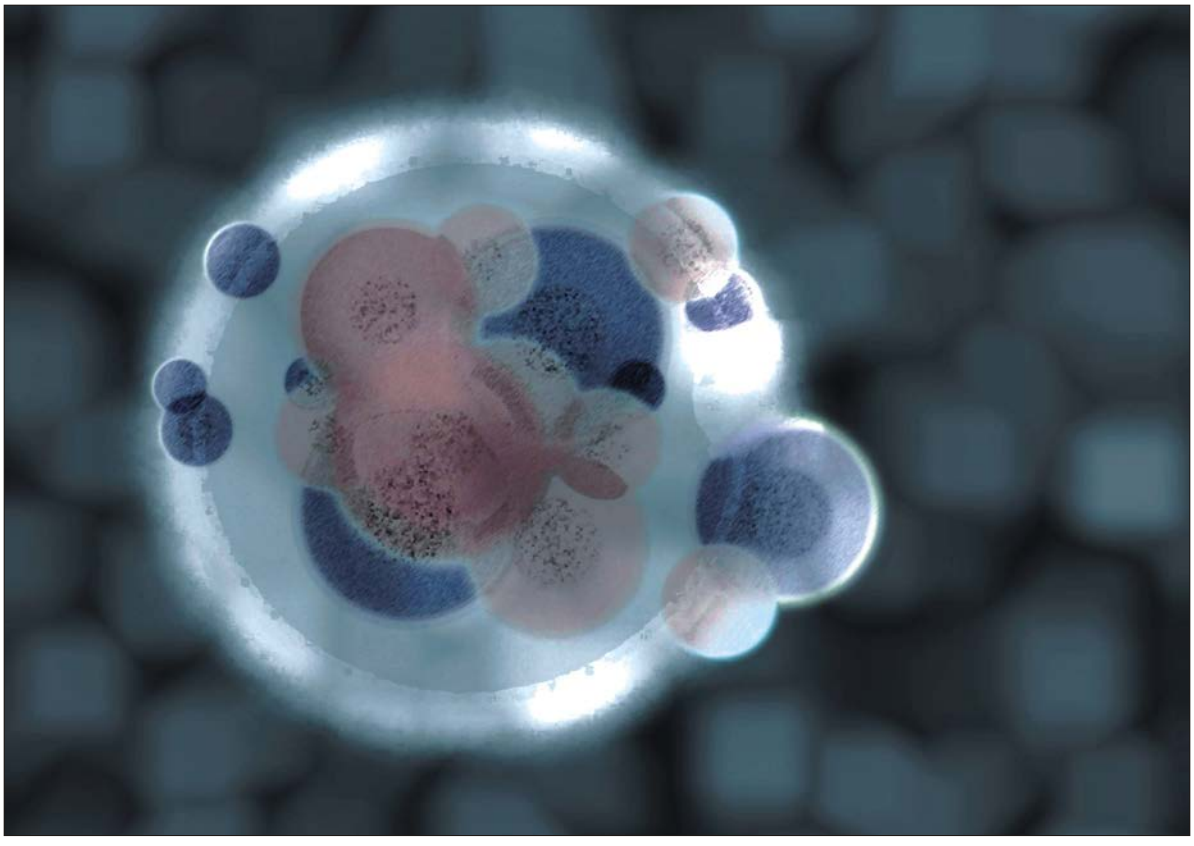

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Comparative Spectrophotometric and Atomic Absorption Determination of Iron Content in Wheat Flour

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Abstract: A simple and sensitive spectrophotometric method has been used for the determination of iron concentration in wheat flour and compare with atomic absorption method. The method is based on the reaction of iron with o-phenathroline reagent at acidic condition. The absorbance-concentration plot is linear over the range 0.3-2 mg mL^{-1} with correlation coefficient of 0.995. This method was applied successfully for determination of iron in flour samples. Quantification of the same flour was determined by atomic absorption for iron content. Values of iron content measured in the flour samples with spectrophotometer method are in good agreement with results obtained with atomic absorption method. It was not statistically significant difference between visible spectrophotometric and atomic absorption methods. This method can be successfully applied determination of iron in flour samples and can use instead atomic absorption.

