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PRODUCTION OF LOW PHYTIC ACID WEANING FOODS BASED ON WHEAT GRAINS:
II - EVALUATION OF PRODUCED NOVEL WEANING FOOD IN COMPARISON WITH COMMERCIAL PRODUCTS

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ABSTRACT

This study aimed to evaluate produced weaning food based on wheat grains low in its content of phytic acid PA which inhibits the minerals absorption such as Ca, Fe and Zn. These minerals are of special importance in infants nutrition. A laboratory formula of weaning food was prepared named N with low content of PA by naturally activation of phytase. It was compared with 6 samples of commercial weaning foods from local markets. Phytic acid, phosphorus and its derivatives and the absorption of previous minerals were determined in all samples.

The results showed that PA in commercial weaning food samples ranged from 73.798 to 352.642 mg/100g., while reduced to be 6.670mg/100g for N sample. The percentage of phytate phosphorus to total phosphorus ranged from 7.150 – 20.161 % for the commercial samples, but it was just 0.520 % for the N sample. The phytate: Ca, Fe and Zn molar ratio ranged from 0.010 to 0.214 , 0.921 to 3.842 and 2.140 to 3.810 respectively for the commercial samples, while the N sample recorded 0.001 , 0.038 and 0.091 respectively.

This study was carried out to evaluate produced novel weaning food based on wheat grains low in PA by activation of naturally occurring phytase. Also, this novel product was compared with some commercial weaning products.

INTRODUCTION

Infancy is one of the most important periods of life as the infants have a restricted number of foods. Weaning foods are the first solid foods, often based on cereal and combined with milk or with legumes, are used to improve the quantity and quality of the protein component.

Weaning foods based on cereal and legumes often contain considerable amount of phytic acid, potentially impairing the mineral and trace element bioavailability.

Phytate of whole-wheat flour has a significant inhibitory effect on calcium, iron, zinc and selenium absorption. The inhibition of minerals bioavailability depends on the total amount of phytate present in the diet (Puspa *et al.,* 1994). Therefore weaning food based on cereal and legumes should be preferably low in phytic acid. Phytic acid can be degraded by the enzyme phytase (*myo*-inositol hexakisphosphate phosphohydrolase), which occurs in cereal grains and legume seed as well as in some microorganisms. Microbial phytase is commonly used for phytic acid degradation of animals feed but is not used for foods prepared for human consumption (Simell *et al.,* 1991 and Cromwell *et al.,* 1993).

MATERIALS AND METHODS

1-Materials

1-1- Commercial weaning foods

Six samples of commercial baby foods based on cereal products were collected from local market at Mansoura City, Egypt. Samples trade names were, Jotis (A) {wheat flour, full cream milk powder, sugar, skimmed milk powder, vitamins, iron fumarate}.,Nestle infant cereal wheat (B) {wheat flour, sucrose, di-basic potassium – phosphate, calcium carbonate, ferrous fumarate, vanillin, and vitamins}., Hipp (C) {apple juice with lower acidity, apple, banana and oranges, juice, wheat, oats, palm oil and vitamin C}., Nestle cereal wheat-4fruits (D) { Wheat flour, skimmed milk powder, sucrose, palm oil, guava, banana, corn oil, apple, mango, calcium, carbonate, iron phosphate, vanillin, and vitamins}., Garber (E) {wheat flour, skimmed milk powder, sucrose, malto dextrine, Soya flour, date puree, lactose, emulsifiers, antioxidants vitamins and minerals}. and Nestle cerelac + 3 vegetables (F) {Wheat flour, skimmed milk powder, sucrose, palm oil, carrots, tomatoes, corn oil, calcium carbonate, ferrous fumarate, vanillin, vitamins}. All samples were stored at (4°C) prior to analysis.

1-2- Wheat grains

Preparation of wheat samples by laboratory whole-wheat grains (Triticum *aestivum,* L.varites sakha 61) were obtained from Agronomy Department, Sakha Agric., sakha El-sheikh, Kafr- El-sheikh Province, Egypt. The amount used in this investigation was 20 Kg. the grains were cleaned and stored at (4° c) till used.

1-3-Skimmed milk powder.

Skimmed milk powder was obtained from Misr Dairy Product Company, Mansoura City, Egypt.

