

# The usefulness of titration and colorimetric analysis



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As a child I was reasoned into taking vitamin supplements as they would make me grow ' tall and strong'. Micronutrients like vitamin C are an empirical base to the formula for maintaining a healthy and balanced body, vitamin C for example has proven to be fundamentally responsible for the regeneration of our teeth, bones, teeth and skin. Moreover its unique regenerative properties have made it a key component of several pharmaceutical products that treat disease that were once thought incurable, namely scurvy. Its biological significance adequately highlights the importance given to its estimation, but one must not forget its chemical relevance. Vitamin C has proven its chemical significance both as an antioxidant and a stabiliser for to the vitamin B complex, two applications where the amount of ascorbate is of the utmost importance. As part of a tenth grade science fair project I went about determining the concentration of vitamin C in freshly squeezed orange juice. The means of volumetric analysis I used to determine the concentration of vitamin C was simple acid-base titration. I titrated vitamin C against a known quantity of analyte - sodium hydroxide. The results I was left with did not successfully reflect the literature values; in fact I was far off. For years practising chemists have attempted to develop an analytical procedure that is simple to operate, rapid, accurate, sensitive and selective and consequently a large number of procedures have sprouted with diverging capabilities[1]. My decision to carry out the following investigation has arisen from the errors present whilst using acid-base titration, as a means of volumetric analysis for vitamin C, and a wish to analyse the varying procedures that have arisen in recent years for vitamin C estimation. I have conducted the following investigation with the following research question in mind:

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How far do colorimetric readings agree with the results acquired through red-ox titration, a means of volumetric analysis, for the amount, in mg, of ascorbic acid in 100g of 3 different cultivars of Brassica oleracea obtained from a local supermarket?

Background Information:

### Vitamin C

Vitamin C or ascorbic acid is a water soluble vitamin that occurs naturally in citrus fruits, leafy vegetables, berries, tomatoes and several other plant sources. It has several beneficial properties that play an imperative role in our body's internal chemistry and therefore consistent intake of this organic molecule is of the utmost importance. Vitamin C's primary role involves helping in the production of collagen a key component in connective tissues such the ligaments, tendons, skin, bones, cartilage, and blood vessels. Along with its biological significance, Vitamin C's unique structure has also made it chemically vital.

Note: one could generalise and say that ascorbic acid and vitamin C are synonyms for each other; Vitamin C is in fact an L- enantiomer of ascorbic acid. An enantiomer is a compound whose molecular structure is not super imposable on its mirror image. The L signifies one of two forms of optical isomers, the second being D-form. Such specifications are irrelevant to this extended essay as the physical and chemical properties of two enantiomers of a chiral compound such as ascorbic acid are identical as long as they do not react with other optically active substances. Therefore for the purpose of

simplifying this essay I have used the terms Vitamin C and ascorbic acid interchangeably.

Vitamin C has an appearance ranging from a whitey to light yellowy powder. A typical molecule of this sugar-acid is composed of 6 carbon, 6 oxygen and 8 hydrogen atoms; this arrangement can be seen below:

File: L-Ascorbic acid. svg

Figure - Ascorbic acid structure[2]

<http://upload.wikimedia.org/wikipedia/commons/d/da/L-ascorbic-acid-3D-balls.png>

Figure - Ascorbic acid 3-D structure[3]

Strictly speaking vitamin C is the enolic[4]form of 3-oxo-L-gulofuranolactone. Most enols are unstable and therefore are quickly de-protonated to form an enolate ion, shown in the centre of figure 1:

[http://wpcontent.answers.com/wikipedia/commons/thumb/5/5c/Ascorbic\\_acidity3.png/500px-](http://wpcontent.answers.com/wikipedia/commons/thumb/5/5c/Ascorbic_acidity3.png/500px-Ascorbic_acidity3.png)

[Ascorbic\\_acidity3.png](http://wpcontent.answers.com/wikipedia/commons/thumb/5/5c/Ascorbic_acidity3.png/500px-Ascorbic_acidity3.png)

