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Mineral bioaccessibility in French breads fortified with different forms iron and its effects on rheological and technological parameters

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ABSTRACT

The present study investigated the effect of different forms of Fe used in the fortification of wheat flour on the rheological characteristics of flour and technological quality of French breads and the bioaccessibility of added Fe, Zn and Ca naturally occurring by *in vitro* assay. The results demonstrated that the wheat flour was suitable for use in bakery products; however, the farinograph and extensograph parameters were affected by different forms of Fe, which also changed the technological quality of breads, with no negative impacts on bread making. The NaFeEDTA and SF microencapsulated proved to be the most effective's forms of iron, due to its higher dialyzed Fe content. Zinc bioaccessibility is not high, thus the NaFeEDTA contributed positively for the absorption of this mineral. In contrast, a high bioaccessibility of calcium was observed, which was not affected by the majority forms of iron. Additionally, Fe, Ca and Zn naturally occurring also presented high bioacessibility. Thus, French bread made with flour fortified with iron can contribute to demand and supply of minerals, and the form of Fe used can affect not only Fe bioaccessibility, but also Zn, leading to changes in the rheological properties of wheat flour and technological quality.

1. Introduction

In Brazil, as in many other countries, fortification of wheat flour and corn flour with iron has been carried out since 2002, to reduce iron deficiency anemia, considered a public health problem due to consumption of a habitual diet low in bioavailable iron (Brasil, 2002; WHO, 2006). Furthermore, the intake of essential minerals such as zinc and calcium is also important, as these minerals are related to the functioning of the body and the deficiency of these can lead to several physiological-pathological conditions of a serious nature (Akhter et al., 2012). Studies on the absorption of minerals have shown that an element can interfere with the absorption of other, competing for the same active sites (Perales et al., 2006). The addition of iron to foods is authorized by the Brazilian legislation, including wheat flour, and the compounds include dehydrated ferrous sulphate (dried), ferrous fumarate, reduced iron, electrolytic iron, NaFeEDTA, ferrous glycinate chelate, among others (Brasil, 2002). Other forms of iron such as ferric pyrophosphate (Kiskini et al., 2007) and microencapsulated ferrous sulfate (Nabeshima et al., 2005) have also been used as iron source for food fortification in other countries.

Some parameters should be considered in fortification with iron for not compromising the quality of the final product as cost (depending on the technology used), rheological properties, and the absorption in the human body (Akhtar et al., 2011; WHO, 2006). The rheological properties of dough, among other parameters, indicate the quality of flour used for bread making, and changes in these properties should not occur after fortification (Akhtar et al., 2011). However, Nabeshima et al. (2005) studied the technological of pan bread fortified with different forms of iron and found that the farinograph and extensograph parameters were affected by microencapsulated ferrous sulfate used in wheat flour fortification. Akhtar et al. (2009) investigated the effects of whole wheat flour fortification with iron on the rheological properties of dough during 60 days of storage. The authors observed greater water absorption in the fortified flour, and the rheological parameters of flour were less affected by NaFeEDTA when compared to elemental iron.

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Although flour fortification with Fe is a common practice in Brazil and other countries, there is no positive correlation with the reduction of anemia in the population (Hurrell et al., 2010). Various authors have studied the bioaccessibility of iron compounds and other minerals, aimed to find more effective compounds in the fortification process. It is noteworthy that some forms of iron such as ferrous sulfate, iron fumarate, NaFeEDTA, and microencapsulated ferrous sulfate are indicated for fortification of cereals, since they are better absorbed by human body (Akhtar et al., 2011). Some researchers have suggested the replacement of ferrous sulfate for its microencapsulated form, with the advantage of protecting food against undesirable organoleptic changes and lipid oxidation (Cocato et al., 2007). *In vitro* assays have been performed to estimate the bioaccessibility of these compounds (Argyri et al., 2009; Cámara et al., 2005; Kiskini et al., 2007; Perales et al., 2006) in addition to studies on Caco-2 cells (Perales et al., 2006).

A comprehensive study on the effect of the addition of different forms of iron to wheat flour on the rheological properties of dough and quality parameters of French bread, and the bioaccessibility of iron, zinc, and calcium have not been found in literature. Therefore, the present study aimed to investigate the effect of different forms of iron on both the rheological properties of fortified flour and technological quality of breads. In addition, the bioaccessibility of different forms of iron used in fortification, zinc and calcium naturally occurring contents in soluble and dialyzed fractions were also investigated.

2. Material and methods

2.1. Material

Refined wheat flour, 100 kg, (Moinho Guaçu Mirim, Mogi Guaçu, Brazil), was fractionated in portions of 10 kg, and fortified with 4.2 $mg \cdot 100\ g^{-1}$ (±20%), according to Normative Resolution 344 (Brasil, 2002), with different forms of iron, totaling 6 samples and the control (without iron addition). The iron sources were: iron sulfate monohydrate (SF), ferrous fumarate (FF), reduced iron (Fe R), (M Cassab, Brazil); sodium iron ethylenediaminetetraacetic acid (NaFeEDTA - Ferrazone) (Vogler Ingredientes, SP, Brazil); microencapsulated ferrous sulfate (SF micro) and microencapsulated ferrous fumarate (FF micro), (Functional Mikron, Valinhos, Brazil). For the fortification process, the sample was homogenized in V-blender (Tecnal Piracicaba, Brazil) for 30 min in portions of 5 kg. The resulting flours were characterized for composition, rheological properties and iron content.