1-4-Minerals

Sodium selenate anhydrous was obtained from Sigma company. zinc sulphate, ferrous sulphate and calcium carbonate were obtained from El Nasr Pharmaceutical Chemicals, Cairo.

2- Methods

2-1-Preparation of novel weaning food (N):

About 250g of wheat grains were cleaned, milled and suspended in water(1 : 2 w/w). The slurry was adjusted to pH 5.1 to activate phytase enzyme by adding different acids (lactic, acetic, ascorbic, citric acids), lime juice and whey. Samples were incubated at 50 – 55 °C for 120 minutes, dried at 70-75°C, ground in Moulinex Wiley, packaged in polyethylene pags then stored at 4°C till analysis.

N formula consists of 70% whole wheat flour + 30% skim milk mixed and enriched with 6.04mg/100g $ZnSo4$, 28.33mg/100g $FeSo4$, 93.75mg/100g $CaCO3$ and 1.8mg/100g sodium selenat.

3- Analytical Methods

3-1- Chemical composition

Moisture, crude fat, crude protein, ash and crude fiber contents were determined according to AOAC (1990).

Total carbohydrates content was calculated by difference between 100 and the sum of (crude protein, crude fat, ash and fiber contents).

3-2-Determination of minerals:

Samples were prepared for minerals determination by digestion in percholoric acid and nitric acid according to Pupsa *et al.* (1994). Calcium, iron and zinc contents were determined by using atomic absorption spectrophotometer (Perkins-Elmer, Model 2380) according to Pupsa *et al.* (1994).

Total phosphorus (TP) content was determined using the spectrophotometric method based on the formation of phosphorus molybdate complex (AOAC, 1990).

For determination of inorganic phosphorus (IP) the sample was extracted with 5% trichloroacetic acid as out lined by Belavady & Banerjee (1953) and phosphorus compound was determined using the spectrophtometric method (Spekol, 11) that is based on the formation of phosphorus molybdate complex (AOAC, 1990).

Organic phosphorus (OP) was calculated by difference between total phosphorus and the sum of inorganic phosphorus and phytate phosphorus.

3-3-Phytate content:

Phytate content was determined calorimetrically for iron present in the ferric phytate with potassium thiyocyanate by the procedure of Wheeler and Ferrel (1971) modified by Harland and Oberless (1977).

3-4- Phytate: Ca, Fe and Zn molar ratio calculation.

These values were calculated according to Morris and Ellis (1980).

RESULTS AND DISCUSSION

Preparation of low PA wheat flour

An experiment was carried out to prepare low PA wheat flour to produce the novel product N. Different acidulents namely acetic, lactic, citric, ascorbic acid, lime-juice and whey were used to adjust the pH to 5.1. The results were given in Table (1).

From these results, it was shown that the best product was obtained by the lactic acid treatment, where PA content was the lowest value (9.534 ± 5.647 mg/100g) compared with other acidulents used (Table 1). Also, it was noticed that retention of PA reduced from 100% in control sample to be 1.476% in the chosen sample. This sample was utilized in production of novel weaning food.

Table (1): Changes in PA content in whole wheat flour as affected by different acidulents.

Samples \ Components	Phytic acid* mg/100g	Phytic acid retention
Control	646.01 ± 17.18	100%
Acetic	22.729 ± 6.840	3.52
Lactic	9.534 ± 5.647	1.476
Citric	300.399 ± 34.720	46.5
Ascorbic	91.453 ± 16.730	14.16
Lime juice	23.840 ± 5.72	3.69
Whey	59.157 ± 17.844	9.16

Chemical composition of novel weaning foods:

Chemical composition and minerals content of both novel and commercial weaning foods according to (lab) analysis were illustrated in Table (2).

Sample N had the higher protein content (18%) comparing with the commercial samples. According to the FAO/WHO (1994) for processed cereal-based foods for infants and children, it defines a minimum crude protein content by 15% for dry cereals.

Table (2) showed that sample N had low content of fat (2.624%) and approximately the same value of carbohydrates (75.266%) compared with the other samples. The fiber content of N sample was 2.990% it was within the recommendations of FAO (1985), where the maximal level of 5% crude fiber had been suggested (Table 2).