Figure - Transfer of electron pairs in ascorbic acid[5]

Figure - Distribution of Electrons in enolate ion[6]

In the case of vitamin C it can further lose a proton as shown on figure 1 to the adjacent double bond, in turn stabilising the enolate ion. The transfer of

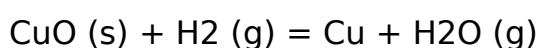
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electrons between the hydroxyl and carbonyl groups and the existence of two resonance structures for the negative enolate ion are also characteristic of a vinylogous carboxylic acid.[7]It is important to note that vitamin C is a reducing agent (electron donor) and has a standard electrode potential of 0.08V. It is also water soluble because of the numerous hydroxyl groups that are found in its structure and can therefore form hydrogen bond with water molecules.

### Redox reactions

For this investigation the effectiveness of colorimetry and titration will be explored by means of redox reactions in acidic media. This family of reactions is fundamentally concerned with the transferral of electrons between the species in a chemical system. Like acid-base reactions this family of reactions is “ antagonistic” in nature, they occur in opposing pairs i. e. every reduction is accompanied by an oxidation reaction and vice versa. A Redox reaction is often characterised by the loss and subsequent gain in oxygen between two species in a system for example:

Copper (II) oxide + Hydrogen = Copper + water



In this case since copper oxide is losing oxygen, it is being reduced and subsequently one could say hydrogen is being oxidised into water due to the gain of oxygen. Similarly we could suggest that Hydrogen is the reducing agent as it takes oxygen away and copper oxide is the oxidizing agent since it gives away oxygen. Similarly a gain in hydrogen characterises a reduced

species and a loss in hydrogen characterises an oxidised species. Yet not all redox reactions will involve oxygen, the reaction between sodium and chloride is redox in nature yet there are no molecules of oxygen involved.

This gives a second set of definitions for redox reactions:

If a species loses one or more electrons during a reaction it has been oxidised(oxidation), and if it gains electrons it has been reduced(reduction)

A species removing electrons is an oxidising agent( species itself is reduced), and if a species donates electrons it is a reducing agent( species itself is oxidised)

Note: In several chemical reactions the aforementioned transferral of electrons may never actually happen and because of this reason a more accurate description of oxidation is, an increase in the oxidation number of a species and therefore a reduction is defined as a decrease in the oxidation number of a species.[8]

Redox reactions can be broken down into half equations to demonstrate the transferral:

[http://upload.wikimedia.org/wikipedia/commons/8/8c/Redox\\_Halves.png](http://upload.wikimedia.org/wikipedia/commons/8/8c/Redox_Halves.png)

Figure - Half equations[9]

## Experiment 1

Though colorimetry is being tested here, the principle reaction here is a redox reaction taking place between ascorbic acid and acidified Potassium

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permanganate. In its oxidised state Potassium permanganate ( $\text{KMnO}_4$ ) is a purple crystallised structure. Considering it is a very strong oxidising agent, acidified  $\text{KMnO}_4$  (aq) was used as the oxidising titrant in this red-ox titration. In an acidic environment[10]it readily oxidises ascorbic acid consequently being reduced to Manganese (II) ions from its Manganate (VII) oxidised state.

Half equation:

Through our experimentation it was found that generally speaking the red-ox reaction harbours a decrease in the intensity of the purple inclining me to believe that eventually the solution would become colourless as all Manganate ions would be transformed into Manganese ions. This colour change was detected as an increase in transmittance, for successive concentrations of ascorbic acid by a colorimeter set to 565nm[11]. This positive correlation is evident on the trend lines presented on graphs 2 and 3. Yet one must account for the so to say false drops in transmittance. The drop can be explained by the formation of  $\text{MnO}_2$  particles which give the solution a dark brownish colour:

Eventually the  $\text{MnO}_2$  particles precipitate under the influence of the sulphuric acid, allowing for the trend to continue. This phenomenon is further explained in the appendix. One should also note the fact that  $\text{MnO}_4$  has a standard electrode potential of 1.51V and considering that vitamin C has a lower one there is sure to be an effective redox reaction.

