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USE OF FERMENTATION AND MALTING FOR DEVELOPMENT OF READY-TO-USE COMPLEMENTARY FOOD MIXES

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ABSTRACT

Two ready-to-use complementary foods (RUCF) were formulated using fermented or malted locally available cereals and pulses. Dehydrated fruit or vegetable powders were incorporated in the RUCF. The RUCF were analyzed for physical properties, proximate constituents, selected nutrients and shelf life was studied. Moisture and protein content of the malted mix was lower than the fermented mix, but fat content was similar. Both RUCF provided 4Kcal/g. Total iron content of the fermented mix was higher (10.1mg%) than the malted mix (8.2mg%). Total zinc content of both RUCF was similar (4mg %). Calcium content of the malted RUCF (234mg%) was higher than the fermented mix (192mg%). Phytate content of both RUCF was 219 – 262 mg%. Packed bulk density of both RUCF was 0.7g/ml, water absorption capacity was 0.3ml/g. Both RUCF had >70% dispersibility. Compared to the RDA for 1 – 3 year olds, the RUCF contributed 40% energy, >60% protein, >90% iron, >80% zinc and >30% calcium of the recommended allowances for these nutrients. Thus, fermentation or malting can be used to produce RUCF that are energy dense, have low bulk density, low water absorption capacity and high dispersibility.

Key words: Ready-to-use complementary foods, fermentation, malting, ionizable iron, dispersibility, bulk density.

INTRODUCTION

Weaning is a critical period of transition from nutritious and uncontaminated breast milk to the regular family diet. During this period, infants are generally given foods made of cereals or tubers. However, these foods are bulky, low in energy and do not provide important micronutrients such as vitamin A, iron and zinc. Further, feeding such foods under poor hygienic conditions increases the risk of gastrointestinal infections and growth faltering in young children (Amankwuah et al., 2009; Khatun et al., 2013; Sharma, 2013). If such foods with low nutrient density are used as staples, children need to be fed large amounts of these foods at each feed in order to fulfil the nutrient requirements. This is generally not practicable given the small stomach capacity of infants (Maleta et al., 2004). Besides using such bulky cereal - based foods, many mothers from urban slums feed commercial weaning foods but often dilute these because of their high cost resulting in inadequate nutrient intakes (Chaudhary and Humayun, 2007, Peerkhan Nazni, Subramaniam Andal and Subramanian Pradeepa, 2009).

These limitations can be overcome by making available affordable, ready-to-use complementary foods (RUCF) that are energy-dense and do not require further processing before consumption. Household processing techniques such as roasting, germination and malting could be adopted for improvement of weaning foods. Fermentation and malting can be used to increase the energy content and nutrient density of weaning foods as well as reduce pathogen contamination of the weaning foods (Simango, 1997). Another advantage is that malting

and fermentation help to reduce anti-nutritional factors such as phytate and thereby increase mineral availability especially that of iron, deficiency of which is a public health problem in developing countries. Also, malting enhances digestibility and protein content with an increase in some amino acids such as lysine, tryptophan and methionine (Ashworth and Draper, 1992). Fermentation is a low-cost, economical technique for production and preservation of foods in developing countries, and several authors (Egounlety, 2002) have reported that simultaneous fermentation of cereals and grain legumes is useful for producing cereal and pulse - based weaning foods.

Therefore, the present study was thus undertaken to formulate two ready - to - use complementary foods (RUCF) using two processing techniques: fermentation and malting, based on locally available cereals and pulses, and spray dried fruit and vegetable powders for improving the iron content. Formulation of the RUCF was done based on the guidelines given by the Food Standards Program Codex Alimentarius Commission (FAO/WHO, 1994) on the nutritional and technical aspects of the production of formulated supplementary foods for older infants (6-12 mo) and young children (12-36 mo).

MATERIALS AND METHODS

FOOD MATERIALS

All food materials were purchased from the Agricultural Produce Marketing Corporation, the largest wholesale market located in Navi Mumbai, India; and fruit

powders were purchased from Aarkay Food Products, Ahmedabad, India.

