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#### **ABSTRACT**

(+) - Glucose rotates the plane of polarised light clockwise, while (-) - Fructose does the opposite and rotates the plane of polarised light anticlockwise. By using a polarimeter, the observed rotation of solutions of each monosaccharide can be measured. However, it is interesting that there is such a lack of data on the combination of two optically active substances and thus investigating the optical rotation of a mixture of two monosaccharides would provide insight into this matter.

Thus, the <u>Research Question</u> for this essay is "When solutions of (+) - Glucose and (-) - Fructose are mixed, is there a relationship between the observed rotation of polarised light caused by this solution and the concentration of the constituent monosaccharide solutions?"

The two monosaccharides used in this experiment were (+) - Glucose and (-) - Fructose. This is because they are two of the most abundant forms of monosaccharides in nature. By making solutions of both monosaccharides at molar concentrations of 0.10moldm<sup>-3</sup>, 0.20moldm<sup>-3</sup>, 0.30moldm<sup>-3</sup>, 0.40moldm<sup>-3</sup> and 0.50moldm<sup>-3</sup>, the observed of each solution was measured using a polarimeter (Philip Harris, Y42200/1). Then the monosaccharide solutions were combined at different concentrations; 0.10moldm<sup>-3</sup> (+) - Glucose with 0.50moldm<sup>-3</sup> (-) - Fructose, 0.20moldm<sup>-3</sup> (+) - Glucose with 0.30moldm<sup>-3</sup> (-) - Fructose etc. From these mixtures, the observed rotation was measured and results were plotted against the concentration of each monosaccharide.

Conclusively, the results of this experiment were indicative of a *linear relationship* between the observed rotation of the mixture and the concentration of the two constituent monosaccharides, hence answering the <u>Research Question</u>. The limitations of this essay are that this relationship has only been found in two very specific organic optical isomers, and it is not possible to generalise from this amount of data.

[291 Words]

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## INTRODUCTION

Since the early 19th century, optical rotation in organic compounds has been observed. In 1848, Louis Pasteur discovered two different types of crystal present in crystallized tartaric acid1 (now called 2,3-dihydroxybutanedioic acid) - these crystals were found to be mirror images of each other. After separating the two enantiomers, he found that one form rotated the plane of polarized light to the right, while the other rotated the plane to the left. Pasteur himself did not understand the reason why these two enantiomers rotated polarized light in different directions. In 1874, J.A le Bel and J.H van't Hoff suggested that these molecules were based around a tetrahedral shape<sup>1</sup>. They showed that if a molecule has four different atoms or groups surrounding a central carbon atom, it is possible to obtain two molecules which are mirror images of each other.1

These optical isomers were non super-imposable and are now said to be chiral - "A molecule will be chiral if it has neither a plane nor a centre of symmetry"2. The diagram below shows how two mirror image forms of lactic acid, a molecule with a central carbon atom surrounded by four different groups, can be produced.

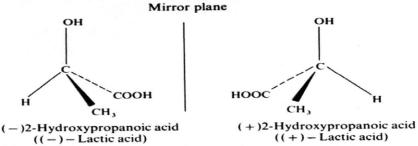


Figure 1.1 Enantiomers of Lactic Acid3 From Principles of Organic Chemistry by Peter Murray. Reprinted by permission of Pearson Education

In this essay, the optical rotation of two of the most naturally abundant monosaccharides will be investigated. A solution of (+) - Glucose rotates the plane of polarised light clockwise, similarly a solution of (-) - Fructose rotates the plane of polarised light anticlockwise. Since the two sugars rotate the plane of polarised light in opposite directions and different magnitudes, I thought that it would be interesting to investigate what the observed rotation of a mixture of the two would be. This led me to the following Research Question;

When solutions of (+) - Glucose and (-) - Fructose are mixed, is there a relationship between the observed rotation of polarised light caused by this mixture and the concentration of the constituent monosaccharide solutions?

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<sup>&</sup>lt;sup>1</sup> Matthews, Philip. 1992. <u>Advanced Chemistry: Organic and Inorganic</u>. Chapter 110: Optical Activity. Cambridge University Press.

<sup>&</sup>lt;sup>2</sup> Matthews, Philip. p732.

<sup>&</sup>lt;sup>3</sup> Murray, Peter R.S. 1972. Principles of Organic Chemistry. Chapter 25: Carbohydrates. Heinemann Educational Books.

As with lactic acid, Glucose has enantiomeric forms due to the nature of the molecular arrangement. However, there are four chiral centers in Glucose, making 16 possible enantiomeric combinations possible<sup>4</sup>. Structures of D-Glucose and L-Glucose are shown below with chiral centers as they are the most naturally abundant.

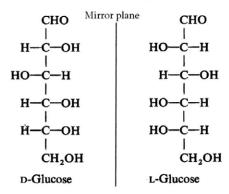


Figure 1.2 Enantiomers of Glucose showing the chiral centers<sup>5</sup>.

From Principles of Organic Chemistry by Peter Murray.

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## Relevance of Optical Isomerism:

Optically active substances are extremely important in medicine and biochemistry: there have been many Nobel Prizes in Chemistry awarded to chemists who have done work in the field of chirality and optical isomerism such as Jacobus H. van't Hoff (1901), D.H.R. Barton (1969) and Vladimir Prelog (1975)<sup>6</sup>. As seen in the case of the pregnancy drug Thalidomide where one enantiomer of the drug treated morning sickness, while the other caused deformities in babies<sup>7</sup>, chirality is important not only in chemistry labs but in the real world. In our bodies, active sites such as neuroreceptors have a certain conformity which must match the correct optically active drug<sup>8</sup>; otherwise the drug will not work or will produce the wrong medicinal effect due to the non super-imposable nature of the optically active drug. In fact, all sugars in our body are in the D-form, whereas all amino acids are in the L-form. Take for example Penicillin; the activity of this drug is said to be *stereoselective*, it only attacks the peptide links of D-alanine, killing the cell walls of bacteria with D-alanine peptides, but not affecting our own membranes as humans have L-amino acids. In the past few decades, there has been pharmaceutical research into developing 100% pure optically active solutions

<sup>&</sup>lt;sup>4</sup> Barton, D.H.R. 1969. The Principles of Conformational Analysis.

