

Extrusion cooking of a high-fibre cereal product

2. Effects on apparent absorption of zinc, iron, calcium, magnesium and phosphorus in humans

BY BARBRO KIVISTÖ, HENRIK ANDERSSON,
GITTE CEDERBLAD, ANN-SOFIE SANDBERG
AND BRITTMARIE SANDSTRÖM

*Department of Clinical Nutrition, Gothenburg University, Annedalsklinikerna,
S-413 45 Gothenburg, Sweden*

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1. The effect of extrusion cooking, using mild conditions, of a high-fibre cereal product on apparent small bowel absorption of zinc, iron, calcium, magnesium and phosphorus was studied.
2. Seven ileostomy subjects were studied during two periods (each of 4 d), on a constant low-fibre diet supplemented with either 54 g/d of a bran-gluten-starch mixture or the corresponding extruded product.
3. The apparent absorption of Zn, Mg and P was significantly decreased ($P < 0.05$) during the period with extruded product compared with the period with bran-gluten-starch. No difference was found for Fe and Ca.
4. The negative effect of extrusion cooking of a product containing phytic acid on availability of Zn, Mg and P was small but could be of nutritional relevance in foodstuffs that are consumed frequently and in infant formulas.

Extrusion cooking is generally a high-temperature short-time (HTST) process using high shear at elevated pressure and temperature and can be used to give desirable texture to many products. It is becoming increasingly popular as a method for giving texture to vegetable proteins and as a heat treatment of starchy products. It is used for a wide variety of foods such as vegetable proteins, breakfast cereals, weaning foods, crispbread, snacks and sweets. Several of these products contain appreciable amounts of dietary fibre and phytic acid, which have been associated with decreased mineral availability (McCance & Widdowson, 1942; Reinhold *et al.* 1976; Sandberg *et al.* 1982; Nävert *et al.* 1985).

Westerlund & Theander (1983) have reported an increase in dietary fibre after extrusion of wheat flour; the more severe the extrusion conditions, the greater the increase in fibre. Formation of amylose-lipid complexes by extrusion cooking of cereal starches have been observed (Mercier, 1980). To what extent minerals are bound to the formed complexes is not known.

The aim of the present study was to determine whether biological availability of zinc, iron, calcium, magnesium and phosphorus is impaired by extrusion cooking of a fibre-rich product. Such an effect could be suspected as we have found that phytic acid is not digested to the same extent after extrusion cooking as in a non-extruded product (Sandberg *et al.* 1986).

SUBJECTS AND METHODS

Subjects

Four men and three women volunteered for the study. Sex, age, diagnosis and serum values at the start of the study are shown in Table 1. All the subjects had previously been proctocolectomized and had well-established ileostomies with only a minor portion of the terminal ileum (30-50 mm) removed. The ileostomies functioned properly and the volumes of ileostomy fluid were not excessively high. No drugs were taken during the study.

All values for serum Zn, Ca and total Fe-binding capacity (TIBC) were within the

Table 1. Sex, age, diagnosis, blood-haemoglobin (B-Hb) and serum values of the ileostomy subjects

Subject no.	Sex	Age (years)	Diagnosis	B-Hb (g/l)	Zinc (μ mol/l)	Iron (μ mol/l)	Calcium (mmol/l)	Magnesium (mmol/l)	Phosphorus (mmol/l)	TIBC (μ mol/l)
1	♂	54	Ulcerative colitis	156	15	18	2.3	0.8	1.0	57
2	♀	30	Crohn's colitis	130	13	6.2	2.3	0.9	1.2	67
3	♂	58	Ulcerative colitis	169	14	18	2.3	0.6	1.0	70
4	♂	49	Ulcerative colitis	144	15	22	2.4	1.0	0.8	61
5	♂	39	Crohn's colitis	151	12	10	2.3	0.8	1.0	77
6	♀	39	Crohn's colitis	137	—	7.2	2.4	0.8	1.5	63
7	♀	23	Crohn's colitis	138	12	18	2.2	0.7	1.5	63
Reference values	♂	—	—	116-149	9-15	10-30	2.2-2.6	0.7-1.2	0.8-1.4	43-90
	♀	—	—	132-166	9-18	15-35				

TIBC, Total Fe-binding capacity.

reference ranges. Three subjects had low serum Fe values, but fell within the reference range for TIBC and haemoglobin (Hb).

