake of 11 mg of non-heme iron for 6–11 months old infants. This amount is suggested under the assumption that the bioavailability of elemental iron in ingested non-heme iron is approximately 10% (Lutter & Dewey 2003). Less dietary iron is necessary from animal-source foods that have heme-associated iron because approximately 30% is bioavailable (Carpenter & Mahoney 1992). Heme proteins in non-insect animals are usually found in muscle in the form of myoglobin and hemoglobin, but heme is also found in cytochrome and catalases (Locke & Nichol 1992). The primary source of heme iron in caterpillars is in cytochromes, and we presume that its bioavailability is similar to the heme iron of myoglobin and hemoglobin (Locke & Nichol 1992, Chapman 1998). Insects also have iron bound to the non-heme molecules, ferritin and holoferritin. Iron associated with these proteins is typically in the ferrous state, which increases its bioavailability, and iron bound to these proteins appears to be more bioavailable than iron in

the form of reduced salts. Therefore, it is likely that the bioavailability of iron in caterpillars is similar to beef, and that the content of iron in our cereal will be sufficient to meet the requirements of infants.

Zinc is critical for cellular growth, and its deficiency is associated with stunting. Our cereal contains 3.8 mg of zinc in a daily portion, which approaches the recommended daily intake of 4–5 mg of zinc in complementary feeding products for infants (Rosado 2003). Although there are no specific quantitative data for the appropriate daily requirements of B vitamins for infants, caterpillars contain riboflavin, niacin, pantothenic acid, pyridoxine, biotin, folic acid and cobalamins (Kodondi et al. 1987).

Human sensory testing of complementary feeding products predicts the acceptability of the introduction of the product's use within target populations (Mensa-Wilmot et al. 2001). Sensory evaluation of the food product including smell, taste, and color, as well as consumption of food products during pilot testing have been described as indicators of positive acceptability (Phuka et al. 2011). We chose to evaluate both maternal acceptability and infant consumption of cereal to provide an appropriate socio-cultural framework for the introduction of this complementary food into infants' diets. Previous studies on acceptability have focused on comparing two food products to each other (Paul et al. 2008, Aaron et al. 2011). Because this cereal is a novel product made from locally available ingredients, we deemed a study comparing caterpillar cereal to a fortified cereal product which was not locally available to be unreasonable. Based on our strategy of evaluation, caterpillar cereal was found to be acceptable to mothers and infants.

Caterpillar cereal appears to be a promising alternative for animal-source foods for complementary feeding; however, we recognize some limitations to this intervention. Although caterpillar cereal is designed to be easily integrated into existing food practices by using it as an additive to the usual diet of children, the volume of cereal that we used may be a challenge for infants at 6–8 months of age to consume. Furthermore, although the iron content of caterpillars is heme-associated, it is not clear if the absorption of this micronutrient will be sufficient to prevent iron deficiency. We described the short-term microbiological and chemical profile of this cereal that was produced at the University of Kinshasa. Understanding the long-term stability and production at external sites needs to be assessed.

Using locally-available food products, we have developed a caterpillar-based cereal that has the appropriate macro-and micronutrient content for infant complementary feeding. This cereal is acceptable to both mothers and infants in a rural area of the DRC. Because the ingredients are locally available and the production of this cereal is simple, this cereal is likely to be a sustainable alternative animal-source food for complementary feeding. However, this cereal will need to be tested in an efficacy trial to determine if it will have positive effects on micronutrient deficiencies and linear growth.

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### **KEY MESSAGES:**

Locally available, micronutrient-rich complementary foods are needed in low-income countries like the Democratic Republic of Congo.

Cereal made from locally available caterpillars has an appropriate macro-and micronutrient content for infant and young child complementary feeding.

The cereal is acceptable to mothers and infants.

A study to investigate whether consuming this cereal may improve stunting of linear growth is warranted.

#### Table 1:

## Content of a 30-gram Portion of Caterpillar Cereal

Macronutrients		Micronutrients		
Energy, Kcal	≈ 132	Iron, mg	3.8	
Protein, g	6.9	Zinc, mg	3.8	
Fat, g	6.3	Magnesium, mg	9.4	
Carbohydrate, g	12.0	Copper, mg	3.7	

#### Table 2:

## Microbiologic Assays

Enrichment Media	Quantifying Media	Result
Peptone Water	Blood Agar (total organism count)	Negative
	MacConkey Agar (Enterobacteria)	Negative
	Mannitol Salt Agar (Staphylococcus aureus)	Negative
	Sabouraud Agar (yeast and fungus)	Negative
Selenite	Blood Agar (total organism count)	Negative
	Hectoen (Salmonella)	Negative
	Salmonella Shigella Agar	Negative
Thioglucolate	Blood Agar (total organism count)	Negative
	Mac Conkey Agar (Enterobacteria)	Negative
	Mannitol Salt Agar (Staphylococcus aureus)	Negative
	Sabouraud Agar (yeast and fungus)	Negative

#### Table 3:

## Maternal Acceptability

	Maternal Opinion, n					
Cereal Characteristics	Dislike very much		Neutral		Like Very Much	
	1	2	3	4	5	
Overall impression	0	0	0	11	8	
Taste	0	0	1	6	12	
Smell	0	0	1	9	9	
Texture	0	0	1	12	6	
Color	0	1	0	7	11	
Consistency	0	0	0	10	9	