<sup>&</sup>lt;sup>5</sup> Murray, Peter R.S. 1972. <u>Principles of Organic Chemistry</u>. Chapter 25: Carbohydrates. Heinemann Educational Books.

<sup>&</sup>lt;sup>6</sup> The Official Website of the Nobel Foundation: All Nobel Laureates. URL:

<sup>&</sup>lt;a href="http://nobelprize.org/nobel\_prizes/lists/all/">http://nobelprize.org/nobel\_prizes/lists/all/</a> Retrieved on 2006-11-02

<sup>&</sup>lt;sup>7</sup> The History of Thalidomide. URL:

<sup>&</sup>lt;a href="http://www.thalidomide.ca/en/information/history\_of\_thalidomide.html">http://www.thalidomide.ca/en/information/history\_of\_thalidomide.html</a> Retrieved on 2006-09-21.

<sup>8</sup> Introducing Amino Acids. URL:

<sup>&</sup>lt;a href="http://www.chemguide.co.uk/organicprops/aminoacids/background.html">http://www.chemguide.co.uk/organicprops/aminoacids/background.html</a> Retrieved on 2006-10-01

containing only one enantiomer<sup>9</sup>. This potentially allows a drug to be twice as effective as it would be if both pairs are present, since usually only one isomer induces the effect of the drug. In the case of nicotine, isolation of the (+) - nicotine has been possible and this enantiomeric form of the drug has been shown to be less lethal on the mice it was tested on compared to (-) - nicotine<sup>10</sup>, indicating that the nicotine receptors in these mice are stereospecfic. Additionally, optically active glucose sensing and monitoring are at the forefront of many medical issues concerning the testing of blood levels for glucose noninvasively<sup>11</sup>. Thus, it can be seen how optical isomerism and optical rotation is important in the developing fields of medicine and pharmacology. Lastly, I found it interesting that literature on the combinations of two different optical isomers in a mixture is scarce, so research into the topic will provide an insight into this matter which is otherwise hard to find. This information could be useful if there is a mixture of two monosaccharides of unknown concentrations. Finding a relationship between the concentration of each monosaccharide and observed rotation would then allow identification of the concentrations of each sugar.

#### Naming Optical Isomers.

Optically active substances either rotate the plane of polarised light clockwise or anticlockwise. If the enantiomer rotates the plane in the clockwise direction, it is said to be *dextrorotary* and is designated a lower case d or a (+) in the naming system. The other enantiomer will rotate the plane of polarized light anticlockwise and with the same magnitude (so a racemic mixture - a mixture of two enantiomers which rotate the plane of polarised in opposite directions, but of the same magnitude - will not rotate polarized light) and is called *laevorotary* and is given a lower case I or a (-) in the naming system. Thus, naturally occurring glucose which is dextrorotary is called dextrose, while naturally occurring fructose is laevorotary and is called levulose 12.

#### The Mutarotation of Monosaccharides.

Since Glucose is being used in the experiment, it is important to note some properties of this monosaccharide in solution. Glucose in acyclic form has four chiral centers; hence there are sixteen optical isomers of the molecule. However, the most common isomer by far is (+) - Glucose, a cyclic isomer which rotates the plane of polarised light clockwise, hence it is *dextrorotary* and also called dextrose<sup>12</sup>. In solution the cyclic (+) - Glucose, also known as  $\alpha$ -glucose, breaks open to take the acyclic form, and occasionally when the cyclic form is remade, the configuration of OH and H groups on the  $C_1$  atom switches to form  $\beta$ -glucose.

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<sup>&</sup>lt;sup>9</sup> Kasche, V., Galunsky, B., Nurk, A., Piotraschke E. and Rieks, A. 1996. <u>The dependency of the stereoselectivity of penicillin amidases-enzymes with R-specific S1- and S-specific S'1-subsites- on temperature and primary structure</u>. Biotechnology Letters, Vol 18, Issue 4 (455-460).

<sup>&</sup>lt;sup>10</sup> Aceto, M.D., Bradshaw, T.J., Dewey, W.L., Harris, L.S., Izazola-Conde, C., Martin, B.R., May, E.L., Uwaydah, I.M., Vincek, W.C. 1979. Optically pure (+)-nicotine from (+/-)-nicotine and biological comparisons with (-)-nicotine. Journal of Medicinal Chemistry, Vol 22, Issue 2 (174-177).

<sup>&</sup>lt;sup>11</sup> Klonoff, DC. 1997. Noninvasive blood glucose monitoring. Diabetes Care, Vol 20, Issue 3 (433-437).

<sup>&</sup>lt;sup>12</sup> Murray, Peter R.S. 1972. <u>Principles of Organic Chemistry</u>. Chapter 25: Carbohydrates. Heinemann Educational Books.