Sulphuric acid

Sulphuric plays a more significant role than simply providing the acidic conditions for the relevant redox reactions. Ascorbic acid itself is a weak acid and therefore would undergo a neutralisation reaction if it were in a basic environment (a pH higher than 7). It would thereafter be oxidised and the resulting product dehydroascorbic acid, would undergo a hydrolysis reaction due to the alkalinity of the solution. The ester bonds in Vitamin C would consequently break - thus the degradation of vitamin C:[12]

vitamin C decomposition

Figure - Degradation of Vitamin C[13]

Furthermore sulphuric acid helps delay the spontaneous oxidation of ascorbic acid brought about by atmospheric conditions by providing a protective layer.

Colorimetry

A colorimeter measures the colour intensity of a sample in terms of the red, blue, and green components of light reflected from the solution. The instrument operates on the basis of Beer-Lambert's law[14] which states that the absorption of light transmitted through a species is directly proportional to the concentration of that species. Effectively we are measuring what fraction of a beam of light passing through a sample reaches a detector on the other side of the sample. The coloured analyte (species being studied) may absorb photons in the beam of light thus reducing the number of photons in the beam of light leaving the solution. The number of photons detected gives a value of the % transmittance which is the independent

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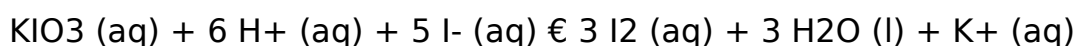


variable in this part of the investigation. A calibrated chart for known concentrations of ascorbic acid vs. %Transmittance was made to help identify unknown concentrations of ascorbic acid in different assays with respect to measured %transmittances.

### Iodometry

The redox reaction between iodine and ascorbic acid can be used to calculate the amount of ascorbic acid in the cultivars of Brassica oleracea.

The pre-prepared solution was made on the basis of this reaction:



As we can see iodine solution can be acquired through this reaction. The iodine is quickly reduced into its iodide ion in the presence of ascorbic acid.

The ascorbic in turn is oxidised into dehydroascorbic acid:

Figure - oxidation of ascorbic acid[15]

Once the ascorbic acid has all been oxidised, iodine will remain in excess. As the iodine molecules build up they will react with the iodine ions to form a blue-black complex with the starch, marking the end-point of the titration.

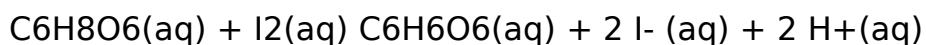
These are the relevant redox half equations:

$\text{I}_2 +$

Reduction half reaction for Iodine at a slightly acidic pH

Oxidation half reaction for vitamin-C ( $\text{C}_6\text{H}_8\text{O}_6$ ) at a slightly acidic pH

Final redox reaction :



(ascorbic acid) (dehydroascorbic acid)

### **Preparing potassium permanganate (KMnO<sub>4</sub>) solution:**

Precisely fill a 1 litre Erlenmeyer flask with 600cm<sup>3</sup> (600ml) of distilled water

Measure out 4g of crushed potassium permanganate crystals, which are to be added into the Erlenmeyer flask containing the distilled water

Swirl the solution until all the KMnO<sub>4</sub> has dissolved and an intense purple solution has been formed.

