THE INFLUENCE OF THE PROCEDURE OF PRODUCING ON THE QUALITY OF THE FORTIFICATED PRODUCTS

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Abstract: *Mineral intake and bioavailability are critical factors for meeting mineral nutritional needs. The present study shows the correlation between the bread making procedures and the iron bioavailability in the fortified white bread. It was elucidated the most appropriate procedure, iron additive amount, and also the most effective recipe considering the organoleptic factors and the iron bioavailability index.*

Key words : *bread making procedure, iron bioavailability, baker's yeast*

1. Introduction

Iron deficiency is a major health problem in the world today [1]. Iron anemia is the severe form of iron deficiency. It can result in low resistance to infection, impaired psychomotor development, and cognitive function at children, poor academic performance, as well as fatigue and poor physical/work endurance.

Minerals present severe problems since the amount absorbed depends not only on the chemical form of the mineral in the food but on other ingredients in that food and of the rest of the diet and also on physiological factors. Phytates inhibit the absorption of non-heme iron. Although cereals contain reasonable levels of trace elements, their degree of absorption has been shown to be low. This may be due to the presence of both phytate and fiber in cereals. In several countries this has been considered with regard to iron and cereals are therefore often fortificated with iron. Wheat flour bread is an appealing vehicle for iron fortification because of its popularity, relative low cost, and widespread consumption [2].

Mineral intake and bioavailability are critical factors for meeting mineral nutritional needs. Whole grain bread contains high levels of potassium, magnesium, iron and zinc, but the presence of phytic acid compromises mineral and trace element absorption in humans. However, during bread making the content of phytic acid decreases due to the action of phytases in the dough. The activity of baker's yeast seems to have no significant effect on these conditions.Nevertheless, if very little phytate is hydrolyzed in unleavened whole meal breads including breads containing bicarbonate of soda, phytic acid hydrolysis occurs during all stages of yeast bread making. Reduction of phytic acid content in different bread types varies between 13% and 100%, with the lowest decrease being in unleavened breads. Phytic acid content of rye bread may be, under optimal conditions, reduced to near-zero values. The substantial decrease of phytic acid in whole wheat products can improve mineral availability in humans.

Considering the exposed problems concerning food fortification and iron bioavailability, the objective of this study was to investigate the influence of the technological procedure of the fortified product on the potential bioavailability of iron. Different procedures of bread making and different ingredients additions were applied to clear up the most appropriate recipe and technological method for the iron fortified bread.

2. Experimental

Preparation ot bread samples. For the bread preparation was used high quality flour, as ferrous additive was taken the ferrous sulfate. As bread making procedures were used the traditional biphase method (yeasts fermentation) and the lactic acid fermentation performed in two stages : the simple mixture of ingredients and further lactic fermentation (I lactic acid method A-L I) (Lopez *et al.*, 2001), and the dough scalding method (II lactic acid method A-L II). The lactic acid fermentation procedure was performend using different durations of fermentation (1, 2, 3 days). Also there was tested yeasts addition in different concentrations (25-75% of the traditional dose). The bread samples were dried until a constant mass, crumbled and portioned (by 10,0±0,2g).

Reagents and Chemical analysis. In a 1-l volumetric flask, 50 g hydroxylamine monohydrochloride was first dissolved in water. Concentrated HCl (100 ml) and 100 g trichloroacetic acid were added and the solution was brought to volume with water.

Bathophephenanthroline disulfonic acid (BPDS; 300 mg) was dissolved in 10-15 ml of water in a 1-l volumetric flask. The solution was brought to volume with 3 M sodium acetate. The solution was stored in an amber colored glass bottle to minimize lightcatalyzed deterioration. Iron standards were perorated, in duplicate, by diluting a stock iron solution (1000 μg Fe/ml in 1% HCl) with iron extracting solution to achieve the following concentrations of iron: 0, 1, 2, 3, 4, 5, 8 and 10 μg/ml. A 1-ml aliquot of the iron standard or the sample filtrate was mixed with 3 ml of the chromogen solution. After standing at room temperature for 15 min, absorbance was measured spectrophotometrically ar 535 nm [3].A standard curve was constructed with the iron standards using linear regression and the food sample readings were plugged into the regression equation to determine their iron concentrations. A filtrate blank was also prepared for each food sample by adding 3 ml of 3 M sodium acetate without the chromogen reagent (BPDS) to 1 ml of the food sample filtrate. The reading obtained from the blank was then subtracted from the reading obtained from the corresponding sample filtrate containing the chromogen reagent.