Table (2) revealed that iron content ranged from 5.203 to 30.400mg/100g for commercial samples but it was 15mg/100g for N sample. For zinc, N sample had the highest content being 7 mg/100g. N sample had moderate contents of both calcium and phosphorus among the commercial samples, its values were 365 , 363.920 mg/100g respectively. Selenium content recorded 0.3μg/g.

Phytic acid and phosphorus derivatives:-

Phytic acid and phosphorus derivatives in commercial and novel weaning foods in the comparison illustrated in Table (3).

From the data in Table (3), in sample A, phytic acid (PA) was 101.764±4.33 mg/100g. depending on its ingredients, wheat variety and extraction rate. Data revealed that low extraction had a lower amount of PA. Because most of PA naturally is located in pericarp or aleurone layer (Fulcher, 1982). With sample B, PA was 120.954±16.644 mg/100g.. The amount of PA considered medium as a whole but higher than that of sample A, this may be due to wheat variety and extraction rate as mentioned before. Fiber content in this sample was 1.65% (Table 2) according to (Lab) analysis therefore, the amount of PA was higher. Sample C, as illustrated in Table (3) PA was 77.689±10.268 mg/100g. PA amount was lower may due to small content of wheat and oats. Similarly, PA content for sample D was presented in Table (3) and its value was 73.798±7.185 mg / 100g.

Table (2): Chemical composition and minerals content of commercial and novel weaning food samples (on dry weight basis).

Component \ Sample		A	B	C	D	E	F	N
Protein	%	14.7	7.8	14.3	13.26	16.61	13.3	18
Fat	%	9.9	1.39	5.09	6.33	2.4	5.17	2.624
Carbohydrate	%	71.82	87.789	85.36	75.79	75.39	78.07	75.266
Energy	Kcal	435.18	394.866	404.49	413.17	389.6	412.01	396.68
Ash	%	3.58	1.301	0.38	3.01	2.9	2.46	1.12
Fiber	%	00	1.72	4.86	1.61	2.7	1.00	2.99
Iron	mg/100g	6.138	8.325	30.4	6.231	5.203	8.390	15
Zinc	mg/100g	4.092	3.122	16	3.115	5.203	4.195	7
Calcium	mg/100g	383.632	260.146	118.9	451.713	426.639	429.995	365
Phosphorus	mg/100g	350.89	217.066	162.7	290.76	329.24	261.35	363.92
Selenium	µ/g	0.205	0.312	0.54	0.207	0.3122	0.21	0.3
Moisture	%	2.25	3.9	81.5	3.7	3.9	4.65	5.33

Table (3): Phytic acid and phosphorus derivatives content in commercial and novel weaning foods (on dry weight basis).

Sample \ Component	Phytic acid* mg/100g	Total phosphorus mg/100g	Phytate * phosphorus mg/100g	In organic phosphorus mg/100g	Organic phosphorus mg/100g **	% PP To TP
A	101.764 ± 4.33	350.89	28.61 ± 1.22	134.63	187.6	8.170
B	120.954 ± 16.664	217.066	34.072 ± 4.694	107.25	75.744	15.696
C	77.689 ± 10.268	162.70	21.884 ± 2.893	47.30	93.516	13.451
D	73.798 ± 7.185	290.76	20.788 ± 2.024	106.45	163.522	7.15
E	235.642 ± 19.174	329.24	66.38 ± 5.4	134.63	128.232	20.161
F	91.08 ± 12.38	261.35	25.6 ± 3.49	106.45	129.243	9.817
N	6.67 ±3.95	363.92	1.895 ±1.1	98.259	263.766	0.52

* Means and SD for four replicates.

** Other than phytate phosphorus.

Data revealed that, the amount of PA considered lower comparing with other sample due to sample ingredients which contain wheat flour and fruits (guava, banana, apple, mango) from labeling data. Tabekha and Domah (1977) stated that, vegetables and fruits are considered very poor sources of phytic acid and many of them are completely devoid of it. PA value for sample E (Table 3) was 235.642±19.174 mg/100g. This sample contained the highest amount of PA comparing with other samples due to soya flour presented in its ingredients (from labeling data) which considered high source of PA (Khokhar *et al.*, 1994).

PA content for sample F was 91.08 ± 12.38 mg/100g. This amount of PA was considered medium in comparing with other samples, it was associated with extraction rate of wheat flour and its ingredients.

The major difference between the novel and commercial weaning foods was PA content, which reduced to 6.67 mg/100g for N sample (Table 3).