Key words: Iron, flour, phenanthroline, atomic absorption, spectrophotometry

INTRODUCTION

Iron deficiency is causing serious damage to social and economic development through poorer pregnancy outcomes, impaired cognition especially in young children, reduced work capacity and increased morbidity from infection disease (McPhail and Bothwell, 1992). The principal cause in children and women of reproductive age in developing countries is inadequate intake of usable iron, which is normally found in a well-balanced diet in the form of heme and nonheme iron. Fortification involves adding nutrients of foods to maintain or improve the quality of diet. The fortification of foods has been shown to be a good and adequate preventive measure for the control of iron-deficiency anemia on a long-term basis (Cook and Reusser, 1993; Yip, 1997; Beinner and Lamoumier, 2003). Flour fortification with iron is an attractive optimum for controlling iron-deficiency anemia in countries such as Iran.

Analyzing the iron content of flour determines whether the required level of iron is present in fortified flour, which is critical to success of flour fortification program. Several analytical methods are available for determining the level of iron in flour ranging from simple tests to ones that required sophisticated instruments. The methods for determination of iron are spectrophotometric and atomic absorption method (AA) (Riganakos and Veltsistas, 2003). The atomic absorption method was used to determine elements in foods. It is sensitive, reliable, accurate and precise technique but this method has some limits such as; required skilled personal, time-consuming, high capital and maintenance costs (Strong and Martin, 1990; Rao, 2005; Davidson and Miller, 2005). The use a rapid, safe, simple and inexpensive method for determining the iron content of flour is perfect. The goal of this study was to compare spectrophotometric and atomic methods for determination of iron content in white wheat flour.

MATERIALS AND METHODS

Materials and solution: Chemicals were obtained from sigma chemicals and Merck Company. Water used in preparation was doubled deionized.

Orthophenanthroline solution was prepared by dissolving 0.1 g o-phenanthroline in about 80 mL water 80°C, cool and dilute to 100 mL. For iron standard solution, 10 μg Fe mL^{-1} was prepared by dissolving 0.1 g analytical grade Fe wire in 20 mL HCl and water and dilute to 1 L. Dilute 100 mL of this solution to 1 L. In a 100 mL volumetric flask, 10 g hydroxylamine monohyrochloride was dissolved in water. Iron working standard solution were prepared by diluting a stock iron solution to achieve

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the following concentration of iron: 0.3, 0.6, 0.9, 1.2, 1.5, 1.8 and 2 mg mL^{-1} for spectrophotometric method and 0.2, 0.4, 0.6, 0.8 and 1 mg L^{-1} for atomic absorption.

Apparatus: Spectrophotometric (PharmacyaBiotec, England) was used for conventional measurements. An atomic absorption spectrophotometr (BRAIC-V FX-130, China) with iron Hallow cathod lamp at wavelength 248.3 nm was used.

A total of five samples of wheat flour silo prepared from Mazandaran province in Iran.

Determination: Spectrophotometric method: Samples of flour (2 g) placed into clean crucible. Ash in muffle furnace at ≤550°C. The crucible remove from furnace and cool to room temperature. One milliliter concentrated HCl add, rinse upper portion of crucible; evaporate to dryness on steam bath. Dissolve residue adding 5 mL concentrated HCl and cover with watch glass and rinse with water, filter quantitatively in to 100 mL volumetric flask and dilute to volume pipet 10 mL aliquate into volumetric flask and add 1 mL hydroxylamine HCl solution. After 5 min, add 5 mL buffer solution and 1 mL o-phenanthroline, dilute to volume and mix thoroughly. After standing 30 min, then measure absorbance of samples, standards and blank solution in spectrophotometric at 510 nm.

Atomic absorption: Two gram of samples place into crucible and heated ≤550°C at 12 h, add 20 mL 2 N HCl dilute to 100 volume. Measure absorption of solution directly with atomic absorption spectrophotometer. The AOAC and AACC official spectrophotometric and atomic absorption methods were used for determination of iron in flour (Davidson and Miller, 2005).

Statistical analysis: The statistical significance between iron content in the samples by two used methods was evaluated with a Mann-Whitney U-test. p-values less than 0.05 was considered to be statistically significant.