Phytic acid content in the extract was determined colorimetrically [4].

Estimation of absorbable iron. Absorbable iron was estimated using the model of Miller et al. [5]. After statistical comparison of the standard deviations, a multiple range test using the Student-Newman-Keuls method was performed to compare the means.

3. Results and discussions

The main problem in iron fortification remains its bioavailability, which, in the case of cereal products, depends mostly on the phytates content and their evolution during bread making, also their digestion in the gastro-intestinal way. In accordance with the bibliographical data, the lactic acid fermentation has a positive impact on the phytates enzymolisis which are progressively degradated into 5,4,3,2 and monoinozitol, the last three being able to pass the gasto-intestinal barrier. The present study shows the influence of different procedures of bread making, namely traditional and lactic acid fermentations with varied fermentation duration and also varied additional amount of bakery yeasts. The main purpose was to investigate the iron bioavailability from the test bread and the phytates enzymolisis, and their interdependence in the fortified product digestion.

The main results of this study are that bread fermentation improves mineral bioavailability in wheat products and that the natural leavening process is more efficient than yeast fermentation to improve iron bioavailability. Losses of phytic acid differ during the manufacturing process. The phytic acid egradation and lactic acid formation improves iron bioavailability from the sourdough bread. In parallel, in the present study, bread making with baker’s yeast contributed to phytic acid degradation. Although a phytic acid decrease was detected during bread making and assigned to yeast phytase activity, the main degradation of phytic acid seemed to be a result of endogenous plant phytases in the flour [6].

The fermentation duration and the additional yeasts amount influence appreciably the iron bioavailability. The best results were established at 2 days of lactic acid fermentation and 25% of baker’s yeast addition.

In the unfortified bread manufactured by the traditional methods the iron solubilization degree is 8-12%, and the soluble iron amount after 4h of *in vitro* digestion represents only 0,2-0,3mg Fe at 100g product. In the case of the fortified bread (4 and 8 mg% Fe), manufactured by the mono and biphase methods, the amount of the soluble iron constitutes 0,9 –06 mg% which represents 24 - 36% from the administrated iron amount. In the bakery products manufactured by lactic acid fermentation I the amount of soluble iron at the end of 4h of gastro-intestinal digestion constitutes 1,5-17mg%.

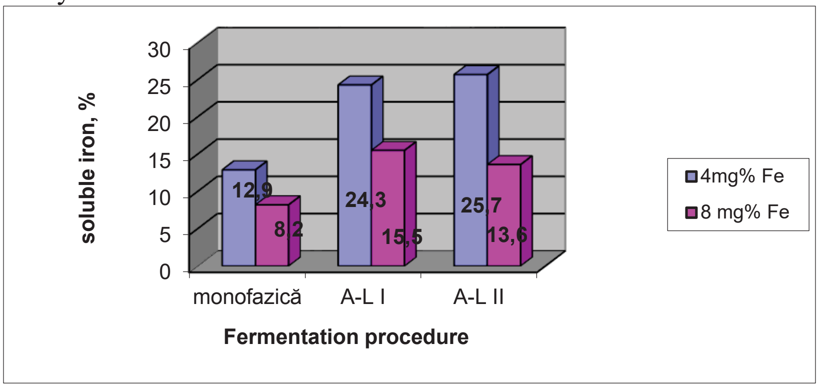
Table 1. The influence of the technologycal parameters of the bread making procedure on the *in vitro iron* bioavailability