### **Preparing standardised solutions of ascorbate:**

Pipette 5ml of 1M sulphuric acid into each of the test-tubes

Carefully Pour 100 ml of distilled water in each of the test-tubes and seal them with a bung

Place all the test-tubes in some form of refrigerated unit to delay the ascorbic acid from being oxidised

### **Calculating calibration chart:**

Set up your data logging equipment and set your colorimeter so that a green LED light of wavelength 565nm is transmitted across cuvettes

Pipette 3ml of distilled water into a fresh cuvette and calibrate the colorimeter to the transmittance through distilled water is a 100%

Pour 30 ml of the pre-prepared potassium permanganate solution into a 200 ml beaker containing 50 ml of distilled water

Pipette 3ml of the 30ml  $\text{KMnO}_4$  (aq) into two fresh cuvettes

Place these cuvettes one after another into the colorimeter

Measure the transmittances for 12 seconds and record them as trial 1 and 2 for the transmittance for when no ascorbic acid is present

Thoroughly clean and dry the 200ml beaker and thereafter carefully pour into it, 30ml of the pre-prepared potassium permanganate solution

Remove test-tube 1 (containing 0.009 grams of ascorbic acid) and pour the contents into the 200ml beaker containing  $\text{KMnO}_4$  (aq)

After allowing the acidified red-ox reaction to continue for 5 minutes pipette 3ml of the product twice into two different cuvettes

Place the cuvettes one after the other into the colorimeter, set to wavelength 565nm, and record the respective transmittances for test tube 1 as trial 1 and 2

Repeat steps 14-17 for each test-tube thus retaining transmittance values for all the different quantities of ascorbic acid

Plot a suitable calibration chart for the average of the two trials of each sample

## **Extracting Vitamin C from cultivars of Brassica oleracea:**

Collect the necessary apparatus and clean it

Set a hot plate to 50 degrees centigrade and place a beaker with 100 ml water and a thermometer on it

Measure out 100g of Cabbage and place them in a beaker

Once the water has reached 50 degrees remove the beaker of water from the hot plate and switch it with the beaker of leaves

Leaves the leaves on the hot plate for 5 minutes to slow down the action of ascorbic acid oxidase

Cut the sample of cabbage as much as possible to facilitate the grinding

Note: Try to perform neat cuts so as to not release too much ascorbic acid oxidase which would obscure the results

Add 5ml (as with the standardised solutions) of sulphuric acid to the leaves, add some sand to help make the grinding of the leaves easier

Grind the mixture using a mortar and pester until an homogenate state is achieved ( you may add 10 ml of water to help achieve the homogenate state faster but this could obscure your results considering vitamin C is water soluble and it must be added before adding the acid)

The homogenate mixture produced can now be filtered through the Mira cloth into a 200ml measuring beaker as shown in Appendix

To attain maximum yield you may squeeze the Mira cloth to squeeze out any remaining fluid into the measuring cylinder, yet ensure that you have precisely 50 ml of the filtered liquid present in the measuring cylinder

Note: ensure that 50 ml is present in each of the measuring cylinders regardless of the source

Precisely dilute the filtrate to a 100 ml, seal the beaker and refrigerate it

Steps 20- 30 will have to repeated for each cultivar so that you will end up with 3 beakers, each with an equal volume of the respective vegetable filtrate solution

### **Analysing cultivars for Vitamin C:**

Repeat steps 8 and 9

Pour 30ml of the aqueous potassium permanganate into a 200ml beaker

Remove the cabbage filtrate from the fridge and pour it into the 200ml beaker containing  $\text{KMnO}_4(\text{aq})$

Allow the reaction to proceed for 5 minutes

Pipette 3ml of this mixture twice, into two different cuvettes

Place the cuvettes one after another into the colorimeter that had been calibrated

Record the transmittance for both cultivar solution for 12 seconds and record the results as trial 1 and 2

Repeat steps 33-38 for each cultivar

### **Carrying out the Red-ox titration – Titrate pre-prepared iodine solution against samples of ascorbic acid extracted from each vegetable:**

Make new samples of cultivar filtrate as outlined by steps 20 - 31

Add the 100 ml of the cabbage filtrate solution with the unknown concentration of ascorbic acid, to a 150 ml Erlenmeyer flask

