Broccoli

EXP. NUMBER	EXPERIMENT/SUBJECT	DATE
2	Determination of Vitamin C in ~~Spinach~~	5/27/09

NAME	LAB PARTNER	LOCKER/DESK NO.	COURSE & SECTION NO.
Jason Largen	Tim Van		Bio Chem Lab

07

Title: Determination of Vitamin C in ~~A~~ Biological Samples

Reference: Experiment 11B in Modern Exp. Bio Chem, 3rd
handout on blackboard: Measurment of Vitamin C

Synopsis: Determine the Ascorbic Acid contained within several Samples (raw & cooked ~~Spinach~~ Broccoli, the cooking water, and a Vitamin C tablet), by comparison to a standardized sample. Using these values, the % RDA can be determined for both raw & cooked ~~Spinach~~ Broccoli, and the ammount loss in the cooking water shown. Standardized DCIP was used to measure ascorbic acid.

Procedure:

Broccoli
raw ~~Spinach~~ was chopped up into a finely grounded mass

the mass of one serving size was recorded (~120 mL)

2 samples were obtained, one for raw and one to cook.

ascorbic acid Standard obtained

disolved in meta phosphoric/ acetic acid (acid Reagent) to 50 mL in a Volumetric flask.

Observations

mostly the flower part was used to see if it would give a higher than listed Vitamin C content.

Beaker 105.8627g
Beaker + Spinach 151.4781g
Spinach 45.6094g in one serving size

Raw Sample: 13.4384g
Cook sample: 12.2115g

0.0252g Standard.

this was to make ≅ 0.50 mg/mL

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2		5/27/09	
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Procedure

1.0mL aliquot of standard was mixed w/ acid reagent (5mL) titrated 3X w/ DCIP

Blank: 1.0 mL DI water + 5 mL acid reagent titrated once w/ DCIP

Raw:
the raw sample was extracted 3X in acid reagent. the extract was diluted to 100mL in a V. Flask.

a 10.0mL aliquot titrated 3 Xs
Blank: 10mL of acid reagent 1X

Boiled:
Sample boiled in 50mL DI water for 7 min,
extracted as w/ Raw into 100mL V Flask,
10.0mL aliquot titrated 3X
Same blank as Raw.

Cooking water returned to 50 mL w/ DI water.
5mL of it + 5mL of acid reagent titrated 3X
Blank: 5mL DI water & 5mL Acid reagent

Observations

See data tables,

I think we forgot to dilute back to 50? was around 40mL?

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Procedure

1 500 mg/tablet tablet was crushed + dissolved to 100 mL in acid reagent

a 1mL aliquote of ↑ diluted to 10.0 mL.

1.0 mL of ↑ titrated w DCIP 3 ×s

Same blank as Std. used.

observations

Data tables

Standard	titration #1	#2	#3	Blank	
Start	0.00	4.41	8.80	13.19	
finish	4.41	8.80	13.18	13.20	≈ 1 drop
added	4.41	4.39	4.39	0.01	

Raw	#1	#2	#3	Blank
Start	0.00	10.01	0.00	10.03
finish	10.01	19.83	10.03	10.12
added	10.01	9.82	10.03	0.09

Cooked	#1	#2	#3	Blank = 0.09 from about
Start	0.00	6.30	12.50	
finish	6.18	12.50	18.83	
added	6.18	6.20	6.33	

Cooking water	#1	#2	#3	Blank
Start	0.00	5.12	10.28	15.41
finish	5.01	10.28	15.41	15.45
added	5.01	5.16	5.13	0.04

SIGNATURE	DATE	WITNESS/TA	DATE

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EXP. NUMBER	EXPERIMENT/SUBJECT	DATE
NAME	LAB PARTNER	LOCKER/DESK NO. / COURSE & SECTION NO.

tablet	#1	#2	#3
Start	0.00	6.80	14,62
Finish	6,80	14,62	22,45
added	6,80	7,82	7,83

