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The determination of iron content in legumes for health and consumption purposes

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Abstract

There is a deficiency problem of Fe in many vegetarians. Because of this they tend to eat higher numbers of legumes, thought to be high in iron. Legumes of different varieties were ashed and the Fe content of each was examined via inductively coupled plasma atomic emission spectroscopy (ICP-AES) in order to come to conclusions of whether or not the Fe amount in average legumes is enough to complete the needs of vegetarians. Garbanzo beans, lentils, black beans, and black eyed peas were tested and it was found that lentils had the amount of Fe per serving furthest from the value of recommended daily intake while black beans had the closest amount per serving.

Introduction

In recent years, more and more people, especially in younger generations, have been adding vegetarian aspects to their diets.[1] Therefore it is important that people find other sources for certain vitamins and minerals normally obtained through meat. One such instance is the high levels of iron deficiency among vegetarians.[2] The accepted and recommended daily intake of iron through diet is considered to be 15 mg.[3] Although it is in low concentrations in the human body, iron is extremely important to the functioning of everyday life. Iron is the central atom of hemoglobin, which is important to the transfer of oxygen throughout the body via red blood cells.[4] However, if iron overwhelms the body, which happens at levels of intake around 20 mg per day, it is possible for adverse effects to occur such as stomach upset, constipation and blackened stools.[3]

It is then important for the correct amount of iron to be ingested. For this purpose, many vegetarians eat large quantities of legumes, which are thought to be high in iron. This begs the question, however, which types of legumes offer the most acceptable level of iron per serving? Because the average person eats three meals a day, each serving of legumes should contain approximately 5 mg of iron.

Common instruments used to measure iron levels in food products are high performance liquid chromatography (HPLC), and an instrument known as a colorimeter, which measures the absorbance of certain wavelengths of light.[5] In this study HPLC was used to determine which type of legume is most appropriate for use as an iron supplement. Four types of legumes: lentils, black, garbanzo, and pinto beans (Great Value), were subjected to dry ashing using a procedure reported by the Agricultural and Environmental Services Laboratory (AESL),[6] followed by analysis with inductively coupled plasma-atomic emission spectroscopy (ICP-AES) to find the levels of iron per gram, also using a procedure created by the AESL.[7] This number for each type of legume was then used to extrapolate the amount of iron per serving of legume, accepted to be 35 g.

The closest level of iron in any given legume to 5 mg per serving size was considered to be the most acceptable, because at this level a vegetarian would be able to consume their daily recommended amount of iron without needing any exterior iron supplements, assuming a three square meal diet.

Experimental

Sample Preparation

Each type of bean was ground into a fine powder using a standard grade mortar and pestle, and the outer coating of the garbanzo beans was removed. One gram of each type of bean was weighed into a 10-mL crucible, and placed in a muffle furnace. The furnace was set to 500°C, and the samples were left to bake for four hours. After the four-hour incubation period was complete, the samples were allowed to cool to room temperature before 5mL 3N HCl was added to each. When the 3N HCl was added to each, they were boiled for five minutes, and allowed to cool down to room temperature once again. At this point, each solution was transferred into a 100-mL volumetric flask and diluted to volume. This process was carried out a total of four times, and the sixteen samples were labeled A through P. These were the samples to be tested.

Standard Preparation

To prepare the standards of calibration, 10 mL 1000-ppm stock Fe in a 4% HNO_3 solution (SCF Science) was diluted to 100 mL. This was further diluted into six standards by diluting 1, 2, 5, 7, 10, and 25 mL of the previous solution into 100 mL each.

Analysis

The standards and samples were run through standard Fe determination procedures in an ICP-AES machine.

Results and discussion

Calibration Plots

The standards created were run through the ICP-AES as calibration, and showed evidence that there was a good level of reproducibility in calibration, however there was some variability in the last calibration plot, pertaining to the last trial for each legume. The calibration plots are shown below in Fig. 1.