Preparation of Mixes: All cereals and pulses were cleaned and foreign matter, damaged and shrivelled grains were removed. The mixes were prepared as shown in Figures 1 and 2. One hundred grams of each RUCF was weighed and packed in polyethylene bags that were sealed using a sealing machine.

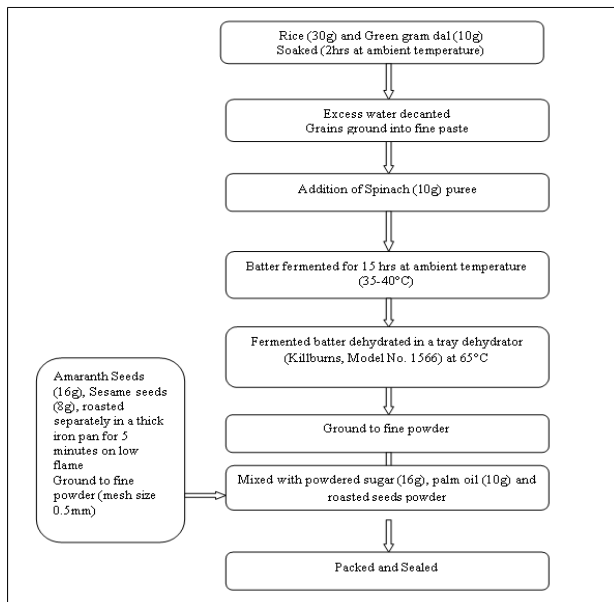


Figure 1: Flow Chart Diagram of Preparation of Fermented Mix

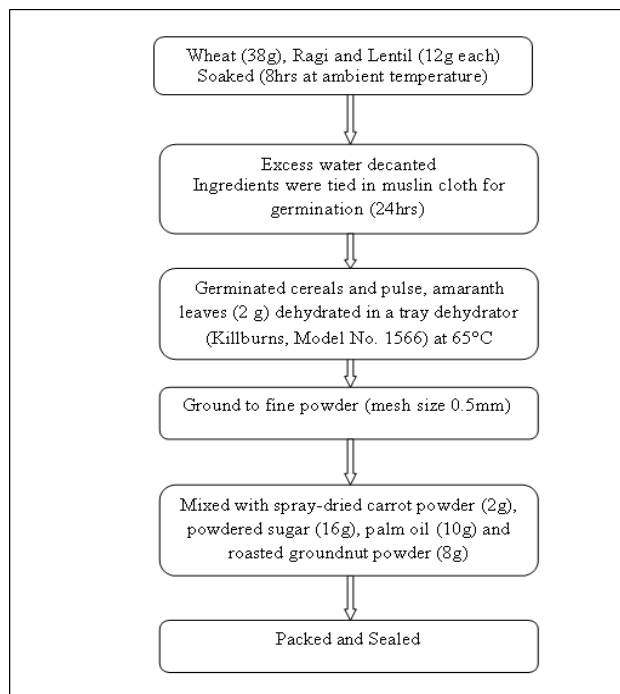


Figure 2: Flow Chart Diagram of Preparation of Malted Mix

ANALYSIS

All analyses were carried out in triplicate and the average of the three readings was calculated. All chemicals used were of AR grade. The two RUCF were analyzed for proximate composition: moisture, crude fat, total ash,

crude fibre using standard AOAC methods (AOAC, 2000). Crude protein content was estimated by the Kjeldahl method and was calculated by multiplying the total nitrogen content using factor of 6.25 (Pearson, 1976). The carbohydrate content was calculated by difference. The total energy content was calculated using Atwater factor: 4 for crude protein and carbohydrate and 9 for fat. *In vitro* starch digestibility and maltose content were estimated using the method given by Singh et al., (1982).

Ash solution was prepared by the AOAC method (2000) and estimation of total iron, zinc and calcium was done by ICP-AAS (Perkin Elmer Analyst 400 Atomic Absorption Spectrophotometer). β -carotene content was estimated spectrophotometrically (LabIndia UV 3000+) using the method given by AOAC (1980). Ionizable iron at pH 1.35 and 7.5 was estimated using the method described by Rao and Prabhavathi (1978). Extractable zinc was determined using the method given by Chrompreeda and Fields (1984) and phytate content was estimated using method given by Haug and Lantsch (1983).