Carefully Titrate[16]the pre-prepared iodine solution[17]to this analyte drop by drop until the end point, a bluish-black colour that persists after 30 seconds of swirling, is reached

Repeat this experiment at least thrice; to obtain three different measurement that should agree with each other within 0. 1 ml - average the three results obtained

Steps 40 to 44 have to be repeated for every cultivar filtrate

Record all your results

Table 18 - Averages for all trials and final average

No of moles of AA present in 100ml of aqueous potassium permanganate

Average % transmittance recorded by colorimeter in trial 1 (+/- 0. 01%)

Average % transmittance recorded by colorimeter in trial 2 (+/- 0. 01%)

Average % transmittance of trials 1 and 2 -used for calibration chart - (+/- 0.01%)

Cultivar of Brassica oleracea

Average % transmittance recorded by colorimeter in trial 1

Average % transmittance recorded by colorimeter in trial 2

Average % transmittance of trials 1 and 2 - used for calibration chart

Estimation for Mg of ascorbic acid in 100g of Broccoli:

Number of moles = 0.00048 moles

Concentration of ascorbic acid = 0.0048 mol dm<sup>-3</sup> (L<sup>-1</sup>)

Number of moles = Concentration Volume

Number of moles = 0.0048 0.1 = 0.00048 moles

Thus both graphs show same data, I shall use transmittance vs. Moles graph from now on as this way it is quicker to calculate the mass of ascorbic acid per 100g of the relevant cultivar of Brassica oleracea.

Number of moles = 0.00048 moles

Mass = Number of moles Mass of one moles (Mr)

Mass = 0.00048 176.12

= 0.08454 g/100ml

$$= 0.08454 \text{ g/100g}$$

$$= 84.54 \text{ mg of AA in 100g of broccoli (+/- 1.04\%)}$$

Estimation for Mg of ascorbic acid in 100g of Cauliflower:

$$\text{Number of moles} = 0.00023 \text{ moles}$$

$$\text{Mass} = \text{Number of moles} \times \text{Mass of one moles (Mr)}$$

$$\text{Mass} = 0.00023 \times 176.12$$

$$= 0.04051 \text{ g/100ml}$$

$$= 0.04051 \text{ g/100g}$$

$$= 40.51 \text{ mg of AA in 100g of cauliflower (+/- 2.17\%)}$$

Estimation for Mg of ascorbic acid in 100g of Cabbage:

$$\text{Number of moles} = 0.00014 \text{ moles}$$

$$\text{Mass} = \text{Number of moles} \times \text{Mass of one moles (Mr)}$$

$$\text{Mass} = 0.00014 \times 176.12$$

$$= 0.02466 \text{ g/100ml}$$

$$= 0.02466 \text{ g/100g (+/- 3.57\%)}$$

$$= 24.66 \text{ mg of AA in 100g of cabbage}$$



Amount of iodine solution titrated into Broccoli cultivar before end-point is reached (+/- 0.05ml)

Amount of iodine solution titrated into Cauliflower cultivar before end-point is reached (+/- 0.05ml)

Amount of iodine solution titrated into Cabbage cultivar before end-point is reached (+/- 0.05ml)

Trial 1

49.00

28.00

21.00

Trial 2

51.00

25.00

16.00

Trial 3

50.00

28.00

20.00

## Averages

50.00

27.00

19.00

Facts to bear:

Molar ratio of iodine to ascorbic acid is 1: 1;

ascorbic acid + I<sub>2</sub> → 2 I<sup>-</sup> + dehydroascorbic acidConcentration of iodine solution is 0.01 mol L<sup>-1</sup>Mr of Ascorbic acid for the purpose of this experiment is rounded to 176.12 g mol<sup>-1</sup>

It is assumed that 100 ml = 100g

End point is where blue colour persists for more than 30 seconds

Broccoli:

On average 50 ml of iodine was used until end point reached

No of moles = Volume concentration

No. Of moles of iodine used = 0.050 0.010

= 5.0 10<sup>-4</sup> moles of iodine solution used

Since ratio is iodine to ascorbic acid is 1: 1 number of moles of ascorbic acid present in solution is equal to iodine used in titration. Number of moles of ascorbic acid present in 100 ml solution of broccoli juice =  $5 \times 10^{-4}$  moles

Mass = Number of moles  $M_r$

Mass of ascorbic acid present in 100 ml of broccoli juice solution =  $5 \times 10^{-4}$  moles  $176.12$

=  $0.08806\text{g} / 100\text{ml}$

=  $88.06\text{ mg of AA in } 100\text{g of broccoli} (+/- 0.10\%)$

Cauliflower:

On average 27 ml of iodine was used until end point reached

No of moles = Volume concentration

No. Of moles of iodine used =  $0.027 \times 0.010$

=  $2.7 \times 10^{-4}$  moles of iodine solution used

Mass = Number of moles  $M_r$

Mass of ascorbic acid present in 100 ml of cauliflower juice solution =  $2.7 \times 10^{-4}$  moles  $176.12$

=  $0.04755\text{g} / 100\text{ml}$

=  $47.55\text{ mg of AA in } 100\text{g of cauliflower} (+/- 0.19\%)$

Cabbage:

On average 19 ml of iodine was used until end point reached

No of moles = Volume concentration

No. Of moles of iodine used =  $0.019 \times 0.010$

=  $1.9 \times 10^{-4}$  moles of iodine solution used

Mass = Number of moles Mr

Mass of ascorbic acid present in 100 ml of cabbage juice solution =  $1.9 \times 10^{-4}$  moles  $176.12$

=  $0.03346\text{g}/100\text{ml}$

=  $33.46 \text{ mg of AA in } 100\text{g of cabbage } (+/-0.26\%)$

Concluding Investigation

The point of this investigation was to explore the following research question:

“ How far do colorimetric readings agree with the results acquired through red-ox titration, a means of volumetric analysis, for the amount, in mg, of ascorbic acid in 100g of 3 different cultivars of Brassica oleracea obtained from a local supermarket?”

Before assessing the proximity of the two sets of results obtained, I have provided a set of literature values to help determine not only the accuracy of

each individual experiment but also to eventually determine which of the two procedure delivered more promising results.

Value of ascorbate in 100g of raw cabbage: 36. 60 mg (20% refuse)[18]

Value of ascorbate in 100g of raw broccoli: 89. 20 mg (39% refuse)[19]

Value of ascorbate in 100g of raw cauliflower: 48. 20 mg (61% refuse)[20]

Surprisingly the experimental values for both experiments weren't too far from the literature values provided above. Through colorimetry means it was found that 24. 66 mg of ascorbic acid was to be found in the 100 gram assay of cabbage. As for the results obtained through iodometry it was found that the 100 g sample of cabbage consisted of 33. 46mg. From the data collected it is evident that the iodometric means of analysis was more effective in determining ascorbate amount in cabbage as it returned a value that was 91. 42% of the literature value whereas the colorimetry means returned a value that was 67. 38% of the literature value. More significantly the error percentage concerning the cabbage was relatively lower for the iodometric titration than the error percentage for the Colorimetry based values, +/-0. 26% and +/-3. 57% respectively. The two aforementioned trends; difference between literature and experimental values and error percentage was similarly evident for both the other cultivars. Iodometry returned a value of 47. 55mg/100g of cauliflower assay with an error percentage of +/- 0. 19% and by means of colorimetry a lower value of 40. 51mg/100g was obtained with a higher error percentage of +/- 2. 17%. Finally the broccoli assay too returned values that fit the aforementioned trend. The experimental values derived through colorimetry suggested a typical 100g assay of broccoli

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contains 84.54 mg of ascorbate with an error percentage of  $\pm 1.04\%$ . The iodometric experiment returned a value in close proximity to the literature value; the results suggested that a typical 100g assay of broccoli may contain 88.06mg of ascorbic acid with a minimal error percentage of 0.10%.