Breads were made by the modified straight dough method. The dry ingredients (100% wheat flour, 1.5% instant dry yeast, 1.8% salt, and 2.5% enhancer) and water were mixed in HAE 10 dough mixer (Hyppolito, Ferraz de Vasconcelos, Brazil) at slow speed for 4 min, and at high speed until development of the gluten network. Dough was divided in portions of 65 ± 1 g, rounded and allowed to rest for 15 min. After this period, the portions were molded in modeling machine HM2 Hp 0.5 (Hyppolito, Vasconcellos Ferraz, Brazil) and fermented in fermentation chamber 20B (Super Freezer, Poços de Caldas, Brazil), for 60–70 min at 27–30 °C and relative humidity of 85–90%. A cut opening was made on the surface of the dough pieces, and then the samples were baked in a HF 4B oven (Haas, Curitiba, Brazil), with top and bottom (hearth) temperatures of 170 °C and 180 °C, respectively, for 16 ± 3 min. After 30 min of cooling, the quality of breads was evaluated. Ten units of each bread was ground, packed, and stored in the freezer (- 20 °C) for analysis.

2.2. Methods

2.2.1. Characterization of the wheat flour

The proximate composition of the control flour was determined as follows: moisture, protein, lipids and ash were determined according to the AACC (2010) methods 44–15.02, 46–13.01, 30–25.01, and 08–01.01, respectively. Total carbohydrates were calculated by difference. Total dietary fiber was determined by AOAC (2000) method 985.29. Gluten index (GI) and gluten content (wet and dry) were determined in Glutomatic equipment (Perten Instruments, Hagersten, Sweden) by AACC (2010) method 38–12.02. Iron contents were determined as reported by Rebellato et al. (2015). All analyses were performed in triplicate.

The wheat flour fortified with different forms of iron, and the control sample were subjected to farinograph analysis, and water absorption (WA), arrival time (AT), dough development time (DDT), stability (EST) and mixing tolerance index (MTI), according to the AACC (2010) method 54–21.01, using farinograph Brabender (Duisburg, Germany), model 827505. The extensograph properties were evaluated for resistance to extension (R), maximum resistance to extension (Rm), extensibility (E) and a ratio number (D = R/E), in the Brabender equipment (Duisburg, Germany), model 860703, according to the AACC (2010) method 54–10.01, in triplicate.

2.2.2. Technological quality of breads

French bread samples (control and fortified) were evaluated for the following parameters: specific volume (VE), 10–05.01 by the AACC (2010); shape, assessed by width and height measurements of the loaves, according to Bodroza-Solarov et al. (2008); oven spring, according to Shittu et al. (2008); cut opening and cut height, according to Almeida and Chang (2012), in four replicates.

2.2.3. Bioaccessibility and minerals contents in breads

Fe, Zn, and Ca were determined in French breads by Flame Atomic Absorption Spectrometry (FAAS), according to Rebellato et al. (2015), in triplicate. For Zn and Ca determinations, the method was validated based on the parameters precision, recovery, sensitivity, linearity, limit of detection, and limit of quantification. In the calcium determination an amount of lanthanum oxide sufficient to obtain a final content of 0.5% was added to eliminate phosphate interferences.

Precision was accepted and expressed as average coefficient of variation of ±10%. Recovery was performed by adding the analytes (Zn and Ca) to the bread samples (without fortification), at 50 and 100%. Percent recovery close to 100% was considered satisfactory. Sensitivity was expressed by the slope of the linear regression equation. Linearity was assessed by the correlation coefficient of the calibration curve and the residual plot was considered satisfactory for $r \geq 0.995$. The limits of detection and quantification were calculated based on the calibration curve, and expressed as $mg \cdot 100\ g^{-1}$ sample.

The control and the sample containing different forms of iron were also subjected to solubility tests according to Cámara et al. (2005), with modifications as described by Rebellato et al. (2015). The volume of enzyme solution used in the digestive trial was adjusted to 6.0 mL (0.4 g of pancreatin from porcine pancreas P-7545, and 2.5 g of bile from B-8631bovine and ovine, Sigma Chemical Co., St. Louis, USA, in 100 mL $NaHCO_3$ 0.1 mol L^{-1}).

The dialysis test was performed according to Perales et al. (2006), with modifications as described by Rebellato et al. (2015). For the digestive trial, 30 mL of PIPES buffer, 0.30 mol L^{-1} (piperazine-N, N0-bis [2-ethanesulfonic acid] disodium salt), adjusted to pH 6.3 with concentrated HCl was used in membrane dialysis (dialysis bag: molecular weight cut-off from 12,000 to 16,000 and porosity of 25 Å, Inlab, São Paulo, Brazil) rather than $NaHCO_3$