PHYSICAL PROPERTIES

Packed bulk density of the RUCF was determined using the method described by Devi et al., (2012). Water absorption capacity was determined using the procedure given by Sosulski (1962) and dispersibility was determined as per the procedure described by Kulkarni, Kulkarni and Ingle (1991).

SHELF-LIFE STUDY

Packets of 100g were made using polyethylene packaging and sealed. The packets were placed at 38°C and 96% RH in a humidity chamber (model CS-03a, Classic Scientific). One week under accelerated laboratory conditions is equivalent to one month at room temperature. The packets were analyzed on days 0, 7 and 14 for moisture and peroxide value using AOAC methods (AOAC, 2000). Microbiological analysis was done using IS methods. Samples were analyzed for total plate count (IS 5402:2002), yeast and mould count (IS 5403:1999) and total coliforms by the method prescribed by Food Safety and Standards Authority of India (FSSAI, 2012).

STATISTICAL ANALYSIS

The Statistical Package for Social Sciences (SPSS Version 20.0) was used for statistical analysis. Analysis of variance was used to determine whether differences between the two RUCF were significant.

RESULTS

PROXIMATE CONSTITUENTS

The moisture content of malted mix was significantly lower than that of the fermented mix (Table 1). The crude fibre content of the fermented mix was more than twice the content in the malted mix. The protein content of both RUCF was a little more than 10g% and the fat content was approximately 11g%. Both RUCF provided 4kcal per gram of the RUCF. The contribution of protein to total energy content was 10.1% for the fermented mix and 10.5% for the malted mix. Fat contribution to total energy content was 25.3% in the

fermented mix and 24.1% in the malted mix. Approximately 65% of total energy was contributed by carbohydrate. Maltose content was approximately 2g with the fermented mix having a slightly but not significantly

higher content than the malted mix. IVSD was almost two times that of the fermented mix.

Table 1: Proximate Constituents and Nutrient Content of Fermented Mix and Malted Mix

Parameters		Fermented mix	Malted mix	F value, p	
Proximate Constituents	Moisture (g%)	7.1±0.7	4.3±0.1	36.423,0.004	
	Ash (g%)	1.3±0.2	1.2±0.03	0.290,0.619	
	Crude fibre (g%)	3.7±0.01	1.5±0.01	32834.571,0.000	
	Energy (kcal/100g)	426±4	436±2	16.149,0.016	
	Crude Protein (g%)	10.7±0.1	11.5±0.2	29.445,0.006	
	Crude Fat (g%)	12.0±0.02	11.7±0.01	369.800,0.000	
	Carbohydrate by Difference (g%)	68.8±0.9	71.2±0.3	13.711,0.021	
	In vitro Starch Digestibility (g%)	7.7±0.8	12.2±0.6	65.533,0.001	
	Maltose (g%)	2.0±0.2	1.7±0.1	2.889,0.164	
	Minerals	Total Iron (mg/ 100g)	10.1±0.5	8.2±1.2	4.337,0.106
Ionizable Iron (mg/ 100g)		pH 1.35	7.0±0.1	2.8±0.1	2002.569,0.000
		pH 7.5	7.3±0.1	5.05±0.6	27.172,0.006
Total Zinc (mg/ 100g)		4.6±0.4	4.1±0.2	2.877,0.165	
Extractable zinc (mg/ 100g)		3.3±0.42	2.1±0.4	8.407,0.044	
Calcium (mg/ 100g)		192.3±31.1	233.7±13.2	2.994,0.159	
Phytate (mg/100g)		219.3±9.2	261.8±6.0	30.009,0.005	
β-Carotene (µg/100g)		40.33±1.4	159.9±2.8	2990.245,0.000	