Through the data provided above one could superficially suggest that the redox titration involving iodine and ascorbic acid has much better served the purpose of determining ascorbic acid quantities in the cultivars of *Brassica oleracea* than using colorimetry to track the redox reaction between  $\text{KMnO}_4$  and ascorbic acid. Though this seems to be the case it is important to highlight the factors that could have affected the reliability of both experiments. The following variables were controlled:

Source of cultivars

Amount of cultivar in each assay

Equal amounts of sulphuric acid used in all assays

All assays were equally heated

The results of two separate redox reaction was being measured in both experiments

The means of extracting vitamin C out of the cultivars

Yet it was not possible to control all the variables. One should primarily note how vitamin C is not equally displaced throughout a species of vegetable or fruit or any other source. Therefore the conjecture stated above about

reliability of both the investigations holds true only if the following is assumed; vitamin C is equally present throughout all the cultivars of Brassica oleracea used. Practically speaking this was not the case as the iodometry experiment was carried out before the colorimetry thus leaving the cultivars more exposed to atmospheric oxidation thus in general there would be less ascorbic acid in the samples of cultivar mixed with  $\text{KMnO}_4$  than in the samples upon which iodine was titrated on. Furthermore the quantities of refuse with our experiments were inconsistent unlike that of the generalised literature values, though not calculated it was evident that our primitive school based techniques led to much more refuse than the stated amounts of the literature values. This applies foremostly to the cabbage, from which it was the hardest to retain an assay from and thus had a refuse of much higher than 20%. Inconsistent refuses would've have meant that the ascorbic acid extracted is not representative of the full 100g assay. One should also take account of the fact that atmospheric oxidation is inconsistent and irregular, because the experiments lasted over days. This could explain the lower values obtained for the longer lasting colorimetry investigation. The length of this investigation means that the sample solutions of vitamin C used to create the calibrated graph were sure to have lost vitamin C, despite being placed in a fridge; this questions the reliability of our calibrated graph and furthermore explains why the first investigation returned lower values of ascorbic acid in the 100g assays. Another problem with the colorimetry experiment was the fact that we used  $\text{KMnO}_4$  solution, which is generally contaminated with  $\text{MnO}_2$ . The  $\text{MnO}_2$  as suggested earlier on in the investigation leads to false drops therefore leading once again to a questionable calibrated graph which has a general inward inclination thereof

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giving lower experimental values for ascorbate concentrations. The  $\text{MnO}_2$  induced drops basically lead to varying readings from the colorimeter which made it difficult to produce an accurate calibrated chart. Furthermore  $\text{KMnO}_4$  has the tendency to react with contaminants found in water due to its oxidative properties, thereby further discrediting the calibration curve made. Another major uncertainty was my use of iodine for the redox titration, not only did I not control the oxidising agent but iodine is evidently not specific enough to estimate amount of ascorbic acid in vegetable cultivars. This is because there are other reducing agents such as uric acid in broccoli, which interfere with the titer values. Since ascorbic acid is not the only one responsible for reducing iodine to iodide ions the assumed end point is questionable. This further suggests the higher values for vitamin C amount found through iodometry and explains why the results are so accurate relative to the literature values. There are several other uncertainties that one could consider such as the errors with the equipment, the loss of vitamin C whilst extracting it from the various cultivars and so forth yet they do not make a difference as both experiments are exposed to these uncertainties and thus in effect they cancel out and can be ignored.

We have assessed the flaws in each of the means of analysis and taking all aspects into consideration I find the permanganate and vitamin C redox reaction tracked by a colorimeter a better method. Firstly because we are not relying on an indicator, permanganate is an indicator itself, and the concentration does not deteriorate if well stopped in amber bottles whereas concentrations of iodine dramatically deteriorate overtime. Furthermore it



seems the flaws with this investigation are more technical in nature and can be m