This is called the mutarotation of glucose and is shown in *Figure 1.3* below.

$$^{\circ}CH_{2}OH$$
 $^{\circ}CH_{2}OH$ 
 $^{\circ$ 

From Principles of Organic Chemistry by Peter Murray. Reprinted by permission of Pearson Education

Both  $\alpha$ -glucose and  $\beta$ -glucose are *dextrorotary* (explained previously on page 3) in detail in the paragraph below), but  $\alpha$ -glucose rotates the plane of polarised light to a greater degree (+112°) than  $\beta$ -glucose (+18.7°). Not all  $\alpha$ -glucose mutarotates and changes configuration to  $\beta$ -glucose so over time equilibrium is reached at +52.7°, thus it is possible from the rotation of glucose in solution to determine if the solution is fresh or has been left and mutarotation has occurred. The implications of mutarotation have consequences for this experiment, since glucose is used in solution. Mutarotation can also be applied to other molecules which are optically active and this is discussed in the evaluation.

#### How is Optical Rotation measured by a Polarimeter?

Ordinary light consists of electric and magnetic waves; both perpendicular to each other and to the direction of travel of the light<sup>14</sup>. Additionally, light from an ordinary source (i.e. a lamp) travels in all planes, so when light falls upon a person's eye, they see light in all planes. Plane polarized light is light which consists of only one plane, and is achievable by passing ordinary light through a Polaroid filter. In a polarimeter, ordinary light from a source is passed through two Polaroid filters; the first filter creates plane polarized light, then the second filter is placed at a right angle to the first, so that the plane polarized light cannot pass through and no light can be seen. This is known as the extinction point of the light - and will subsequently change when the plane polarized light is passed through an optically active substance, as seen in *Figure 1.4* on the next page. A tube of the optically active substance is placed in the polarimeter so that light travels through the first polarimeter, becomes plane polarized, and then is rotated by the optically active substance, before the new extinction point is found, relative to the initial 90°.

<sup>13</sup> Murray, Peter R.S. p286

<sup>&</sup>lt;sup>14</sup> Giancoli, Douglas C. 1980. <u>Physics: Principles with Applications</u>. Chapter 23: Electromagnetic Waves. Pearson Education, Inc. (Sixth Edition)

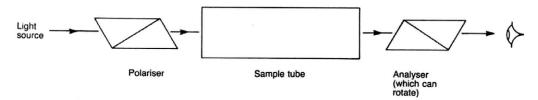


Figure 1.4 A schematic representation of a polarimeter15.

In order to find the specific rotation of a substance in solution, we must measure the observed rotation with a polarimeter, the length of the polarimeter tube and the concentration of the solution in grams per cm<sup>3</sup> of water. The equation below can be used to find the specific rotation  $[\alpha]^{T_{\lambda}}$  (Specific rotation is defined as "[the] rotation produced by a solution of length 10 centimetres and unit concentration ... for the given wavelength of light at the given temperature"<sup>16</sup>):

$$\left[\alpha\right]_{\lambda}^{T} = \frac{\Theta}{cL}$$

Equation 1 The specific rotation of a substance

Where  $[\alpha]$  = the specific rotation of the substance (°).

T =the Temperature of the solution (°C).

 $\lambda$  = the wavelength of light passing through solution (nm)

 $\Theta$  = the observed rotation of the solution (°).

L = the path length of the light traveling through the solution (dm).

c = the concentration of the solution (gcm<sup>-3</sup>).

Although the specific rotation is not calculated for either monosaccharide in solution, it is useful and relevant because of the above equation which allows us to re-arrange it and hypothesise a relationship between concentration (c) and observed rotation ( $\Theta$ ). *Figure 1.4* below shows a schematic representation of a polarimeter, consisting of a monochromatic light source, two Polaroid filters and a tube of the optically active solution.

<sup>&</sup>lt;sup>15</sup> Matthews, Philip. 1992. <u>Advanced Chemistry: Organic and Inorganic</u>. Chapter 110: Optical Activity. Cambridge University Press.

Murray, Peter R.S. 1972. <u>Principles of Organic Chemistry</u>. Chapter 4: Naming Organic Compounds. Heinemann Educational Books.

## **HYPOTHESIS**

Since optical rotation is due to the spatial arrangement of chiral molecules which rotate the plane of polarized light, the magnitude of observed optical rotation should increase proportionately with an increase in concentration of the optically active solution. This can be shown in <u>Equation 1</u> below:

$$\left[\alpha\right]_{\lambda}^{T} = \frac{\Theta}{cL}$$

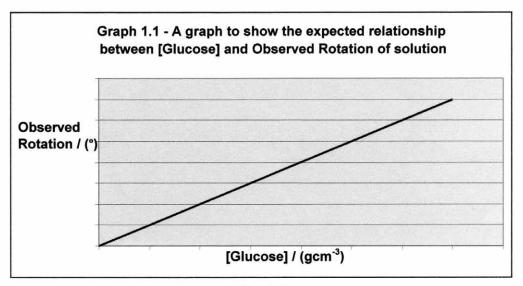
Equation 1 The specific rotation of a substance

Rearranging the formula we can obtain *Equation 2* as shown below:

$$\Theta = \alpha cl.$$
Equation 2 The observed rotation of a substance

Thus, while keeping the length of the solution and specific rotation of the solution constant,  $\Theta$  = kc where k is a constant.

Thus, it should be the case that the following graph will be obtained when observed rotation  $(\Theta)$  is plotted on the y-axis against the concentration of glucose on the x-axis:



Graph 1.1

Additionally, the specific rotation may be calculated by dividing the gradient of the straight line by L, the path length (in dm) of the light traveling through the solution. Obviously, a similar graph can be drawn with the concentration of fructose on the x-axis, and these are the expected results for the individual monosaccharides in solution.