MICRONUTRIENT CONTENT

The iron content of the fermented mix was higher than the content in the malted mix, but there was no significant difference between the two RUCF. However, ionizable iron was significantly higher in the fermented mix compared to the malted mix. As a percentage of total iron content, it was observed that the fermented mix had a higher ionizable iron content (72.3%) than did the malted mix (61.6%). Phytate content of the fermented mix was significantly lower compared to the malted mix. Zinc content of the two RUCF was approximately 4.0mg/100g, with no significant difference between the two RUCF. Extractable zinc was significantly higher in the fermented mix compared to the malted mix and constituted 71.7% of total zinc in the fermented mix and was significantly lower (51.2%) in the malted mix. There was an inverse relationship between phytate content and ionizable iron at pH 7.5 ($r = -0.873$, $p=0.000$), but not with extractable zinc ($r = -0.664$, $p=0.151$) of the two RUCF. The malted mix had a higher calcium content compared to the fermented mix, but the difference was not statistically significant. β-carotene content in the malted mix was significantly higher compared to the fermented mix.

PHYSICAL PROPERTIES

Complementary foods are generally given in the form of gruels and prepared by mixing with water or milk. Therefore, physical properties such as bulk density, dispersibility and water absorption capacity were examined (Table 2). Bulk density and dispersibility of the fermented mix were slightly but significantly higher

compared to the malted mix. Water absorption capacity of both RUCF was similar.

Table 2: Physical Properties of the Two Ready-To-Use Complementary Foods

Parameters	Fermented Mix	Malted Mix	F value, p
Bulk Density (g/ml)	0.73±0.02	0.68±0.01	15.000,0.018
Water Absorption Capacity (ml/g)	0.35±0.05	0.33±0.05	0.192,0.684
Dispersibility (%)	75.0±0.5	73.4±0.3	21.943,0.009

SHELF LIFE STUDY

The moisture content and peroxide value of both RUCF increased during the accelerated storage (Table 3). The shelf life of both RUCF under accelerated conditions was about six weeks from the date of manufacture. At the end of two weeks, the peroxide value exceeded the limit of 10 mEq/kg as per ISI specifications (1981).

Results of microbial analysis indicated that there was an increase in the bacterial count of both RUCF at Day 14 of the shelf life study, although the count was within the limit given by FSSAI (2012) of less than 10000 per gram (Table 3). Yeast and mould count should be absent in 0.1 gram of sample as given by FSSAI (2012). In the present study, yeast and mould count in both RUCF increased beyond the limit specified by FSSAI. No coliforms were detected.

Table 3: Results of Accelerated Shelf Life Study of the Two Ready-To-Use-Complementary Foods

Parameters	Fermented Mix			Malted Mix		
	Day 0	Day 7	Day 14	Day 0	Day 7	Day 14
Chemical Analysis						
Moisture (g%)	3.50	5.60	7.38	4.10	4.30	6.20
Peroxide Value (mEq/kg)	3.70	6.90	10.99	5.12	6.60	9.94
Microbial Analysis						
Total Plate Count (cfu/g)	1.1x10 ²	5.90x10 ²	21.38x10 ²	4.7 x 10 ²	31.27x10 ²	62.27x10 ²
Total Coliform Count (cfu/g)	Absent	Absent	Absent	Absent	Absent	Absent
Total Yeast and Mould Count (cfu/0.1g)	140	827	2110	170	2636	3636

Table 4: Percent Recommended Dietary Allowances (RDA) Fulfilled by 100 g of the Ready-to-Use-Complementary Foods

Nutrients	RDA, 2010	% RDA Fulfilled	
		Fermented Mix	Malted Mix
Energy (Kcal)	1060	40.2	41.2
Protein (g)	16.7	64.0	68.8
Fat (g)	27	44.4	43.4
Iron (mg)	9	112.0	91.2
Zinc (mg)	5	92.0	82.0
Calcium (mg)	600	32.0	40.0
β-carotene (µg)	3200	1.2	5.0

DISCUSSION

Low moisture content in complementary foods is important to prevent nutrient losses and to ensure shelf life of the product (Amankwah et al., 2009). The Protein Advisory Group (PAG, 1971) recommended that moisture content should not exceed 10% in order to keep a floury product for a reasonably long time (PAG, 1971). The moisture content of the RUCF developed in the present study was below the limit, recommended by the PAG. The moisture content of both RUCF was higher than values of 1.5g% to 2.5g% reported by Kumkum et al., (2010) and Salve et al., (2011) who developed mixes containing roasted ingredients. The higher moisture content of the mixes in the present study may be due to the processing technique used. Kumkum et al., (2010) roasted each ingredient that may have reduced the moisture content. Recently, Ijarotimi and Keshinro (2013) and Ikujuola and Adurotoye (2014) reported that the moisture content of weaning foods prepared from fermented popcorn, bambara groundnut and malted quality protein mix and cowpea ranged from 5.68% to 10.70%. These values are in the range of values observed in the present study.