Table (3) indicated that phytate phosphorus (PP) content was associated with PA therefore, the high amount of PA resulting in high amount in PP. It was obvious that, PP content ranged from 66.38±5.4 to 20.788±2.024mg/100g for E and D samples respectively comparing with N sample which contained 1.895 ± 1.100mg/100g.

Data presented in Table (3) showed that, the total phosphorus (TP) content ranged from 162.700 to 363.920mg/100g for C and N samples respectively, meaning that N sample had the highest value of TP. The variation in TP content is due to samples components and addition of chemical sources of phosphorus.

As shown in Table (3), inorganic phosphorus (IP) content for commercial samples ranged from 47.300 to 134.630mg/100g for samples C and both A and E respectively, while N sample recorded 98.259mg/100g. IP content essentially depending on the presence chemical sources of phosphorus.

From the same Table it was noticed that N sample had recorded the highest content of organic phosphorus (OP) (263.766 mg/100g) whereas B sample recorded the lowest value 75.744mg/100g

Finally % PP to TP was calculated and presented in Table (3), indicating that N sample realized the lowest percent of PP to TP.

From Fig. (1) It was illustrated that the novel weaning food N had the lowest amount of PA and % PP to TP was the better one and the high amount of TP comparing with the other commercial samples.

Phytate: Ca, Fe and Zn molar ratio.

The phytate: Ca molar ratio of the six tested commercial And novel weaning foods used in this study were shown in Table (4). It was ranged from 0.001 to 0.214 for N and C samples respectively. Infants consuming the sample No (C) may be suffering from calcium deficiency. Because the availability of calcium increases with the reduction index of phytate: Ca molar ratio. These results are in agreement with those reported by (Morris and Ellis, 1985) who stated that, persons consuming diets with phytate: Ca molar ratio > 0.2 may be at risk of calcium deficiency.

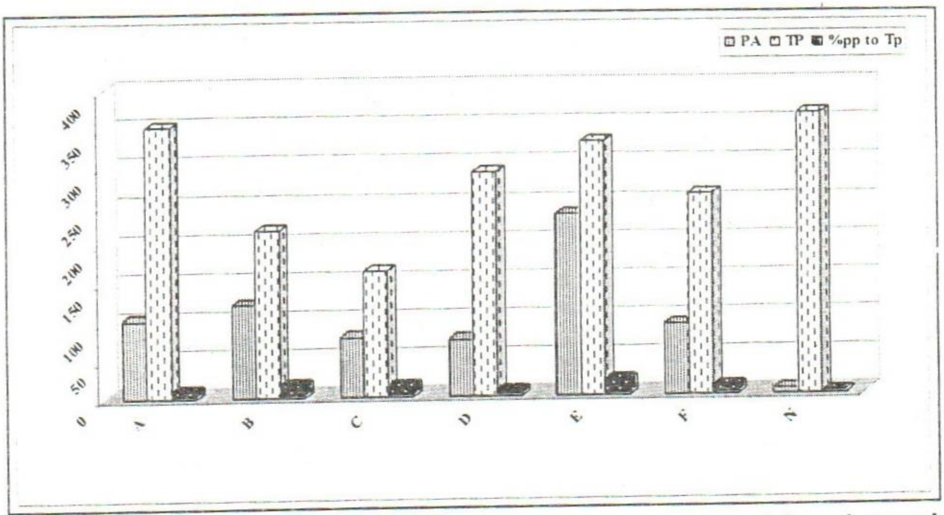

Fig. (1): Phytic acid, TP and % PP to TP in commercial and novel weaning food.

Table (4): Phytate: Ca, Fe and Zn molar ratio

Samples	Phytate: Ca molar ratio	phytate: Fe molar ratio	Phytate: Zn molar ratio
A	0.016	1.406	2.450
B	0.029	1.232	3.810
C	0.214	1.098	2.550
D	0.010	1.005	2.330
E	0.034	3.842	2.460
F	0.013	0.921	2.140
N	0.001	0.038	0.091

As shown in Table (4) phytate: Fe molar ratio was calculated for all samples used in this study. It was ranged from 0.038 to 3.842 for N and E samples respectively. As for Ca, PA decreases Fe solubility (Sandberg & Svanberg, 1991) and the inhibition of Fe absorption is closely related to the content of phytate in bread as example (Brune *et al.,* 1992).

