EFFECTS OF FeSO₄- Na₂EDTA MIX, FERROUS FUMARATE Na₂EDTA MIX AND THE RINSING CONDITION ON THE FERROUS FORTIFICATION IN PARBOILED RICE

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ABSTRACT

The objective of this study were to comparison the adsorption of Fe fortified in parboiled rice from FeSO₄ and Ferrous fumarate, both in the presence of Na₂EDTA and to evaluate the retention of iron under the different rinsing conditions of parboiled rice. The sensory evaluation of Fe – fortified parboiled rice was also studied.

There was no significant difference in the concentration of iron in parboiled rice (IR64 cultivar) between two approaches: fortifying Fe from FeSO₄ and from Ferrous fumarate, both in the presence of Na₂EDTA. The adsorption rates of Fe increased with the concentration of ferrous solution and soaking time. The loss of Fe after rinsing was quite low. Sensory evaluation of Fe fortified parboiled rice was at high consumer acceptance level..

Keywords: FeSO4- Na₂EDTA, ferrous fumarate - Na₂EDTA, Fe fortification, parboiled rice, Fe retention

1. INTRODUCTION

Iron-deficiency is the most serious health problem in Asia and Africa countries where the diets based on rice. Together with rice, parboiled rice is the stable food in South Asian countries and in Africa. However, the content of iron in raw rice and parboiled rice are not high enough for human body needs. Fortification of Fe into food has become a suitable approach to reduce the prevalence of Fe – deficiency anemia [8, 14, 15, 11].

Iron fortification of parboiled rice was a novel technique to increase Fe content in rice [11]. The parboiling conditions include soaking time, temperature were determined as the key factors, among other, affecting the density of Fe in Fe-fortified-parboiled rice [13]. The penetration and the retention of fortifying Fe in grain and parboiled grain various among Thai's cultivars [11].

FeSO₄, ferrous fumarate were used to fortify Fe in some food such as milk, bread, Indian prepared rice...The relatively poor adsorption of iron from food related to the nature of food [2]. The presence of Na₂EDTA enhance the intrinsic or added Fe adsorption from low iron bioactivity food when the molar ratio of EDTA to iron of \leq 1.00 [1], in comparison to the ferrous solution without EDTA used. Na₂EATA was also demonstrated a significantly enhancement of the iron adsorption from the high iron bioactivity food in comparison to the ferrous sulfate used alone. Another study showed that the bioactivity of iron fortified in rice was various among rice cultivars and enhanced in presence of acid ascorbic or EDTA [10, 12].

In Vietnam, researching on Fe fortification into rice has not received proper concentration although rice is our stable food, the iron deficiency anemia is still present and the export of rice from Vietnam into the world is in top-5. Among other, the cultivar IR64 is one of the most importance rice cultivar for exporting. The goal of this study was evaluating effect of the nature of the Fe solution (FeSO₄ - Na₂EDTA or C₄H₂FeO₄ -Na₂EDTA) and the soaking condition (Fe concentration in soaking solution and soaking time) on the content of Fe in parboiled rice (the cultivar IR64). The desired content of Fe in finished rice product was about 21.3 μg Fe/g rice. The fortification in whole grain (paddy rice) and in milled rice were also concerned. The retention of Fe after various times of rinsing was also checked.

2. MATERIALS AND METHODS

2.1. Fe fortification through parboiling process

Samples of unfortified paddy rice IR64 or unfortified milled rice were collected from Dong Nai Province. The Fe concentration of rice samples is 1.3 mg Fe per 100 g milled rice. For the Fe - fortification, 10 g of paddy rice or milled rice were soaked with 30 ml of various Fe solution (FeSO₄ - Na₂EDTA or $C_4H_2FeO_4$ -Na₂EDTA).

To study the adsorption of Fe into endosperm, two ferrous solutions were used in rice soaking: ferrous sulfate (FeSO₄) and disodium ethylenediaminetetra-acetic (Na₂EDTA) mixed in molar ratio of 1:1, ferrous fumarate ($C_4H_2FeO_4$) and ethylenediaminetetra-acetic acid disodium salt (Na₂EDTA) mixed in molar ratio of 1:1.

Preliminary studies showed that the soaking rice and paddy rice 60°C gave the highest moisture in rice and paddy rice and the grains still kept a good shape (results not shown). Therefore the soaking temperature chosen for this study was 60°C.