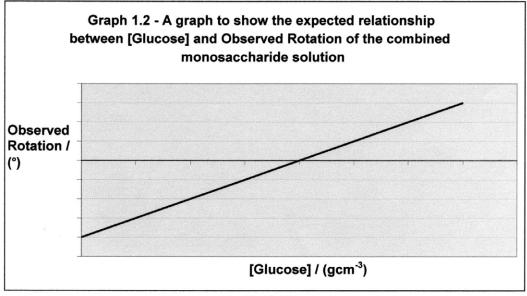
However, in order to investigate the Research Question, a combination of two different monosaccharide solutions will need to be used. The two monosaccharides used will be (+) - Glucose and (-) - Fructose. This is because they are two of the most naturally abundant monosaccharides, so I thought that they should be studied. In order to investigate the effect of combining both solutions on the resulting optical rotation, solutions of different concentrations for both monosaccharides must be made up. The molar concentrations used will be 0.10moldm<sup>-3</sup>, 0.20moldm<sup>-3</sup>, 0.30moldm<sup>-3</sup>, 0.40moldm<sup>-3</sup> and 0.50moldm<sup>-3</sup>. Since the units for concentration required for calculation of specific rotation are gcm<sup>-3</sup>, conversions were made so that the correct mass of monosaccharide could be weighed out. An example of such a calculation is shown below in *Table 1.1*, the rest can be found in *Table 2.1* in Appendix I: Raw Data on page 21.

<u>Table 1.1</u> A Table of results to show how the mass of monosaccharide per 100cm<sup>-3</sup> of water was calculated.

| 1 | Concentration of<br>Monosaccharide<br>Solution / (moldm <sup>-3</sup> ) | Molar Mass of C <sub>6</sub> H <sub>12</sub> O <sub>6</sub> / (gmol <sup>-1</sup> ) | Concentration of<br>Monosaccharide<br>Solution / (gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Mass of<br>Monosaccharide in<br>100cm of water / (g)<br>±0.001g |
|---|---|---|--|---|
|   | 0.1000  | 180.18  | .01802   | 1.802   |

Firstly, the 0.10moldm<sup>-3</sup> solution of (+) - Glucose will be mixed with the 0.50moldm<sup>-3</sup> solution of (-) - Fructose. Then the 0.20moldm<sup>-3</sup> solution of (+) - Glucose will be mixed with the 0.40moldm<sup>-3</sup> solution of (-) - Fructose and so on. The results of mixing the monosaccharides should yield a linear relationship, because when a more concentrated solution of one sugar is added to a less concentration of the other, the prevalent rotation will be similar to that of the more concentrated solution. Thus, the following graphical relationships should be found:

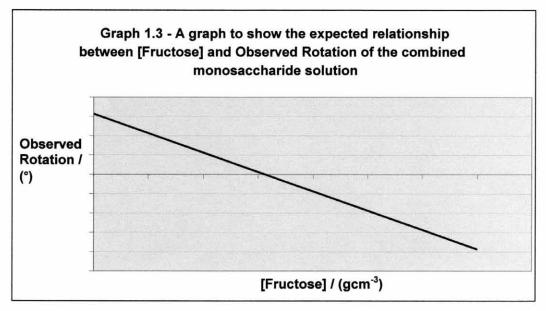
#### 1) When plotting overall observed rotation against concentration of (+) - Glucose:



Graph 1.2

Here it can be seen as the concentration Glucose increases, the observed rotation becomes more positive (as (+) - Glucose is *dextrorotary*). When the concentration of (+) - Glucose = 0gcm<sup>-3</sup>, the observed rotation should be exactly the same as the observed rotation of (-) - Fructose at the corresponding concentration since no optically active (+) - Glucose molecules are present in the solution to cause an observed rotation in the clockwise direction.

2) When plotting overall rotation against concentration of (-) - Fructose:



Graph 1.3

*Graphs 1.1, 1.2* and *1.3* are all hypothesized and are to be supported by the results of this experiment.

## **APPARATUS**

The following apparatus was used in this experiment.

## Equipment and glassware

- (10) Volumetric flasks (100± 0.08cm<sup>3</sup>)
- (10) Volumetric flask stoppers
- (2) Glass Stirring Rods
- (2) Glass funnels
- (2) Beakers  $(50 \pm 5 \text{cm}^3)$
- (2) Measuring cylinders (10± 0.1cm³)
- (2) Spatulas
- (1) Thermometer ( $-10^{\circ}$ C to  $100^{\circ}$ C  $\pm 0.5^{\circ}$ C)
- (1) Polarimeter (0°-360°±0.5°) containing the following pieces of equipment:
  - (2) Polaroid filters
  - (1) Yellow light filter
  - (1) Glass Polarimeter tube (14cm in length)
- (2) Wire connectors
- (1) Power pack (0-12V)

#### Chemicals

(+) - Glucose

Specific Rotation: 111° to 52.5° @20°C

Impurities: 0.3% H<sub>2</sub>O

(-) - Fructose

Specific Rotation: -90° to -92° @ 20°C

Impurities: 0.3% H<sub>2</sub>O Distilled Water

## **METHOD**

Measuring out accurate concentrations of monosaccharide solution.

- 1) Using the mass top balance, measure the mass of the glass container, record this mass and re-zero the scale.
- 2) Accurately weigh out 1.802g of the crystallized (+) Glucose
- 3) Transfer the (+) Glucose to a 100cm<sup>3</sup> beaker and wash out any residual sugar into the beaker using distilled water.
- 4) Dissolve the sugar in the beaker using approximately 80cm<sup>3</sup> of distilled water.
- 5) Using a funnel, transfer the dissolved sugar solution into a 100cm<sup>3</sup> volumetric flask.
- 6) Wash out residue from beaker into the volumetric flask and make up to 100cm3.
- 7) Repeat the above steps for the four other concentrations of (+) Glucose and for each of the concentrations for (-) Fructose.