The ash content of the two RUCF was within maximum limit of 5g% specified by the FSSAI (2011). The Protein Advisory Group (1971) has recommended that the crude fibre content of infant foods should be less than 5g%. The crude fibre content in the two RUCF prepared in the present study is within the recommended level, although the fermented mix had a significantly higher fibre content than the malted mix. This was due to incorporation of spinach and sesame seeds in the former. The low crude fibre content would enable children to consume food that is more nutrient dense and help to meet their daily energy

and other vital nutrients requirements (Ijarotimi and Keshinro, 2013).

The energy content of the two RUCF was as per the guidelines given by the Codex Alimentarius for complementary foods i.e. at least 4kcal/g of dry mix (FAO/WHO, 1994). Also it was comparable to the energy content of weaning foods prepared from fermented popcorn by Ijarotimi and Keshinro (2013).

Germination, malting and fermentation has been reported to improve the protein content and quality of food products (Fasasi, 2009). In the present study, the malted mix had a higher protein content compared to the fermented mix. Inyang and Zakari (2008) reported that the increase in protein content during malting is attributable to net synthesis of enzymic protein by the germinating seeds.

Fat content of the two RUCF met the minimum requirement of 10 – 25g% recommended by WHO (2001) for infant foods. Fat content of similar mixes developed by other investigators (Amankwah et al., 2009; Ghasemzadeh and Ghavidel, 2011; Ahmad, Gupta and Srivastava, 2013; Khatun et al., 2013; Sharma, 2013) varied widely from as little as 0.78g% to as much as 10.6g%. Higher fat content is nutritionally advantageous because it can increase the energy content. However it can reduce the shelf life and stability of the product, since unsaturated oils are vulnerable to oxidative rancidity. One possibility for the relatively short shelf life of the RUCF in present study, as indicated by their peroxide value could be due to the amount of oil used. It would be worthwhile to examine whether shelf life can be extended by replacing oil with fat powder and using packaging material that is impermeable to gases and moisture. This would be worthwhile also from the perspective of controlling microbial growth. In the present study high moisture content may have been responsible for rapid microbial growth during the shelf life study.

The carbohydrate content of the RUCF in the present study is in line with the values reported in germinated formulations (60.2g% to 69.6g%) by Imtiaz et al., (2011). However, some investigators have reported lower carbohydrate content (Amankwah et al., (2009) - 60.9g%-62.0g% and Khatun et al., (2013) - 33.4g% – 64.1g%). These differences in the carbohydrate content are because these investigators have used less sugar and ingredients that may contribute more protein such as soy and fish by Amankwah et al., (2009) and milk powder by Khatun et al., (2013). Other investigators have reported a higher carbohydrate content than the values observed in the present study: 77.2g% – 85.2g% (Adebayo et al.,

2012), 73.6g% (Ahmad, Gupta and Srivastava, 2013) and 81.8g% (Sharma, 2013).

One important advantage of fermentation and germination is the activation of α – amylase by which starch is hydrolyzed to simpler sugars. (Ikujenlola and Adurotoye, 2014). Gokavi and Malleshi (2000) reported that during germination, partial degradation of amylopectin occurs and amylose content increases. This process would be reflected in higher starch digestibility and formation of maltose. Exposure to heat causes hydrolysis of amylose and amylopectin to maltose and dextrin that also reduces viscosity of thick gruels (Gibson et al., 2006). In the present study, IVSD of the malted mix was higher compared to the fermented mix. Chau and Cheung (1997) reported that germination, which is a primary step in malting, significantly improved the IVSD content in sorghum. Also, solubility of the cell wall, partial digestion of starch and degradation of amylase inhibitors enhance the overall carbohydrate digestibility on malting (Choudhary, Das and Baroova, 2011).