Blank = 0,01 (from Standard)

Calculations

Standard ~~0,1025g~$\frac{25,2\text{ mg}}{50\text{mL}}$~~ $= 0.504\ \frac{\text{mg}}{\text{mL}}$

Average DCIP $= \frac{(4,41-0,01)+(4,39-0,01)+(4,39-0,01)}{3} = 4,387\text{ mL}$

titrant factor $= \frac{(0.504\ \frac{\text{mg}}{\text{mL}})\times(1,0\text{mL})}{4,387\text{mL}} = 0,1149\ \frac{\text{mg ascorbic acid}}{\text{mL DCIP}}$

for avg $\frac{(,1145+,1151+,1151)}{3} = 0,1149$

$S = \sqrt{\frac{-.0004^2+,0002^2+,0002^2}{3-1}} = 3,464\times10^{-4}$ $S_m = \frac{3,464\times10^{-4}}{\sqrt{3}}$

$= 2,00\times10^{-4}$

t at 95% 2df = 4,303

titrant factor = $0,1149 \pm 0,00086$

Raw #1 $\frac{0,1149\ \frac{\text{mg aa}}{\text{mL DCIP}} \times (\text{corrected DCIP } 10,01\text{mL} - 0,09\text{mL}) \times \left(\frac{100,0\text{mL}}{10,00\text{mL}}\right)}{13,4384\text{g}} = 0,8290\ \frac{\text{mg ascorbic acid}}{\text{1g Veg.}}$

13,4384g = Starting Sample Size; $\frac{100,0\text{mL}}{10,00\text{mL}}$ = aliquot adjustment

#2 $\frac{(0,1149)\times(9,82-0,09)\times\left(\frac{100,0}{10,0}\right)}{13,4384} = 0,8319$

#3 $\frac{(0,1149)\times(10,03-0,09)\times\left(\frac{100}{10}\right)}{13,4384} = 0,8499$

SIGNATURE	DATE	WITNESS/TA	DATE

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2		5/27/09

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Raw cont.

$$Avg = \frac{.8290 + .8319 + .8499}{3} = 0.8369 \ \frac{mg\ aa}{g\ veg}$$

$$S = \sqrt{\frac{(.8290-.8369)^2 + (.8319-.8369)^2 + (.8499-.8369)^2}{3-1}} = 0.01132$$

$$S_m = \frac{0.01132}{\sqrt{3}} = 6.536 \times 10^{-3} \quad x(t@95\%@2df) \ 4.303$$

$$0.8369 \pm 0.0281 \ \frac{mg\ aa}{1g\ veg}$$

RDA = $0.8369 \times 45.6094 = 38.17$ mg / serving size

$$\%\ RDI = \frac{38.17\ mg}{90\ mg} \times 100\% = 42.41\%$$

Cooked

$$\#1 \ \frac{(6.18 - 0.09) \times (0.1149) \times \left(\frac{100.0}{10.0}\right)}{12.2115\ g} = 0.5730 \ \frac{mg\ aa}{1g\ veg}$$

$$\#2 \ \frac{(6.20 - 0.09) \times (0.1149) \times \left(\frac{100}{10}\right)}{12.2115} = 0.5749 \ \frac{mg}{g}$$

$$\#3 \ \frac{(6.33 - .09) \times (.1149) \times \left(\frac{100}{10}\right)}{12.2115} = 0.5871 \ mg/g$$

$$Avg = \frac{0.5730 + 0.5749 + 0.5871}{3} = 0.5783 \ mg/g$$

$$S = \sqrt{\frac{(.573 - .5783)^2 + (.5749 - .5783)^2 + (.5871 - .5783)^2}{3-1}} = 7.651 \times 10^{-3}$$

$$S_m = \frac{7.651 \times 10^{-3}}{\sqrt{3}} = 4.417 \times 10^{-4} \times 4.303 = 1.9 \times 10^{-3}$$