Measuring the observed rotation of sugar solutions using a polarimeter.

- 1) Measure 14cm<sup>3</sup> of distilled water using the 10cm<sup>3</sup> measuring cylinder (twice) and pour into the glass polarimeter tube.
- Connect polarimeter to power pack and attach glass tube, then switch the power pack on to 6V.
- 3) Record the extinction point of water (the point where the least light can be seen through the polarimeter eyepiece.)
- 4) Remove glass polarimeter tube and pour out distilled water.
- 5) Invert the volumetric flask containing 1.802g of (+) Glucose twice to ensure complete mixing of solution.
- 6) Pour out approximately 50cm<sup>3</sup> of solution into one of the beakers.
- 7) Measure 14cm³ of the (+) glucose solution using measuring cylinders.
- 8) Pour this solution into the glass polarimeter tube.
- 9) Insert filled glass tube into holder and insert holder into the polarimeter.
- 10) Measure the extinction point of the solution.
- 11) Repeat for all other concentrations and for (-) Fructose.
- 12) Once complete, conduct four repeat measurements of each solution.
- 13) Then measure 7cm³ of (+) Glucose (0.01802gcm-³) and 7cm³ of (-) Fructose (0.09009gcm-³) and pour both solutions into the glass polarimeter tube.
- 14) Record extinction point of mixture and repeat using other combinations of the two monosaccharides as shown in *Table 1.4* on page 13.

## RESULTS

In this section, the results of the experiment will be displayed graphically and explained through analysis of the data. The results displayed below are from the initial data showing the relationship between observed rotation and concentration of (+) - Glucose in solution. The raw data for one concentration of (+) - Glucose is shown below in <u>Table 1.2</u> and the rest of the data can also be found in <u>Table 2.2</u> in <u>Appendix I: Raw Data</u> on page 21.

Table 1.2 A table of results to show the extinction point of (+) - Glucose at 0.01802gcm<sup>-3</sup>

| Concentration of<br>(+) -Glucose /<br>(gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Maximum<br>Extinction<br>Point (1) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (2) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (3) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (4) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (5) / (°)<br>±0.5° | Average<br>Maximum<br>Extinction<br>Point <sup>17</sup> / (°)<br>±0.5° |  |
|---|---|---|---|---|---|--|--|
| 0.01802   | 354   | 353   | 354   | 353   | 354   | 354  |  |

<u>Table 1.3</u> A table of results to show the observed rotation of (+) - Glucose at different concentrations

| Concentration of (+) -Glucose / (gcm <sup>-3</sup> ) ±0.001gcm <sup>-3</sup> | Extinction Point of H <sub>2</sub> O / (°) ±0.5° | Average<br>Maximum<br>Extinction<br>Point / (°)<br>±0.5° | Observed<br>Rotation <sup>18</sup> / (°)<br>±1° |
|--|--|--|---|
| 0.01802  | 353  | 354  | 1   |
| 0.03604  | 353  | 355  | 2   |
| 0.05405  | 353  | 357  | 4   |
| 0.07207  | 353  | 358  | 5   |
| 0.09009  | 353  | 36119  | 8   |

In order to analyse the results, a graph was drawn to display the optical rotation against the concentration of (+) - Glucose. The random errors incorporated into the error bars in the above measurement are from the concentration of sugar in solution ( $\pm 0.001 \text{gcm}^{-3}$ ), the rotation measurement of the polarimeter ( $\pm 0.5^{\circ}$ ) multiplied by two since the measurement was taken for water and for the solution, and the volume of solution in the glass tube used in the polarimeter ( $\pm 0.2 \text{cm}^3$ ). The y-axis error for each data point was found to be  $\pm 1^{\circ}$  and is shown on each point as y-error bars. The x-axis error for each data point was found to be  $\pm 0.001 \text{g}$  and is shown on each point as x-error bars.

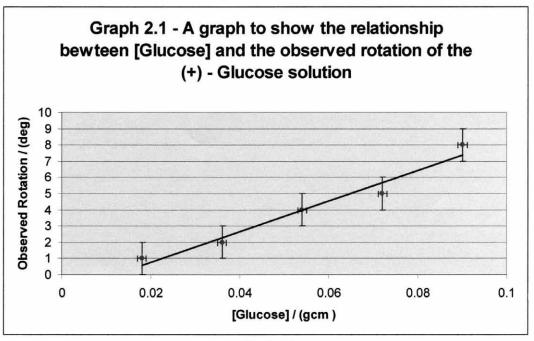
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 $<sup>^{\</sup>rm 17}$  The Average Maximum Distinction point is an average of the five repeat readings.

<sup>&</sup>lt;sup>18</sup> Observed Rotation = (Ave. Extinction Point of (+) - glucose - Extinction point of H<sub>2</sub>O)

<sup>&</sup>lt;sup>19</sup> The value as observed using the polarimeter was 1°, however for mathematical purposes is written as 361° in the table.



Graph 2.1

This graph shows a linear relationship between observed rotation and [Glucose], as hypothesized in *Graph 1.1* on page 6.