RESULTS AND DISCUSSION

Iron content of wheat flour samples was determined by spectrophotometric method and compared with atomic absorption method. The mean iron concentrations determined visually were plotted against standard concentrations of iron spectrophotometrically and linear regression was performed (Fig. 1). The linear regression equation was y = 0.0765x+0.0057 with a correlation coefficient (R^2) 0.9947 (n = 7) indicating a good linearity. Quantification of the same wheat flour samples that assayed with spectrophotometric method was determined by atomic absorption. The linear regression equation was

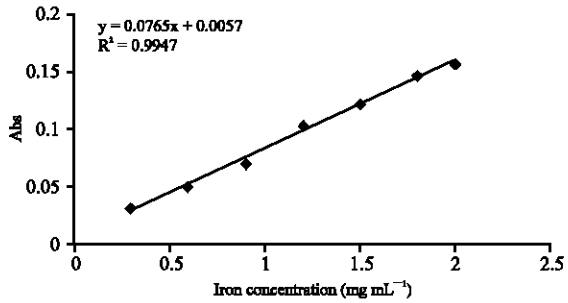

Fig. 1: Correlation of iron concentration (x) vs absorbance (y), determined by the UV/Visible spectrophotometric method at 510 nm

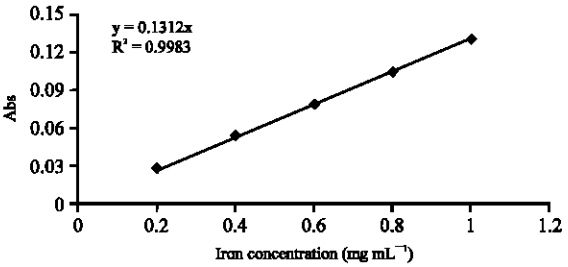

Fig. 2: Correlation of iron concentration (x) vs absorbance (y), determined by the atomic absorption method

Table 1: Comparison of iron concentration in flour samples determined by spectrophotometric and atomic absorption methods

	Iron concentration (mg kg^{-1})[a]	
Flour samples	Spectrophotometric method[b]	Atomic absorption
1	21.71±0.09	22.87±0.28
2	35.93±0.15	34.78±0.13
3	46.92±0.01	44.97±0.56
4	28.70±0.06	29.09±0.56
5	25.58±0.02	26.62±0.23

[a]Values are means±standard deviations (n = 3) [b]p>0.05 between two methods for all values

y = 0.1312x with R^2 0.998 for atomic absorption (Fig. 2). The mean iron content of flour samples was showed in Table 1. The simple method (spectrophotometric reading) produced accurate results that were not significantly different from the atomic absorption method for the flour samples tested.

In recent years, awareness that trace elements play a very important role, either beneficial or harmful, in human health has increased. Because most essential trace metals especially iron are present in food products in very low concentration, precise and accurate analysis is most essential if meamingful results are to be obtained (Rao, 2005). A method of determining iron in food is flame atomic absorption spectrophotometry with slurry nebulization into an air-acetylene to various flame has been developed. This method has been applied to various

kinds of trace elements in foods (Rao, 2005). This method required skills personal and expensive maintenance. This study showed that spectrophotometric method can used to determine iron content in flour and this method has not significantly difference with atomic absorption. The o-phenanthroline was used a colour reagent. The spectrophotometric method is based on a complex formation between iron and phenanthroline to give an orange-red product showing maximum absorbance at 510 nm (Tesfaldet and Staden, 2004). The colour is developed 30 min. This method is inexpensive and safely compared to atomic absorption. The results (Table 1) showed that distribution of iron content between samples flour are different, it was between 22.8 to 44.97 ppm (mg kg^{-1}). It is necessary to determine iron level in each sample if the fortified food with iron is one of goal of developed country. Before add iron to food it is important to determine iron level. The actual availability of iron in food may depend on their condition such as genetic and physiologic of food and soil (Rao, 2005). The simple method (spectrophotometric method) is practical, perfect and not very skilled personal for determination of iron in flour for all laboratories that has simple equipment such as spectrophotometer. Values of iron content measured in the flour samples with spectrophotometer method are in good agreement with the results obtained with atomic absorption method.

In conclusion, the spectrophotometric method with phenanthroline reagent can be successfully applied to the determination of iron in flour and it can use instead atomic absorption.

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