In study of the effect of Fe concentration and soaking time, the concentration of Fe in the suitable ferrous solution used was in range of $8.33~\mu g/mL$ to $66.67~\mu g/mL$. Grains were soaked at 60° C and in various period of time (in range of 15 min to 150 min for the milled rice, in range of 1 hour to 8 hours for paddy rice) and various concentration of Fe solutions; after draining off free water, were steamed at 100° C for 5 minutes. The treated grains were cooled and dried to the moisture content of approximately 13%, milled into rice and then analysed for Fe content. Each treatment was replicated twice.

2.2. Retention of Fe

Samples were rinsed to simulate rice washing behaviour when cooking parboiled rice. The milled samples of raw Fe-fortified parboiled rice were thoroughly rinsed in 1, 2, 3 times of clean water (2:1 v/w (water/rice) ratio) [5, 16] and then drained off water. Samples were cook for 30 minutes with water and then drain to simulate the cooking process for preparing cooked rice samples. The cooked rice samples were then analysed for Fe content.

2.3. Solubility of Fe

For dilute acid extraction, approximately 0.5 g of milled Fe-fortified-parboiled rice grains was weighed into a 50 ml flask. Each sample was treated with 10 ml of 0.1 M HCl solution and placed on a hotplate at 60°C for 30 minutes. After the extraction of soluble Fe, the residue was analysed for insoluble Fe by the method described in total Fe analysis. Soluble Fe in the fortified

Fe rice was calculated by the deduction from total Fe concentration of insoluble Fe concentration.

2.4. Analysis of Fe content

Analysis method of Fe absorbed in paddy rice was AOAC 986.15. The analysis was carry out at Hai Dang sorptography Center for a week.

2.5. Data analysis

The analysis of variance was carried out to detect the differences of Fe concentration in Fefortified paddy rice and milled rice using FeSO₄:Na₂EDTA and $C_4H_2FeO_4$:Na₂EDTA and in treated rice among different molar ratios of Ferrous solution. The significant difference among treatments was compared using the least significant difference (LSD) at p < 0.05.

3. RESULTS AND DISCUSSION

3.1. Comparison the effects of two ferrous compounds, Ferrous sulfate and ferrous fumarate (in the presence of Na2EDTA) on Fe fortification in parboiled rice

In this study, the soaking of rice (paddy rice or milled rice) were carried out with two solutions of ferrous: the mix of sulfate $FeSO_4$ and Na_2EDTA (in molar ratio 1:1) or the solution of $C_4H_2FeO_4$ in the presence of Na_2EDTA at the molar ratio of 1:1.

Firstly, the experiment was carried out with milled rice. The Fe concentration in soaking solution was 50 μg Fe/ml. The soaking time was 120 minutes. Table 1 shows the Fe concentration in Fe – fortified - rice after treating milled rice. There are no significant difference in Fe content in two samples of milled rice which were treated in the same condition but difference Fe compound (p < 0.05).

Table 1. Concentration of Fe in parboiled rice after Fe fortification carried in milled rice

Ferrous solution	Fe concentration (μg/g milled rice)
FeSO ₄ :Na ₂ EDTA (1:1)	28.40 ± 0.47^{a}
$C_4H_2FeO_4$: Na_2EDTA (1:1)	28.75 ± 0.39^{a}

^{*} Value are means \pm SD (n=2);

Table 2. Concentration of Fe in parboiled rice after Fe fortification carried in paddy rice

Ferrous solution	Fe concentration (µg/g milled rice)
FeSO ₄ :Na ₂ EDTA (1:1)	25.56 ± 0.10^{a}
C ₄ H ₂ FeO ₄ : Na ₂ EDTA (1 : 1)	25.73 ± 0.14^{a}

^{*}Value are means \pm SD (n = 2);

^{*} Means with no common letters differ, p < 0.05 (n = 2).

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Secondly, paddy rice was used to study the effect of ferrous solution nature. The experiment was carried out at the same condition of soaking for two ferrous solution nature. Table 2 shows that there are no significant difference in Fe content in two samples of paddy rice which were treated in the same condition but difference Fe compound (p < 0.05), in the presence of Na₂EDTA.