No available data obtained for phytate: Fe molar ratio index as comparing with phytate: Ca molar ratio. From the results, it could be concluded that a lower phytate molar ratio is better for Fe absorption.

The phytate: Zn molar ratio values of commercial weaning food samples presented in Table (4) showing that N sample was the lowest value (0.091) comparing with the commercial. The inhibitory effect of phytate on Zn absorption observed by several researchers (Oberleas, 1983; Turnlund *et al.,* 1984; Sandstrom *et al.,* 1987, Kivisto *et al.,* 1989 and Sandstrom *et al.,* 1990). They observed that, the abcorption of extrinsically labeled Zn^{65} was negatively correlated to the phytic acid content of the meal. The molar ratio between phytic acid and Zn was suggested as an index of Zn bioavailability the same situation noticed with the obtained data.

Morris and Ellis, (1980) reported that with moderate Ca intake, phytate: Zn^{2+} molar ratio < 10 are likely to provide an adequate available Zn and molar ratio > 10 are associated with symptoms of Zn deficiency such as growth rate depression. However, Wise (1995) doubted the predictive use of this ratio, derived from animal studies, for humans.

On our observation samples differ among them in chemical composition and minerals content. Although these samples contain high amounts of minerals they contain significant amount of phytic acid either, which reduces minerals availability for infants. These results are in agreement with those reported by Egli (2001) who stated that the PA content (0.4g/100g) of non-dephytinized weaning foods represents a level, which can be expected, in many commercial weaning products. Analysis of thirty cereal-based weaning foods available in Switzerland resulted in a mean PA content 0.3g/100g (range 0.02 to 1.02g/100g). Low zinc bioavailability from many of these products can be expected. So PA should be completely reduced (<3 mg/100g.) in the weaning foods to avoid its effect on minerals bioavailability (dephytinized weaning foods).

From the phytate: Ca, Fe and Zn molar ratios of novel weaning food it was predicted that there was no influence of low phytic acid on minerals absorption comparing with commercial weaning foods samples, (Table 4).

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إنتاج أغذية الفطام المكونة أساسا من حبوب القمح ومنخفضة فى حامض الفيتيك:
٢ – تقييم منتج غذاء الفطام الجديد ومقارنة بالمنتجات التجارية

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هذه الدراسة تهدف إلي تقييم تركيبة من أغذية أطفال (أغذية الفطام) مصنعة أساسا من حبوب القمح ومنخفضة في محتواها من حامض الفيتيك الذي يثبط امتصاص المعادن مثل الكالسيوم والحديد والزنك. هذه المعادن لها أهمية خاصة في تغذية هؤلاء الأطفال

تم تحضير تركيبة من أغذية الأطفال في المعمل ويرمز لها بالرمز (N) منخفضة في محتواها من حامض الفيتيك عن طريق تنشيط أنزيم الفيتيز طبيعيا. وقد جري مقارنة هذه العينة مع ستة عينات من أغذية الأطفال التجارية الموجودة في الأسواق المحلية. وتم تقدير حامض الفيتيك ومشتقاته ومقدار امتصاص المعادن السابقة في كل العينات.

وقد أظهرت النتائج أن كمية حامض الفيتيك في أغذية الفطام التجارية تراوحت ما بين ٧٣,٧٩٨ – ٣٥٢,٦٤٢ مجم/١٠٠جم بينما انخفضت هذه النسبة في التركيبة المعملية (N) إلي ٦,٦٧مجم/١٠٠جم . أما بالنسبة للفوسفور فقد بلغت النسبة المئوية لفوسفور الفيتات إلي الفوسفور الكلي من ٧,١٥ – ٢٠,١٦١ % بينما العينة المعملية (N) سجلت ٠,٥٢% فقط. و تراوحت النسبة المولارية بين الفيتات وكلا من الكالسيوم والحديد والزنك بين (٠,٠١ – ٠,٢١٤) و (٠,٩٢١ – ٣,٨٤٢) و (٠,٢١٤ _ ٣,٨١) علي الترتيب بالنسبة للأغذية التجارية وسجلت التركيبة المعملية (N) ٠,٠٠١ و ٠,٣٨ و ٠,٠٩١ علي التوالي.