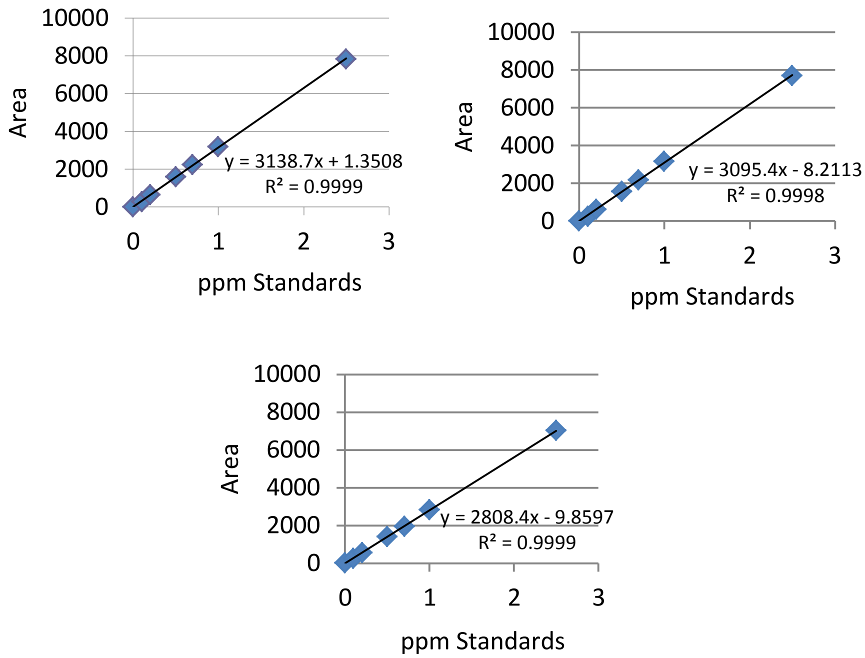

Figure 1. Calibration plots for each set of trials run, in order from first to last.

Although the plots are accurate in themselves, when all are looked at together, there is a slight variance in the slope of the third on from the first and second. This variance is likely due to the levels of argon being depleted in the ICP-AES, meaning that an equal decrease in integration for the samples run under these conditions can be noted. From this, it can be assumed that with the numbers are still accurate, as long as the new slope is used to calculate them.

The final levels of iron in each type of legume after four trials each is shown in Table 1 below. Out of the different levels of iron found, only one was found to be an outlier. This outlier is trial 2 of the black eye pea samples. The %RSD values for all findings are abnormally high, which can be attributed to the original grinding of the legumes being possibly not as fine as possible, as well as the ashing process which did not bring all samples to the level of ash necessary in order to dissolve it completely into the HCl solution. A third reason for the variance is that an internal standard was not used. Because of this, any possible fluctuations due to varying levels of argon supply would not be accounted for.

It is possible to drastically reduce the standard deviations as well as the %RSD values by removing the value in each legume category that caused the most variance. However, because of the small pool size of samples, only the one outlier was found, and thus all other possible outliers were unaccounted for under these conditions.

Table 1. The amount of iron in grams per serving in each type of legume. The highlighted box shows a statistical outlier. When the outlier is removed, the average for black eye pea becomes 0.0019, the standard deviation becomes 0.0001, and the %RSD becomes 5.3%.

	Black Bean	Lentil	Garbonzo Bean	Black Eye Pea
Trial 1	0.00390	0.00184	0.00108	0.00200
Trial 2	0.01084	0.00081	0.00167	0.00425
Trial 3	0.00259	0.00144	0.00191	0.00180
Trial 4	0.00164	0.00118	0.00195	0.00168
Average	0.0047	0.0013	0.0017	0.0024
Standard Deviation	0.0042	0.0004	0.0004	0.0012
%RSD	88.0	32.9	24.3	50.1

Conclusions

This experiment used ICP-AES in order to determine the levels of iron in different legumes. Although there was a high level of irreproducibility in the samples, the black bean was shown to contain iron levels closest to what would allow a vegetarian to obtain 15mg in a day, assuming three square meals, and no other iron sources. This is not in line with what the values on each packaging for the legumes stated. According to the packaging, the lentils did still have the least amount, however the black eye peas had the highest levels of iron. In order to improve the accuracy of these results, it would be beneficial to run more trials, as well as use a machine to more evenly ground the legumes.

References

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