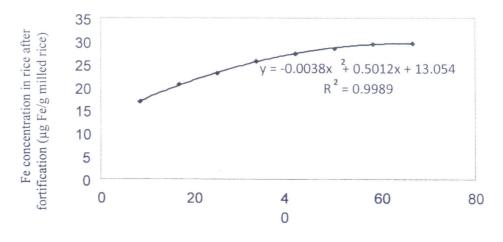
The role of Na_2EDTA in Fe fortification in meal has been studying [1, 6, 11]. It enhances the bioavailability of Fe in Fe-fortified cereal-based meal [6], in Babali bread and in low iron bioavailability meals [1]. It was expected to enhance the adsorption of Fe into rice. In current study, there was no significant difference in Fe concentration in treated rice between Fe sulfate and Fe fumarate treatment.

The slightly difference between the content of Fe in parboiled rice after treating paddy rice and milled rice may due to the presence of inhibitors in husk of paddy rice such as phytate,... However this difference was not concerned in the current study.

The mix of ferrous fumarate and Na₂EDTA was subjected to next experiments. The role of Na₂EDTA in the enhancement of Fe adsorption from poor soluble or insoluble ferrous compound such as ferrous fumarate has been in controversy [1, 6, 17, 9, 3]. However, using FeSO₄ for anemia treatment showed unpleasant side effects including the increase of plasma malondialdehyde [4, 7]. Therefore, the mix of ferrous fumarate and Na₂EDTA (molar ratio of Fe and EDTA of 1) was chosen for the next experiments.

3.2. Effect of Fe concentration in soaking solution on the concentration of Fe in fortification rice after treating milled rice

Firstly, milled rice was used tested to find out whether relation between the concentration of Fe in soaking solution (in the tested concentration) and the concentration of Fe in treated milled rice is present. Figure 1 and Figure 2 illustrated that the concentration of Fe in the soaking solution showed significant effect to the Fe content of treated rice. The concentration of iron in Fe fortified – parboiled – milled rice increased when the Fe concentration in soaking solution increased from $8.33~\mu g$ Fe/mL to $66.67~\mu g$ Fe/mL.

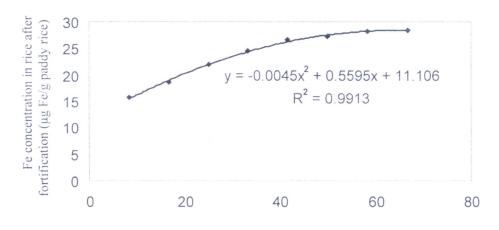


Ferrous concentration in soaking solution (µg Fe/mL)

Figure 1. Influence of Fe concentration in soaking solution on Fe content in Fe fortified-parboiled rice after treating milled rice

At the concentration in the range of 8.33 μg Fe/mL to 58.33 μg Fe/mL, the content of Fe in treated milled rice samples were marked difference. From the level of 58.33 μg Fe/mL, the detected Fe concentration had no significant change. The expected Fe concentration in Fe fortified rice (21.305 μg Fe/g rice) was obtained by using the iron concentration of soaking solution at 19.29 μg Fe/mL in case of milled rice and 22.19 μg Fe/mL in case of paddy rice which were in range of this experiment. The largest concentration of Fe in end - rice product (the Fe concentration of Fe solution was 66.672 μg/g rice) in this experiment was higher than expectation (29.54 μg/g rice in case of milled rice and 28.43 μg/g rice in case of paddy rice).

In the same period time of soaking, the concentration gradient affected the Fe loading rate. The high concentration gradient of Fe promote the Fe loading of rice due to the osmotic pressure gradient at the surface of endosperm. Increasing of Fe content in to soaking solution at Fe high concentration reduced the Fe loading rate due to less available of space in the endosperm. The same phenomenon occurred in cased of paddy rice and milled rice.



Ferrous concentration in soaking solution (µg Fe/mL)

Figure 2. Influence of Fe concentration in soaking solution on Fe content in Fe fortified-parboiled rice after treating paddy rice

3.3. Effect of soaking time on the concentration of Fe in fortification rice after treating rice

The influence of soaking time on the Fe loading of rice was studied on both of paddy rice and milled rice. The results (Figure 3 and Figure 4) showed that soaking time had a strong effect to the content of Fe in finished rice product. There was significant difference between soaking time of milled rice and paddy rice. The desired content of Fe in final rice product, as mentioned above, in case of milled rice was obtained after about 2 hours while the paddy rice needed about 8 hours to reach that Fe content. This difference in soaking time due to the barrier cause by husk and bran of paddy rice.

