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Article

Effect of different cooking methods on the levels of iron and ascorbic acid in green vegetables

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SUMMARY

Many people assume that vegetables will provide their daily nutrients irrespective of the cooking method. Various cooking methods have an impact on nutrients have an impact on nutrients such as iron and ascorbic acid. This experiment investigated the loss of iron and ascorbic acid (vitamin C) when vegetables were cooked using three different cooking methods: steaming, microwaving, and boiling. The concentration of iron was evaluated using thiocyanate colorimetry, while the concentration of ascorbic acid was determined using a titration process with iodine solution. Microwaving showed the smallest loss of iron and ascorbic acid across all vegetables. Boiling resulted in the greatest loss in iron and ascorbic acid. Considering the importance of vital nutrients such as ascorbic acid and iron, selecting the optimal cooking method will ensure the right level of nutrients after cooking. Future use of this study could include examining the effect of cooking methods on other essential nutrients that have high deficiency rates in the US such as ergocalciferol (vitamin D), calcium, and other minerals in other vegetables and food items.

INTRODUCTION

Vegetables such as *Spinacia Oleracea* (Spinach), *Brassica Oleracea* (Brussel Sprouts), and *Brassica Oleracea Italica* (Broccoli) contain several nutrients, including ascorbic acid (Vitamin C) and Iron. According to a nationwide survey by the Nurses' Health Study (NHS) and the National Cancer Institute (NCI) (1), adopting a healthy eating pattern and eating the recommended daily servings of vegetables, fruits, nuts, whole grains can lower the risk of death from chronic diseases, such as obesity, high cholesterol, and type 2 diabetes. Iron deficiency is a top nutritional disorder in the world and as many as 80% of the world's population does not have enough iron in their body (3). There are multiple forms of iron that can be ingested from vegetables, and one of these forms is non-heme iron (Fe3+) iron form that require ascorbic acid to be absorbed by the body (4). Approximately 21 million Americans have a serious ascorbic acid deficiency and fewer than 30 million people achieve optimal ascorbic acid levels (6). Iron is a key component of Heme, which is needed to transport oxygen in the blood to tissues (15). Ascorbic acid is a water-soluble vitamin capable of reduction and hydrolysis. As a reducing agent, ascorbic acid supports the transformation of ferric iron to ferrous iron (an iron that is necessary to bind, transport, and release molecular oxygen (16). Ascorbic acid facilitates iron absorption by forming a chelate with ferric iron at only an acidic pH that remains soluble at the alkaline pH of the duodenum.

A research study done by the NHS showed that an excess amount of heat can cause a loss of nutrients (3) According to a Cornell research study, microwaving does not have a major effect on ascorbic acid levels in vegetables. Exposing foods like vegetables to irradiation (such as radio waves in microwaves) produces very little chemical change in foods and doesn't change the nutritional value of food (7). Exposure to large amounts of water and heat results in strong interactions between H_2O and water-soluble vitamins (ascorbic acid) or minerals (iron), resulting in a release of nutrient content from the vegetable (8-10).

This study will test if there is a relationship between the amount of heat that a vegetable is cooked in and the concentration of iron and ascorbic content in the selected vegetables, specifically for microwaving, boiling, and steaming of vegetables. The hypothesis is based on the reasoning that (i) iron is a mineral deemed labile to large amounts of water and (ii) ascorbic acid is a water-soluble and heat-sensitive vitamin; therefore, it is more likely that ascorbic acid will decrease more than the other nutrients. Results from this study showed a decrease of almost 75% of ascorbic acid and iron in vegetables tested.

RESULTS

Selection of vegetables for the experiment

The green vegetables selected for this experiment were *Spinacia Oleracea* (spinach), *Brassica Oleracea* (Brussels sprouts), and *Brassica Oleracea Italica* (broccoli). These green vegetables were chosen for their high iron content (18). Based on USDA data, the nutritional content of 100 mg of each vegetable varies: Spinach (Iron 2.7 mg, Ascorbic acid 28.1 mg), Brussel Sprouts (Iron 1.4 mg, Ascorbic acid 85 mg), and Broccoli (Iron 0.7 mg, Ascorbic acid 91.3 mg).