The iron contents of both the RUCF were much higher, than the 5 mg% specified by the FSSAI (2011), with the fermented mix having significantly higher iron than the malted mix. Also the content was higher than iron content (3.0 mg% to 6.5 mg%) reported by other researchers in composite weaning foods containing cereals, malted legumes and vegetable powders (Ghasemzadeh and Ghavidel, 2011).

Bioavailability of iron is an important consideration since the absorption of iron from typical mixed cereal – pulse vegetarian diets is 3 – 5% and is a contributory factor for iron deficiency anaemia (ICMR, 2010). Hence, fermentation and malting were employed in order to improve availability of iron. The fermented mix had higher ionisable iron compared to the malted mix at pH 7.5 which is close to the pH of the site of iron absorption from the human intestines. In the two mixes percent ionizable iron was very high 72.3% in the fermented mix and 61.6% in the malted mix. Several reports in the literature indicate that iron availability increases after germination and fermentation (Rao, 1994; Deosthale, 2002; Afify et al., 2011).

Germination and fermentation decrease the phytin phosphorus content (Afify *et al.*, 2011; Suma and Urooj, 2014, Nazni *et al.*, 2010). In the present study, a strong correlation was seen between available iron and phytate content of RUCF, although the same trend was not seen for zinc. The decrease in phytin phosphorus content during sprouting can be due to leaching out of phytate into water used for soaking under the influence of a concentration gradient. Further, during cooking, phytic acid may decrease due to the formation of insoluble complexes between phytates and other components and also to the breakdown of phytic acid content at high temperatures. Also activation of phytase by both process probably contribute to degradation of phytate.

Bulk density is an important physical property. It depends on the particle size of the ingredients, with smaller particle size, being associated with lower bulk density (Okorie et al., 2011). High bulk density is a disadvantage in case of RUCF because it can limit the nutrient intake per feeds (Isah, Danladi and Ejike, 2013).

Bulk densities of the RUCFs developed in the present study were similar to the values reported by other investigators (Kumkum et al., 2010; Ghasemzadeh and Ghavidel, 2011; Ikujenlola et al., 2013; Khatun et al., 2013; Ikujenlola and Adurotoye, 2014). Both fermentation and germination have been shown to reduce bulk density (Ikujenlola and Adurotoye, 2014). The decrease in bulk density after malting and fermentation would be advantageous in the preparation of infant gruels from such mixes. Germination and fermentation also reduces water absorption capacity (Ikujenlola and Adurotoye, 2014). Lower water absorption capacity is desirable for making gruels in which more flour can be added per unit volume of the gruel. This would help to increase the energy density and nutrient content of the infant foods. The reduction in bulk density and water absorption capacity reflects the breakdown of starch to its constituents such as dextrin and maltose that absorb less water when cooked (Desikachar, 1980; Ikujenlola et al., 2013).

Further, the percentage of the nutrient daily allowances recommended by the Indian Council of Medical Research (2010) for 1 to 3 year old children supplied by the two RUCF were compared (Table 4). The mixes were similar in their contribution to nutrient intakes and were dense enough because 100g of the mixes could meet two – thirds of the protein requirement, at least one – third of calcium requirement, approximately 80% of zinc requirement and almost all of the iron requirements.

CONCLUSION

The results of the present study showed that ready-to-use complementary foods prepared by either fermentation or malting and using locally available food ingredients can be produced as they are energy dense, and have low bulk density, low water absorption capacity and high dispersibility. These RUCF need to be popularized among mothers especially. Inclusion of spray dried fruit and/ or vegetable powders can improve the micronutrient content of the RUCF. The advantage of such RUCF is that they can be stored; ready for consumption and malted foods improves nutritional value by reducing the antinutritional factors. However, extension of shelf life of the RUCF needs to be studied by using different packaging material after analysis in the laboratory under simulated conditions. Further to determine the feasibility of producing the RUCF commercially at a large scale merits attention.

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