Experimental model

Each subject was studied for two 4 d periods eating a constant low-fibre diet with the addition of either 54 g/d of a bran–gluten–starch mixture (period A) or the corresponding extruded product (period B) as described previously (Sandberg *et al.* 1986). The subjects were weighed at the start and end of each period and fasting blood samples were obtained at the beginning of the study for Zn, Fe, Ca, Mg, P, TIBC and Hb analyses. The volunteers collected all ileostomy contents during the two periods. They changed ileostomy bags every 2 h except for the bag used during the night, which was kept until the morning. Each bag was immediately sealed and frozen in a vessel containing dry-ice kept by the volunteer. The deep-frozen ileostomy bags were delivered to the laboratory daily. The contents of each bag were freeze-dried in the ileostomy bag. The freeze-dried ileostomy contents from each separate day were pooled and homogenized and portions taken for analysis.

Diets

The same daily menu was served each day during the two periods. The diet was a constant low-fibre diet with the addition of 32.4 g wheat starch, 16.2 g wheat bran and 5.4 g gluten (period A) or 54 g of the extruded bread-like product (period B). Gluten and starch were mixed and heated with rice for 5 min to permit gelatinization of the starch. Just before serving, the bran was added. The extrusion was done using mild conditions. For details of the diet and the extrusion cooking, see Sandberg *et al.* (1986). All food was prepared in advance in the metabolic-ward kitchen and care was taken not to contaminate the food with minerals. The utensils used for preparing the meals were made of plastic, Teflon or stainless steel. Only deionized water was allowed for cooking purposes. Windows and doors to the metabolic-ward kitchen were kept closed during preparation of the food to keep the kitchen as free from dust as possible and no food was left uncovered longer than necessary. Each subject was given daily 2 litres deionized water in a plastic bottle for drinking. The individual food portions were kept frozen and reheated in aluminium vessels. The subjects were instructed to bring back any uneaten food.

Two duplicate portions of the daily menu of each period per subject were homogenized and freeze-dried and portions taken for analysis.

Chemical analyses

Portions of the freeze-dried food and ileostomy contents were analysed in duplicate for their contents of Zn, Fe, Ca, Mg and P. The extruded product and the corresponding raw material were analysed separately. All glassware was washed in 2.5 M-hydrochloric acid and rinsed in deionized water before use. Zn and Fe were determined by atomic absorption spectrophotometry (Perkin Elmer Model 360), after dry-ashing in Pyrex beakers overnight (450°) of the freeze-dried ileostomy contents (0.3 g) or of the diet (0.6 g). Three drops of nitric acid (430 ml/l) were added and the ashing continued until a white residue was obtained. The ash was digested in 5 ml 5 M-HCl, the beakers covered with Parafilm and left overnight. The contents were then transferred quantitatively to 25-ml (ileostomy contents) or 100-ml (diet) flasks, diluted to volume and left for at least 4 h before analysis. Ca and Mg were determined by atomic absorption spectrophotometry after the wet-ashing (290–300°, 15 min) of 0.1 g freeze-dried samples in 1 ml concentrated sulphuric acid with the addition of 3 ml hydrogen peroxide (300 ml/l). If necessary, another 2 ml H₂O₂ were added and the tube heated again until the digest was clear and colourless. Deionized water was added to volume and the samples analysed after addition of lanthanum oxide. The same digest was used to

determine P according to Fiske & Subbarow (1925). Reference standards for Zn, Fe, Ca and Mg were prepared from Titrisol® (Mercks). Reference standard materials for Zn and Fe with concentrations representative of those found in the diet and ileostomy samples were run simultaneously and fell within the certified range (Orchard Leaves SRM 1571 and Bovine Liver SRM 1577(a); National Bureau of Standards, USA). Reference materials from our laboratory were used as controls for Ca, Mg and P analyses. The coefficients of variation for control materials of Zn, Fe, Ca, Mg and P were 3.1, 4.4, 4.1, 3.7 and 5.4% respectively.

Calculations and statistical methods

The apparent daily absorption was calculated as the difference between the intake and the amount found in the ileostomy contents.

For statistical comparison of the results from the two periods, Student's paired *t* test was used.

Ethical considerations

This project was approved by the Research Ethical Committee at Sahlgren's Hospital.

RESULTS

The daily intake of Zn, Fe, Ca, Mg and P from the low-fibre diet with the extruded product and the bran-gluten-starch mixture as well as the excretion in the ileostomy contents are shown in Table 2. The individual intakes were similar in the two periods. The extruded product and the bran-gluten-starch mixture contributed about 35% of the total intake of Fe and Mg, 15% of Zn and P and only 2% of the intake of Ca. The day-to-day variation in ileal mineral excretion in each 4 d period was small, the coefficient of variation being less than 15% in fifty-eight of the seventy 'balances'. The apparent daily absorption of Zn, Fe, Ca, Mg and P for each subject is shown in Table 3. Significantly lower apparent absorption ($P < 0.05$) from the extruded product than from the bran-gluten-starch mixture was found for Zn, Mg and P but not for Fe and Ca. The same differences were found for the mineral ileal excretions.