Concentration level of Iron and Ascorbic Acid

A standard iron thiocyanate colorimetry test (12) was used for absolute iron concentration determination. Based on the absorbance of iron in the standard solutions, the level of concentration of iron was assessed taking average of four (4) samples. The average concentrations and absorptions of iron content under three cooking methods for three sampled vegetables emphasizes the results of this experiment (**Table 1**). For the control, a uncooked vegetable was used. This data clearly showed that the average concentration of iron was highest in microwaving and boiling has the lowest level of iron concentration. The average loss of iron concentration was highest at 60% for broccoli and low at 28% for brussels

	Iron Concentration (1e-5 mol/L)					% Loss of Iron Due to Cooking Methods					Absorption of Iron under Cooking Methods				
	Sample 1	Sample 2	Sample 3	Sample 4	Average	Sample 1	Sample 2	Sample 3	Sample 4	Average	Sample 1	Sample 2	Sample 3	Sample 4	Average
Spinacia Oleracea (Spinach)															
Uncooked (Control)	6.74	6.60	6.65	6.50	**6.62**						0.250	0.240	0.225	0.235	**0.238**
Microwaving	5.05	5.02	4.99	4.80	**4.97**	25%	24%	25%	26%	**25%**	0.190	0.181	0.170	0.174	**0.179**
Steaming	4.17	4.01	4.10	4.03	**4.08**	38%	39%	38%	38%	**38%**	0.160	0.152	0.140	0.147	**0.150**
Boiling	3.25	3.21	3.20	3.04	**3.18**	52%	51%	52%	53%	**52%**	0.120	0.113	0.111	0.113	**0.114**
Brassica Oleracea (Brussel Sprouts)															
Uncooked (Control)	4.01	3.90	3.80	3.75	**3.87**						0.150	0.140	0.135	0.138	**0.141**
Microwaving	3.77	3.70	3.60	3.62	**3.67**	6%	5%	5%	3%	**5%**	0.140	0.135	0.130	0.133	**0.135**
Steaming	3.28	3.20	3.15	3.20	**3.21**	18%	18%	17%	15%	**17%**	0.130	0.128	0.122	0.123	**0.126**
Boiling	2.87	2.76	2.75	2.74	**2.78**	28%	29%	28%	27%	**28%**	0.110	0.107	0.105	0.106	**0.107**
Brassica Oleracea Italica (Broccoli)															
Uncooked (Control)	2.79	2.70	2.65	2.55	**2.67**						0.110	0.106	0.105	0.108	**0.107**
Microwaving	2.35	2.30	2.28	2.20	**2.28**	16%	15%	14%	14%	**15%**	0.090	0.087	0.085	0.089	**0.088**
Steaming	1.70	1.65	1.62	1.60	**1.64**	39%	39%	39%	37%	**39%**	0.070	0.069	0.065	0.066	**0.068**
Boiling	1.10	1.11	1.04	1.02	**1.07**	61%	59%	61%	60%	**60%**	0.050	0.047	0.048	0.049	**0.049**

Table 1: Average Iron Concentration and loss by Cooking Method. These tables show the values of the absorption, concentration, and the loss of iron with four samples for each vegetable and each cooking method. The absorption value was found utilizing the Iron Thiocyanate test. The concentration value was found utilizing the line of Best Fit and linear regression. The loss of Iron is determined utilizing the control value of the uncooked vegetable.

sprouts (**Table 1**). The average loss of iron concentration for spinach was 52%.

During the experiment, the levels of concentration and absorption of ascorbic acid were assessed under three cooking methods. The average concentration of ascorbic acid was lowest in boiling and highest in microwaving which is displayed through **Table 2**. The average loss of ascorbic acid concentration was highest at 73% for spinach and lowest at 57% for broccoli) (**Table 2**). The loss of iron concentration averaged around 70% for brussels sprouts (**Table 2**).

DISCUSSION

The results of the experiment were consistent across all three vegetables. We collected raw data of from the four samples.

Microwaving showed the least loss of iron and ascorbic acid for all three selected vegetables. *Brassica Oleracea* Italica (Broccoli) showed the greatest loss of ascorbic acid across all cooking methods and Spinach showed the greatest loss of iron across all cooking methods. After quantifying the temperature and the amount of water for each cooking method, the tests showed that higher temperatures and larger amounts of water cause a decrease in ascorbic acid and iron, making microwaving the best cooking method for preserving ascorbic acid and iron content. The boiling method used the most water and showed the greatest decrease in iron and ascorbic acid content across all three vegetables. The steaming and microwaving methods showed a lower loss of ascorbic acid content and iron content because the vegetables had lesser contact with high temperatures and lower water content.