$$0.5783 \pm 1.90 \times 10^{-3} \ \frac{mg\ aa}{1g\ veg}$$

RDA = $0.5783 \times 45.6094 = 26.38$ mg/serving

$$\%\ RDI = \frac{26.38}{90\ mg} \times 100\% = 29.31\%$$

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EXP. NUMBER	EXPERIMENT/SUBJECT	DATE
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Cooling water #1 $(5.01-0.04)\times(0.1149)\times\left(\frac{100.0}{10.0}\right)\left(\frac{50.0}{5.0}\right) = 5.711\ mg^{aa}/50mL$

~~12.2118~~

#2 $(5.16-0.04)\times(.1149)\times\left(\frac{50}{5}\right)$ $5.883\ mg/50mL$

#3 $(5.13-0.04)\times(.1149)\times\left(\frac{50}{5}\right)$ $5.848\ mg/50mL$

$Avg = \frac{5.711+5.883+5.848}{3} = 5.814$

$S = \sqrt{\frac{(5.711-5.814)^2+(5.883-5.814)^2+(5.848-5.814)^2}{3-1}} = 0.0909\ mg^{aa}/50mL$

$S_m = \frac{0.0909}{\sqrt{3}} = 0.05248 \times 4.303$ $5.814 \pm 0.2258\ mg^{aa}/50mL$

lost in cooking

Tablet

#1 $(6.80-0.01)\times(0.1149)\times\left(\frac{10.0}{1.0}\right)\times\left(\frac{100}{1}\right) = 780.2\ mg^{aa}/tablet$

#2 $(7.82-0.01)\times(.1149)\times\left(\frac{10.0}{1.0}\right)\left(\frac{100}{1.0}\right) = 897.4\ mg^{aa}/tablet$

#3 $(7.83-0.01)(.1149)\left(\frac{10}{1}\right)\left(\frac{100}{1}\right) = 898.5\ mg^{aa}/tablet$

$Avg = \frac{780.2+897.4+898.5}{3} = 858.7$ $S = \sqrt{\frac{(780.2-858.7)^2+(897.4-858.7)^2+(898.5-858.7)^2}{3-1}}$

$S_m = \frac{67.99}{\sqrt{3}} = 39.25(4.303)$ $= 67.99$

$858.7 \pm 168.9\ mg^{aa}$ in tablet.

358.7 mg more than the 500 listed!!

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Conclusions/Discussion

Our sample was of raw broccoli, which according to http://www.hoptechno.com/nightcrew/sante7000/Sante7000_Detail2.cfm?ID=72201100 should have 41.01 mg of ascorbic acid per half cup. Our determination found 38.17 mg per half cup. We used mostly the flower of the broccoli and expected it to rate higher than the listed value, as it was believed to contain more of the nutrients there. They list a ½ cup as 44 grams, where we had 45.6094 grams. This makes the difference even larger, as it is .932 mg/g on the site verse our .8369 mg/g. Perhaps there were more nutrients in the stalk than we expected? your data is very comparable

Given a 90 mg per day recommended intake, our sample proved to be 42.41% of that in one serving. When compared to cooked, it was only 26.38 mg/serving and 29.31% of the RDI. We found that 5.814 mg per serving was lost in the cooking process but still within the water used to boil. This still leaves a difference of 5.976 mg between the raw and cooked/water. The cooking process may degrade the ascorbic acid during heating, as more may be converted to dehydroascorbic acid which lacks the vitamin C effects.

The 500 mg tablet was found to have quite a bit more ascorbic acid than listed, at 858.7 mg. This is 358.7 mg more than expected. Our procedures had a bit of an outlier with the first titration, but the +/- of 168.9 still doesn't even get us close to the 500 mg listed. This is somewhat scary in just how unregulated the vitamin industry is. While it is doubtful that this is harmful (and perhaps could be seen as a benefit due to getting more for your money), how many pills are on the other extreme and have only a fraction of what is listed?