DISCUSSION

Earlier balance studies from our laboratory (Isaksson & Sjögren, 1967) have shown reliable mineral balances not to be reached until after 1–2 months on a constant diet in normal subjects. We have recently reported that undigested food components are completely excreted in the ileostomy contents on the day of consumption (Sandberg *et al.* 1981, 1983). Moreover, on a constant mineral intake, ileal excretion shows much less day-to-day variation compared with mineral faecal excretion in subjects with an intact colon (Isaksson & Sjögren, 1967). Therefore it is possible to obtain reliable results from short-term balance studies on subjects with an ileostomy (Sandberg *et al.* 1982).

Our present study shows the absorption from the small bowel only, where the main uptake takes place. Mineral absorption from the human colon remains to be established, but considerable uptakes from the colon of pigs are reported (Partridge, 1978).

While the intake of dietary fibre and phytic acid (1.5 mmol/d) was the same during the two periods of the present study, the digestion of phytic acid differed significantly (Sandberg *et al.* 1986). During the period with the bran-gluten-starch mixture, 44% of the phytic acid was recovered in the ileostomy contents, while the corresponding value for the period with the extruded product was 72%. It is therefore probable that the observed effect in the present study on mineral absorption of the extrusion-cooked product was mainly due to the presence of undigested phytic acid. Phytate-mineral complexes unavailable for absorption

Table 2. Daily intake and ileostomy output of minerals in seven subjects on a low-fibre diet with addition of 54 g bran-gluten-starch mixture (period A) or the corresponding extruded product (period B)

	Zinc		Iron		Calcium		Magnesium		Phosphorus	
	($\mu\text{mol}/24\text{ h}$)		($\mu\text{mol}/24\text{ h}$)		(mmol/24 h)		(mmol/24 h)		(mmol/24 h)	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Intake:										
Period A: Low-fibre diet	153	14	95	10	31.9	4.4	7.2	0.6	47.9	3.7
Bran-gluten-starch mixture	24	—	50	—	0.7	—	3.8	—	9.4	—
Period B: Low-fibre diet	153	14	96	10	29.1	3.3	7.2	0.6	47.6	3.7
Extruded product	24	—	50	—	0.7	—	3.8	—	9.4	—
Output:										
A	146	15	120	10	23.5	2.7	8.1	0.5	20.8	2.3
B	159	15	124	10	25.7	3.3	9.0	0.6	24.6	2.4

(Mean values with their standard errors)

Table 3. The apparent daily absorption of minerals in seven ileostomy subjects on a low-fibre diet with addition of 54 g bran-gluten-starch mixture (period A) or the corresponding extruded product (period B)

Subject no.	Zinc		Iron		Calcium		Magnesium		Phosphorus	
	($\mu\text{mol}/24\text{ h}$)		($\mu\text{mol}/24\text{ h}$)		(mmol/24 h)		(mmol/24 h)		(mmol/24 h)	
	A	B	A	B	A	B	A	B	A	B
1	48.4	58.2	34.2	32.5	8.8	4.0	4.5	2.8	46.9	44.0
2	10.2	-5.5	38.6	66.0	1.7	0.8	0.4	-0.2	28.9	25.8
3	14.4	7.8	10.3	11.9	3.0	3.9	2.8	2.4	28.5	27.7
4	50.5	40.5	0.6	0.8	9.8	8.5	4.2	3.7	40.6	37.5
5	4.4	-11.2	8.8	-8.1	8.3	0.1	1.5	1.0	34.7	27.8
6	70.2	53.1	53.9	37.6	6.9	4.2	5.0	3.6	40.3	36.5
7	15.1	-16.4	36.5	11.7	25.3	7.1	2.0	0.6	35.2	27.3
Mean	30.5	18.1	26.1	21.8	9.1	4.1	2.9	2.0	36.4	32.4
SE	9.6	12.0	7.4	9.6	2.9	1.1	0.6	0.6	2.5	2.6
Statistical significance of difference	$P < 0.05$		NS		NS		$P < 0.01$		$P < 0.01$	

NS, not significant.

may be formed during extrusion cooking or in the small bowel. A contributing factor would be that extrusion cooking as performed in the present study deactivated the phytase naturally present in bran.

The difference in P absorption after extrusion cooking can be ascribed to the difference in digestibility of phytic acid. The formation of a phytate complex with Zn and Mg might explain the difference in uptake for these minerals.

The impairment of mineral absorption after extrusion cooking was small and probably of minor nutritional significance when extruded products constitute a small part of the total diet. However, if extruded products were more commonly used and when total diets are extruded as, for example, in infant formulas, the effect could be significant. Studies of reasons why phytic acid is not broken down in the small bowel after extrusion cooking are therefore of interest. With modern technology it should be possible to use processes and conditions during extrusion cooking that render all nutrients available in fibre-rich products.

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