This study raised several questions or thought process whether the method of cooking affected the concentration of iron or ascorbic acid. The results could have been impacted by level of temperature of boiling or microwaving or quantity of

	Level of Ascorbic Acid (mg/L)- Various Cooking Methods					% Loss of Ascorbic Acid Due to Cooking				
	Sample 1	Sample 2	Sample 3	Sample 4	Average	Sample 1	Sample 2	Sample 3	Sample 4	Average
Spinacia Oleracea (Spinach)										
Uncooked (Control)	278	276	265	260	**270**					
Microwaving	144	144	140	135	**141**	48%	48%	47%	48%	**48%**
Steaming	130	130	125	120	**126**	53%	53%	53%	54%	**53%**
Boiling	78	76	72	70	**74**	72%	73%	73%	73%	**73%**
Brassica Oleracea (Brussel Sprouts)										
Uncooked (Control)	300	299	290	294	**296**					
Microwaving	172	166	164	163	**166**	43%	44%	43%	45%	**44%**
Steaming	155	157	150	147	**152**	48%	48%	48%	50%	**49%**
Boiling	90	89	85	87	**88**	70%	70%	71%	70%	**70%**
Brassica Oleracea Italica (Broccoli)										
Uncooked (Control)	338	340	345	350	**343**					
Microwaving	198	199	201	209	**202**	41%	41%	42%	40%	**41%**
Steaming	175	174	178	179	**177**	48%	49%	48%	49%	**49%**
Boiling	146	143	150	153	**148**	57%	58%	57%	56%	**57%**

Table 2: Average Ascorbic Acid Content and loss by Cooking Method (mg/L). These tables show the values of the level of the ascorbic acid in the vegetable and the loss of ascorbic acid with four samples for each vegetable in each cooking method. This value was found with the described ascorbic acid test and the formula utilized to determine this concentration.

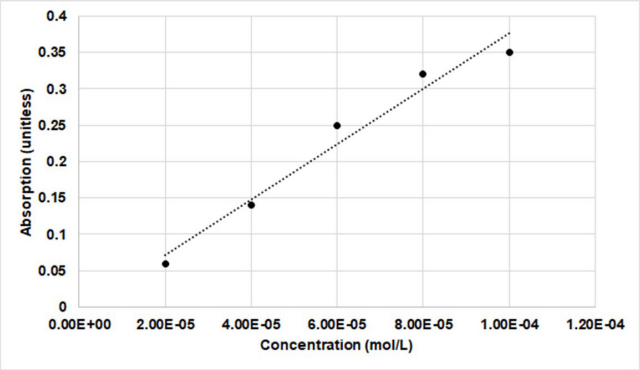

Figure 1: Measuring iron concentration in Ferric Ammonium Sulfate. Standard Solution with Linear Regression. The line of best fit was used to model the absorption measurements and iron concentration and the iron thiocyanate test was utilized.

water used for boiling. The results of the method matched the original expectation and supports the hypothesis the project as it was clear that the increased level of temperature of the boiling/microwaving or the quantity of the water used affected the change in Iron and Ascorbic Acid in the vegetables. Furthermore, the type of method of cooking could have varying impact on different types of vegetables and fruits. Additional trials within each experiment could continue to increase the accuracy of results. Furthermore, testing with other fruits and vegetables of different brands in freshness can continue to increase the accuracy of the project's results.

MATERIALS AND METHODS

This experiment analyzed the impact of three cooking methods--boiling, steaming, and microwaving--on iron and ascorbic acid (Vitamin C). The three vegetables examined were *Spinacia Oleracea* (Spinach), *Brassica Oleracea* (Brussel Sprouts), and *Brassica Oleracea Italica* (Broccoli) with a sample size of 100 mg from the standard grocery store. Four samples of each vegetable were tested to review the concentration of iron and ascorbic acid. A consistent power wattage of 1,000 watts of cooking power was used while microwaving.