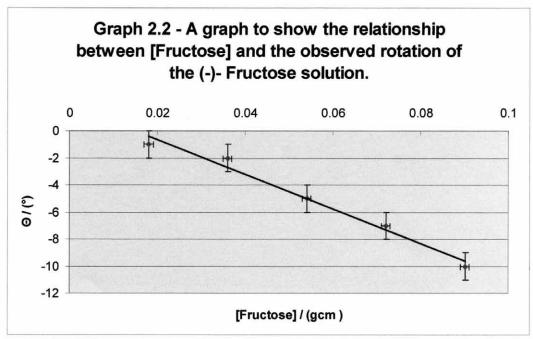
Next the same method for (-) - Fructose was used; the results are displayed in <u>Table 1.4</u> and <u>Table 1.5</u>. Again, in order to analyse the results, a graph was drawn to display the optical rotation against the concentration of (-) - Fructose.

<u>Table 1.4</u> A table of results to show the extinction point of (-) - Fructose at different concentrations.

| Concentration<br>of (-) -<br>Fructose /<br>(gcm <sup>-3</sup> ) | Maximum<br>Extinction<br>Point (1) / (°) | Maximum<br>Extinction<br>Point (2) / (°) | Maximum<br>Extinction<br>Point (3) / (°) | Maximum<br>Extinction<br>Point (4) / (°) | Maximum<br>Extinction<br>Point (5) / (°) | Average<br>Maximum<br>Extinction<br>Point / (°) |
|---|--|--|--|--|--|---|
| ±0.001gcm <sup>-3</sup>   | ±0.5°                                    | ±0.5°                                    | ±0.5°                                    | ±0.5°                                    | ±0.5°                                    | ±0.5°   |
| 0.01802   | 352                                      | 353                                      | 352                                      | 352                                      | 353                                      | 352   |
| 0.03604   | 350                                      | 349                                      | 349                                      | 349                                      | 349                                      | 349   |
| 0.05405   | 348                                      | 349                                      | 348                                      | 349                                      | 348                                      | 348   |
| 0.07207   | 347                                      | 347                                      | 346                                      | 346                                      | 346                                      | 346   |
| 0.09009   | 343                                      | 344                                      | 343                                      | 343                                      | 344                                      | 343   |

<u>Table 1.5</u> A table of results to show the observed rotation of (-) - Fructose at different concentrations

| Concentration of<br>(-) - Fructose /<br>(gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Extinction Point of H <sub>2</sub> O / (°) ±0.5° | Average<br>Maximum<br>Extinction<br>Point / (°)<br>±0.5° | Observed Rotation / (°) |
|---|--|--|-------------------------|
| 0.01802   | 353  | 352  | -1                      |
| 0.03604   | 353  | 349  | -2                      |
| 0.05405   | 353  | 348  | -5                      |
| 0.07207   | 353  | 346  | -7                      |
| 0.09009   | 353  | 343  | -10                     |

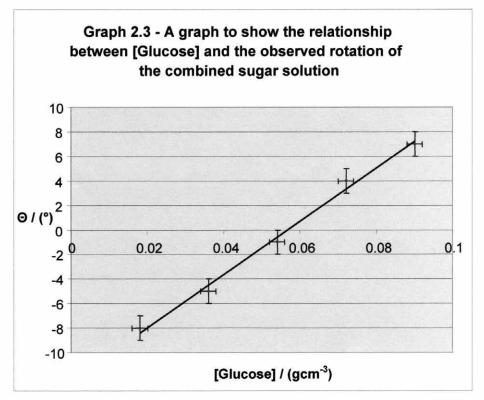


Graph 2.2

Finally, solutions of (-) - Fructose and (+) - Glucose were mixed together in different concentrations in order to establish a relationship between the resulting rotation and the concentrations of the two monosaccharides in solution. The results of this part of the experiment are shown below in <u>Table 1.6</u> and <u>Table 1.7</u> and represented graphically in <u>Graph 2.3</u> and <u>Graph 2.4</u>.

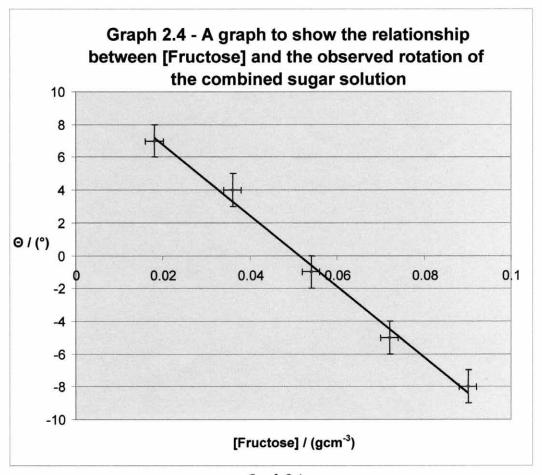
<u>Table 1.6</u> A table of results to show the observed rotation of mixtures of (-) - Fructose and (+) - Glucose at different concentrations

| Concentration<br>of Glucose /<br>(gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Concentration<br>of Fructose /<br>(gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Maximum<br>Extinction<br>Point (1) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (2) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (3) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (4) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (5) / (°)<br>±0.5° | Average<br>Maximum<br>Extinction<br>Point / (°)<br>±0.5° | Observed<br>Rotation<br>(°)<br>±0.5° |
|--|---|---|---|---|---|---|--|--------------------------------------|
| 0.01802  | 0.09009   | 345   | 344   | 345   | 345   | 345   | 345  | -8                                   |
| 0.03604  | 0.07207   | 348   | 347   | 348   | 349   | 348   | 348  | -5                                   |
| 0.05405  | 0.05405   | 352   | 353   | 352   | 352   | 352   | 352  | -1                                   |
| 0.07207  | 0.03604   | 356   | 356   | 357   | 358   | 356   | 357  | 4                                    |
| 0.09009  | 0.01802   | 359   | 363   | 360   | 360   | 359   | 360  | 7                                    |



Graph 2.3

Next, a graph was drawn plotting the concentration of (-) - Fructose against the overall observed rotation of the combined sugar solution.