The Figures also illustrated relation between soaking time versus Fe loading rate of samples. The Fe loading rate was faster at the beginning of soaking. The longer soaking the slower increasing of Fe content in end-product.

To reach the content of Fe in final rice product at $21.305 \mu g$ Fe/g rice, it took 118.7 min and 7.25 hours at soaking step for milled rice and paddy rice respectively.

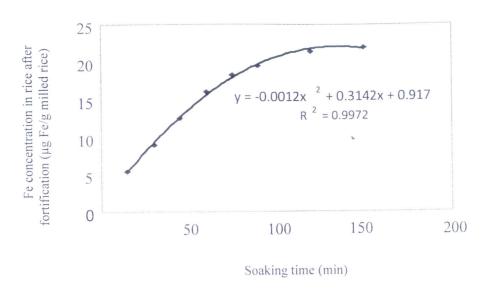


Figure 3. Influence of soaking time on Fe content in Fe fortified-parboiled rice (milled rice)

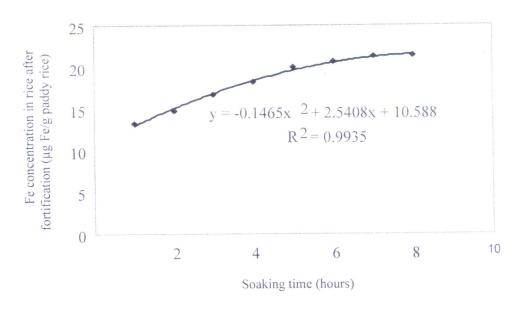


Figure 4. Influence soaking time on Fe content in Fe fortified-parboiled rice (paddy rice)

3.4. Effect of rinsing on retention of Fe in Fe - fortified - parboiled rice

The Fe fortified parboiled rice was tested for the retention after rinsing. The results showed that the increase the rinsing times of parboiled rice cause decrease in the amount of Fe in milled rice. The rinsing was carried out in 1, 2, 3 times resulted in Fe remaining in milled rice was 20.9 μ g Fe/g rice, 20.4 μ g Fe/g rice and 20 μ g Fe/g rice respectively in compare to the content of 21.305 μ g Fe/g rice before rinsing.

The Fe concentration in parboiled rice is easily extracted in dilute acid. Concentration Fe which was extracted was very high, it up to 85.51%.

The sensor analysis showed that iron parboiled in this study were accepted by consumers (data not showed).

3.5. Testing Fe concentration in parboiled rice after dilute acid extraction

The Fe in parboiled rice in the current study is easily extracted in dilute acid. Concentration Fe which was extracted was very high, it up to 85.51%.

4. CONCLUSIONS

This study has established that Fe fortification of paddy rice during the parboiling process increased the Fe concentrations in milled rice grain about 21.75 μ g/g rice compared to those in unfortified raw rice (13 μ g/g rice).

The mixes of FeSO₄ and Na₂EDTA or Fe fumarate and Na₂EDTA were not significantly influent the Fe loading of rice (in both of milled rice and paddy rice).

Increase concentration of solution also increase adsorption speed. At a certain concentration of solution (\geq 60 μg Fe/ml), the amount of iron in milled rice didn't rise more over and it had constant value. This is the limit of the adsorption ability of paddy rice. However, the value which was expected to had good amount of iron in milled rice was 22.3 μg Fe/ml.

It is found that if the soaking time is increasing, result in rising the amount of adsorbed ion. However the longer the time is, the lower the adsorption speed is and had constant value after certain period. It's about 6 hours. This was because there is no difference between iron concentration inside and outside paddy rice grain, thus there was not enough motion for iron to diffuse into paddy rice grain. The longer soaking time showed no significant change of Fe concentration in rice.

In other researches, it is found that a part of solute iron has been dissolved into water if milled rice grains were washed several times before cooking, known as the lost iron. The amount of lost iron was not high, it is from about 4% to 6% increase by increasing the washing times. This lost iron was very small compare to lost iron in un-parboiled rice (70%).

The previous studies found that there was a relation between levels of bioavailability Fe in the Fe-fortified parboiled rice grains and the high solubility of the fortified Fe retained in the grains. In the current research, about 85.51% of the fortified Fe remained soluble in dilute acid compare to 60% of the previous study [12].

The sensory test was carried out and was highly acceptable. Although sensor points of iron fortified parboiled rice is not as high as normal IR64 rice, but they was enough for the consumers accept it. So this method is high practicability and application.

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