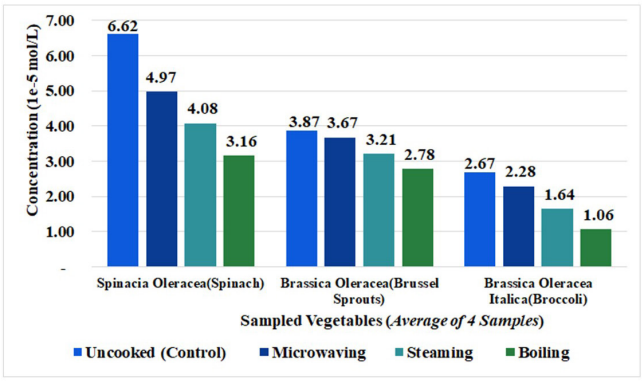

Figure 2: The Effects of Various Cooking Methods on the Concentration of Iron. The bar graph shows the concentrations of iron in the different vegetables, based on the cooking method. Boiling had the largest decrease in iron content, followed by steaming. Microwaving had the least decrease in the concentration of iron. Four replicates were performed.

Four trials were performed to account for the variability in the results and determine the concentration of iron (mol/liter) and ascorbic acid levels (mg/liter). The levels of iron and ascorbic acid of an uncooked vegetable were the control in this experiment that we compared our values against for various methods of cooking like microwaving, steaming, and boiling. The other parts including the time used for each step, test technique, time and day of testing, and various chemicals were used as a control variable (cooked and tested at the same time) in this experiment. Each vegetable sample was weighed to equal 4 g and was cooked for 8 minutes.

Measuring Iron Content

a) Preparation of Standard Solutions: Ferric ammonium sulfate was grinded in a mortar and pestle. After grinding the powder form of 2.41 g was poured into a beaker and sulfuric acid of 20 mL was added to it to sit overnight. Later 100 mL of distilled water was added to the solution to sit for two days. After two days, transferred 20 mL of ferric ion solution into a 200 mL flask and added 180 mL of distilled water. Each standard solution was prepared by adding 10mL of sulfuric acid to 10 mL of the ferric ion solution. Solid ammonium thiocyanate (38 g) was added into a 100mL volumetric flask with distilled water to make standard solution to the top of the flask.

b) Food Sample for Analysis: A vegetable sample of (4 g) was poured into a crucible and heated over a Bunsen burner until it was reduced to ash (about 8 minutes). After sample had cooled, the ash was crushed into powder. Thereafter hydrochloric acid of 10 mL) was added to the same and stirred for 5 minutes until the ash was completely soaked. Distilled water (5 mL) was later added and filtered the solution into a flask to remove the ash. This sample filtered solution was used for colorimetric analysis.

c) Colorimetric Analysis: The sample solution (10 ml) was poured into a test tube and ammonium thiocyanate (10 mL) was added it. After 15 minutes of adding thiocyanate, use the colorimeter and measure the colored solution at a wavelength of 490 nm and record the absorption measured

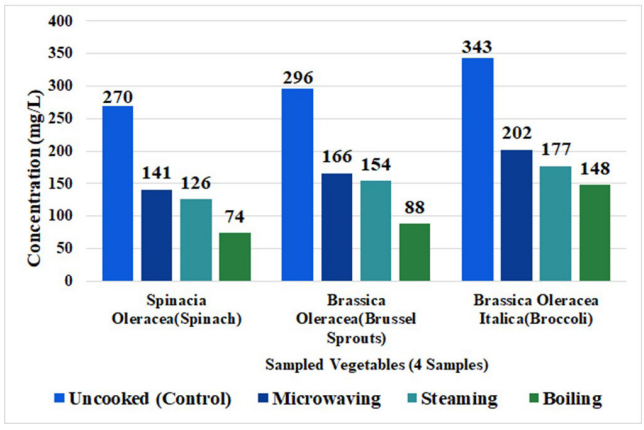

Figure 3: The Effects of Various Cooking Methods on the Concentration of Ascorbic Acid. The bar graph that shows the concentrations of ascorbic acid in the different vegetables, based on the cooking method. Boiling had the largest decrease in ascorbic acid content, followed by steaming. Microwaving had the smallest decrease in the concentration of ascorbic acid. Four replicates were performed.

by colorimeter.

The above steps were repeated for each cooking method per vegetable. Using the sample solution, the titration was repeated for four replicates.

Measuring Ascorbic Acid Content

A titration process was used to determine the number of moles of ascorbic acid that reacted with the iodine solution. Iodine and starch solutions were prepared along with the crushed vegetable samples cooked under the three methods. Titration was performed by mixing the vegetable sample (3g sample) with potassium iodate solution (5mL) and distilled water (75 mL) and the starch indicator solution (1mL) into the burette.

After completion of the titration process utilizing the Titration Test (13), the concentration level of the ascorbic acid was computed using the formula displayed in Figure One.

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