Graph 2.4

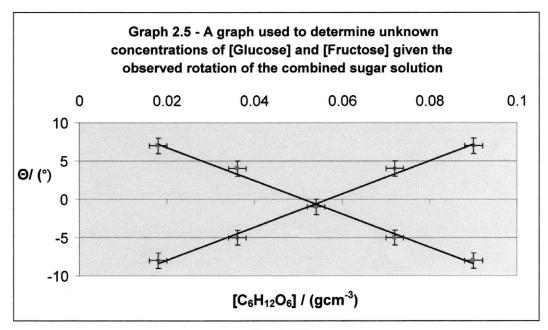
Thus, the linear relationship between the concentration of each of the constituent monosaccharides and the observed rotation of the mixture of the two can be seen and the hypothesis is supported by the experimental results.

## **CONCLUSION**

As mentioned previously, the Research Question for this essay is as follows:

When solutions of (+) - Glucose and (-) - Fructose are mixed, is there a relationship between the observed rotation of polarised light caused by this mixture and the concentration of the constituent monosaccharide solutions?

By looking at the graphs obtained in the results section of this essay, it is possible to answer the Research Question. It appears that there is indeed a relationship between the observed rotation of polarised light caused by this solution and the concentration of the constituent monosaccharide solutions. This relationship is of a linear nature as shown in *Graph 2.3* and *Graph 2.4* in the previous section. It has been shown that as the concentration of one monosaccharide increases (and thus the concentration of the other decreases), the observed rotation of the combined solution becomes more similar to that of the individual sugar solution. The implication of this linear relationship is that if given a solution containing a mixture of these two monosaccharides of unknown concentrations, it is possible due to the above linear results to identify the concentrations or concentration ratios of each constituent monosaccharide. By combining Graph 2.3 and Graph 2.4, this identification is possible and is shown in Graph 2.5 below. If the observed rotation of an unknown solution is known, the point on the line of best fit for each monosaccharide corresponding to this observed rotation can be found. From this, the x-axis value can be determined (which is measured in grams of monosaccharide per cm3) and thus the concentration of (+) - Glucose and (-) - Fructose can be identified.



Graph 2.5

#### Limitations and unresolved questions

Unfortunately, there are limitations to the implications of the results of this experiment. The experiment provides insight only into the combination of these two monosaccharides at specific concentrations. It is limited in order to answer the research question in the specified word limit and to allow an in depth analysis within the time allocated to the experiment. In order to establish a general relationship when adding together two optically active substances with different rotations, more experimental data must be collected.

#### Further Investigations

Firstly, it would be ideal to collect data on a combination of all three isomers of glucose - i.e. both fructose and galactose since they all have the same molecular mass. Furthermore, it is impossible to know whether the same linear relationship would hold if two optically active substances of different molecular mass are used, but this could be a further investigation into the topic.

Another possible further investigation in the topic of optical isomerism is to attempt to investigate the mutarotation of glucose in solution through polarimetry, a topic discussed in the Introduction on page 3. Since we know that  $\alpha$ -glucose rotates the plane of polarised light to a greater degree (+112°) than  $\beta$ -glucose (+18.7°), we can use the linear relationship obtained above to determine the concentration of each of these isomers by simply finding the observed rotation of the solution of glucose. The practical implications of this relationship are that pharmacological companies, schools and other institutes using glucose can determine how 'fresh' the glucose in solution is, allowing them to know when they need to order new glucose.

Thus, it can be seen that the essay has successfully answered the <u>Research Question</u> by investigating the optical rotation of (+) - Glucose and (-) - Fructose using a polarimeter. Some of the errors and limitations of the experiment are explained in the Evaluation on the next page.

## **EVALUATION**

As it can be seen from the variance of data of the above graphs (i.e. plotted points lying outside the maximum and minimum lines of best fit), there are systematic errors inherent in this experiment. These will be discussed in the following section.

The mutarotation of glucose possibly was occurring while doing the experiment. Since the results were recorded on a series of days, the observed rotation of glucose or fructose may have varied. The collection of data would ideally have been conducted on the same day, so as to minimise the possibility of mutarotation of glucose. If this is not possible, it would be alternatively ideal to make up the monosaccharide solutions and use them immediately rather than leave them in solution for the duration of the experiment. An alternative solution would be to prepare the solutions and leave them for a specific amount of time before conducting the experiment.

The polarimeter was the source of much of the error in this experiment. Firstly, the polarimeter was manual and only had divisions of 1 degree (which were extremely close together). Thus, if any deviation occurred while measuring the observed angle (i.e. it was not measured from directly above), the results would be rendered inaccurate. To solve this specific problem, a more accurate polarimeter, or one which records data digitally, would be ideal. This is, of course, an expensive alternative in most cases (i.e. in a basic experiment investigating optical rotation of a single substance) but would greatly improve the accuracy of the results while simultaneously minimising the errors.

Measuring the maximum extinction point by eye is a very subjective method of measurement. It is, in a sense, qualitative rather than quantitative. In order to solve this problem, it would again be ideal to use a digital polarimeter where the percentage of light transmission could be measured for a range of angles, and hence the analysis would be quantitative. This would minimise human error and thus make the measurements of the observed rotation more accurate.