Fe total bread=2,5 mg% total amount of phytates –578mg phytic acid/100g product

Bread making procedur	Fermentation period (days)	Yeasts addition %	Fe,mg% (sol)			Fe_{SOl} %	Phytates,mg%(sol)			Soluble phytates %
			Gastric	Tripsic	Total		Gastric	Tripsic	Total.	
Trad.	-	100	0,23	-	0,23	9,2	30,8	53,1	83,9	14,5
A-L I	1	-	0,29	0,25	0,54	21,6	121,9	83,7	205,6	35,37
A-L I	1	25	0,32	0,24	0,56	22,4	129,6	81,5	211,1	35,89
A-L I	1	50	0,26	0,08	0,34	13,6	123,5	78,6	202,1	34,36
A-L I	1	75	0,22	0,24	0,46	18,4	125,2	89,5	214,7	36,5
A-L I	2	-	0,19	0,32	0,41	16,4	112,6	87,5	300,1	51,02
A-L I	2	25	0,51	0,5	1,01	40,4	197,9	99,7	297,6	50,59
A-L I	2	50	0,29	0,28	0,57	22,8	111,4	75,3	186,7	31,74
A-L I	2	75	0,08	0,02	0,1	4,0	129,3	85,6	214,9	36,56
A-L I	3	-	0,42	0,18	0,6	24,0	204,1	22,6	226,7	39,2
A-L I	3	25	0,52	0,12	0,64	25,6	97,5	87,6	185,1	31,47
A-L I	3	50	0,29	0,18	0,47	18,9	98,7	112,7	211,4	35,94
A-L I	3	75	0,29	0,24	0,53	21,2	217,4	29,1	246,6	42,7
A-L II	1	-	0,28	0,23	0,51	20,2	114,4	15,0	129,4	22,4
A-L II	1	25	0,40	0,36	0,76	30,4	131,5	75,8	207,3	35,24
A-L II	1	50	0,35	0,23	0,58	23,2	182,5	47,6	230,1	39,8
A-L II	1	75	0,34	0,25	0,59	23,6	122,6	46,2	168,8	24,2
A-L II	2	-	0,27	0,12	0,39	15,6	108,7	75,8	184,5	31,37
A-L II	2	25	0,19	0,13	0,32	12,8	121,5	73,8	197,3	33,54
A-L II	2	50	0,22	0,15	0,37	14,8	119,6	109,7	229,3	38,98
A-L II	2	75	0,26	0,22	0,48	15,2	165,1	32,8	197,9	34,3
A-L II	3	-	0,14	0,17	0,31	12,2	95,6	112,7	208,3	35,41
A-L II	3	50	0,29	0,16	0,45	12,4	122,9	43,5	166,4	28,8
A-L II	3	75	0,22	0,26	048	15,2	82,7	136,5	219,2	37,26

The additional amount of baker's yeast (50-75%) has no considerable influence. The most significantly on the iron bioavailability influences the additive amount. Previous studies concerning the fortified bread manufactured by traditional method (monophase), show that the most propitious amount of iron additive is 4 mg%, that is 6,5 mg% as total iron amount in bread.

a) 50% baker's yeast



b) 75% baker's yeast

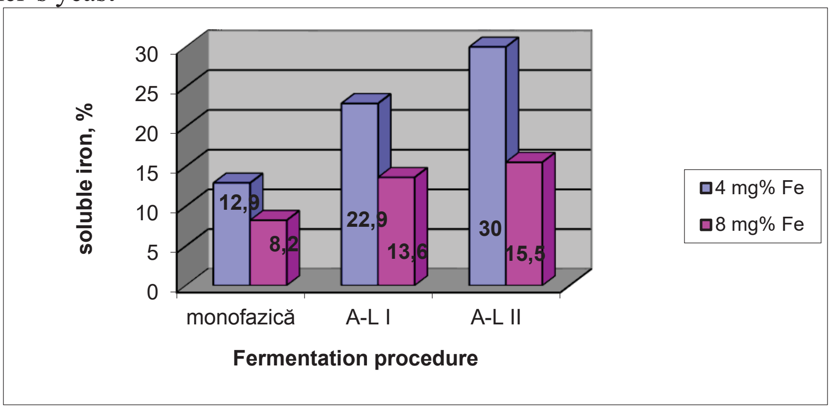


Fig 1. Influence of the manufacturing procedure and of the additive amount on the iron bioavailability.

Thus, iron bioavailability in the 3 different bread making procedures at an additive amount of 4mg% is higher comparing to the additive amount of 8mg%). Comparing the obtained results for the lactic acid fermentation I and lactic acid fermentation II, shows that in the second case iron bioavailability is a little higher, especially for 4 mg% Fe additive.

4. Conclusions

The present study concludes the most propitious conditions for the iron fortification of bread:

- The iron additive amount - 4 mg%
- The addition of 50% of baker's yeast for the lactic acid fermentation I or 75% of baker's yeast for the lactic acid fermentation II
- The lactic acid fermentation II ensures a higher phytates solubilization degree and a higher bioavailability of the non-hemic iron.

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