Glucose is only soluble in water up until a certain point (91g per 100cm<sup>3</sup> @ 25°C)<sup>20</sup>. When the glucose is at high concentrations in water, it begins to make the solution more opaque and thus light is transmitted to a lesser extent. This renders the experiment almost impossible. However, the problem with using low concentrations of glucose is that the observed rotation of the solution was small, thus the percentage error was relatively high. A solution would be to increase the temperature of the solution, as 244g per 100cm<sup>3</sup> of glucose dissolves at 50°C. The following equation<sup>21</sup> can be used to account for this change in temperature (and also can be used to account for slight variance in laboratory temperature):

$$[\alpha]\lambda(20^{\circ}C) = [\alpha]\lambda(T)[1+0.0001(T-20)]$$

<a href="http://www.mpcfaculty.net/mark\_bishop/supersaturated.htm">http://www.mpcfaculty.net/mark\_bishop/supersaturated.htm</a> Retrieved on 2006-11-03

<sup>&</sup>lt;sup>20</sup> Supersaturated. URL:

<sup>&</sup>lt;sup>21</sup> Synthesis and Optical Resolution of Co(en)<sub>3</sub><sup>3+</sup>. URL:

<sup>&</sup>lt;a href="http://www.uiowa.edu/~c004153a/Co(en)3-2004.pdf">http://www.uiowa.edu/~c004153a/Co(en)3-2004.pdf</a>> Retrieved on 2006-11-03

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## **APPENDIX I: RAW DATA**

Table 2.1

A table of data showing the concentrations of monosaccharide solutions by mass/volume and mol/volume.

| Mass of<br>Monosaccharide in<br>100cm of water / (g)<br>±0.001g | Concentration of<br>Monosaccharide<br>Solution / (gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Molar Mass<br>of C <sub>6</sub> H <sub>12</sub> O <sub>6</sub> /<br>(gmol <sup>-1</sup> ) | Concentration of<br>Monosaccharide<br>Solution /<br>(moldm <sup>-3</sup> ) |  |
|---|--|---|--|--|
| 1.802   | .01802   | 180.18  | 0.1000   |  |
| 3.604   | .03604   | 180.18  | 0.2000   |  |
| 5.405   | .05405   | 180.18  | 0.3000   |  |
| 7.207   | .07207   | 180.18  | 0.4000   |  |
| 9.009   | .09009   | 180.18  | 0.5000   |  |

<u>Table 2.2</u>
A table of results to show the observed rotation of glucose at different solutions.

| Concentration<br>of Glucose /<br>(gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Maximum<br>Extinction<br>Point (1) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (2) / (°<br>±0.5° | Maximum Extinction Point (3) / (°) ±0.5° | Maximum Extinction Point (4) / (°) ±0.5° | Maximum Extinction Point (5) / (°) ±0.5° | Average<br>Maximum<br>Extinction<br>Point / (°)<br>±0.5° | Observed<br>Rotation /<br>(°)<br>±1° |
|--|---|--|--|--|--|--|--------------------------------------|
| .01802   | 354   | 353  | 354                                      | 353                                      | 354                                      | 354  | 1                                    |
| .03604   | 354   | 355  | 354                                      | 356                                      | 355                                      | 355  | 2                                    |
| .05405   | 358   | 358  | 356                                      | 357                                      | 357                                      | 357  | 4                                    |
| .07207   | 359   | 358  | 358                                      | 357                                      | 358                                      | 358  | 5                                    |
| .09009   | 361   | 360  | 361                                      | 361                                      | 361                                      | 361  | 8                                    |

Table 2.3

A table of results to show the observed rotation of fructose at different concentrations.

|                         | Extinction Point (1) / (°) |       |       |       | Maximum<br>Extinction<br>Point (5) / (°) |       | Extinction<br>Point of<br>H₂O / (°)<br>±0.5° | Observed<br>Rotation / (°) |
|-------------------------|----------------------------|-------|-------|-------|--|-------|--|----------------------------|
| ±0.001gcm <sup>-3</sup> | ±0.5°                      | ±0.5° | ±0.5° | ±0.5° | ±0.5°                                    | ±0.5° |  | ±1°                        |
| 0.01802                 | 352                        | 353   | 352   | 352   | 353                                      | 352   | 353  | -1                         |
| 0.03604                 | 350                        | 349   | 349   | 349   | 349                                      | 349   | 353  | -2                         |
| 0.05405                 | 348                        | 349   | 348   | 349   | 348                                      | 348   | 353  | -5                         |
| 0.07207                 | 347                        | 347   | 346   | 346   | 346                                      | 346   | 353  | -7                         |
| 0.09009                 | 343                        | 344   | 343   | 343   | 344                                      | 343   | 353  | -10                        |

<u>Table 2.4</u>
A table of results to show the observed rotation of mixtures of fructose and glucose at different concentrations

| Concentration<br>of Glucose /<br>(gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Concentration<br>of Fructose /<br>(gcm <sup>-3</sup> )<br>±0.001gcm <sup>-3</sup> | Maximum<br>Extinction<br>Point (1) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (2) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (3) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (4) / (°)<br>±0.5° | Maximum<br>Extinction<br>Point (5) / (°)<br>±0.5° |     | Observed<br>Rotation /<br>(°)<br>±0.5° |
|--|---|---|---|---|---|---|-----|--|
| 0.01802  | 0.09009   | 345   | 344   | 345   | 345   | 345   | 345 | -8                                     |
| 0.03604  | 0.07207   | 348   | 347   | 348   | 349   | 348   | 348 | -5                                     |
| 0.05405  | 0.05405   | 352   | 353   | 352   | 352   | 352   | 352 | -1                                     |
| 0.07207  | 0.03604   | 356   | 356   | 357   | 358   | 356   | 357 | 4                                      |
| 0.09009  | 0.01802   | 359   | 363   | 360   | 360   | 359   | 360 | 7                                      |