<table>
<thead>
<tr>
<th><strong>IB Diploma Programme subject in which this extended essay is registered:</strong></th>
<th><strong>Group Biology</strong></th>
</tr>
</thead>
</table>

(For an extended essay in the area of languages, state the language and whether it is group 1 or group 2.)

**Title of the extended essay:** How can respiration help in the search for extraterrestrial life?

**Candidate's declaration**

This declaration must be signed by the candidate; otherwise a grade may not be issued.

The extended essay I am submitting is my own work (apart from guidance allowed by the International Baccalaureate).

I have acknowledged each use of the words, graphics or ideas of another person, whether written, oral or visual.

I am aware that the word limit for all extended essays is 4000 words and that examiners are not required to read beyond this limit.

This is the final version of my extended essay.

Candidate's signature: ___________ Date: 03/12/15
It seems appropriate in the International Year of Soils that a student should attempt to devise their own study of extra-terrestrial soils. As a keen hobby astronomer the student has linked an interest from outside school to one of his academic higher level subjects, using age appropriate techniques and school equipment to generate some interesting data. In extending class syllabus and lab work this way, the student was hugely challenged by the difficulties of controlling variables to effect repetition of trials. His subsequent wealth of data showed some unexpected results. The difficulty of deciphering complex published material from academic studies at a tertiary level was also hugely challenging, in trying to make sense of the data found in this study.
## Assessment form (for examiner use only)

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Examiner 1 maximum</th>
<th>Examiner 2 maximum</th>
<th>Examiner 3</th>
<th>Achievements level</th>
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<td>1</td>
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<td>B introduction</td>
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<td>C investigation</td>
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Total out of 36: **18**

Name of examiner 1:  
(CAPITAL letters)  
Examiner number:  

Name of examiner 2:  
(CAPITAL letters)  
Examiner number:  

Name of examiner 3:  
(CAPITAL letters)  
Examiner number:  

IB Assessment Centre use only:  
B:  

IB Assessment Centre use only:  
A:  

Extended essay

One small step in the search for extraterrestrial life

Investigating the affect of varying sugar type and concentration on respiration in Clay soil

Word Count: 3927
Abstract
This extended essay investigates which sugar out of glucose, fructose and sucrose is most effective in increasing the rate of respiration in a clay soil sample and at which concentration, 12, 8 or 4 grams per litre. The topic was chosen as it helps in the search for extra terrestrial life, in that it could allow for the accelerated growth of extra-terrestrial life, assuming they are in the form of micro-organisms, and easier study. It led to the research question: “How can respiration help in the search for extraterrestrial life?”

The experiment were conducted in a school laboratory, with the blinds closed and each sugar was tested separately to begin with varying the concentration from 12, 8 and 4 grams per litre while leaving one soil sample as a control, only adding water. Concentrations were made by mixing 0.12, 0.08 and 0.04 grams with 10 ml of distilled water. The results were recorded as carbon dioxide production (PPM) on the computer software logger pro using carbon dioxide probes. Logger pro produced a graph which was analysed as well as calculation of the rate of carbon dioxide production (referred to as the rate of respiration) to help with further analysis.

The results attained corresponded with the initial hypothesis as there was a increase in carbon dioxide production as the concentration of glucose was increased, shown by the increase in rate in trial one from 4g/l to 12g/l as 406.9233 to 953.9046 PPM per hour. This raised the question of whether other sugars may be more effective than glucose at increasing the rate of respiration, which led to further experimentation of sucrose and fructose at the same concentrations. These results showed glucose to be most effective at 12 g/l and sucrose and 4 g/l which means these would be the two most effective sugars, depending on the length of the trip, to be taken to accelerate the growth of extra terrestrial life.

Word Count: 320
Introduction
Since the beginning of civilization, man has looked to the heavens and asked “are we alone in the universe?” Early research was limited to optical observations and the search for “little green men” but once space exploration became a reality the search intensified on a more scientific basis, so far however with little success. The modern search for life is focused on the building blocks of life in the form of micro organisms and fossil records. Soil and rock samples are very difficult to study not only because they are so far from Earth but also because they would be of limited size and, most likely, lack the conditions and nutrients for growth. However, if it were possible to promote growth of any organisms discovered by providing additional nutrients, they would become far easier to study. Micro-organisms discovered on far off planets wouldn’t survive the journey back to Earth and samples could even be contaminated by the very machines collecting them. The answer, if it were possible, is to promote growth within the organism while keeping it in its natural conditions. Analysis would become far more accurate and much easier. Robots could be sent to remote planets, carrying nutrients required to promote growth, performing analysis and reporting data, in the form of gas analyses, back to Earth bound scientists. Current NASA Mars exploration is conducted in a similar manner using the Curiosity probe. Mass spectrometers are employed to analyse gases produced by reactions in soil and rock samples. This technique has to be treated with caution as material from Earth, no matter how rigorously screened, is involved and could lead to imperfect results. Following this process “outer planet farms” could be established where any newly discovered micro-organisms are grown for analysis.

Let us start by considering which processes within organisms are responsible for growth, i.e cell division. Although many processes are involved I will be focusing on respiration as it is an important pathway for production of energy and can be easily studied and measured using simple laboratory methods.
This leads to my research question: How can respiration help in the search for extraterrestrial life?

Cellular Respiration is defined as:” A series of metabolic processes that take place within a cell in which biochemical energy is harvested from organic substance (e.g. glucose) and stored as energy carriers (ATP) for use in energy-requiring activities of the cell.” (Biologyonline)

It occurs within all living organisms, and can be written out in simple terms as:

\[ C_6H_{12}O_6 \text{(sugar)} + O_2 \rightarrow CO_2 + H_2O + \text{Energy}. \]

Increase respiration and the organism will produce more energy in the form of ATP (Adenosine triphosphate) and potentially allow for increased cell division as there is more available energy for use by the cell. For the purpose of my experimentation I must find an organism which uses anaerobic or aerobic respiration.

In order to investigate respiration an organism with some relevance to other planets is needed. I chose soil as it contains various micro-organism such as bacteria, protozoa and Fungi, which I am assuming are most likely to be found on other worlds. The terrestrial soil I chose did not contain any arthropods or nematodes as these would most likely not be present in extra-terrestrial samples.

Therefore I need to focus on the respiration of Bacteria, protozoa and Fungi. Bacteria and fungi can respire both anaerobically and aerobically while Protozoa respire mostly aerobically.

Both respiration pathways start with glycolysis (figure 7). Glyclosis occurs in 4 steps:

1. Phosphorylation: Where a hexose sugar (e.g. glucose) becomes phosphorylated by two ATP molecules and turned into Hexose biphosphate.
2. Lysis: Hexose Biphosphate splits into two triose phosphates (3 carbon sugars)

3. Oxidation: Triose phosphates are oxidised, losing hydrogen and NAD is reduced into NADH+H⁺

4. ATP formation: Four ATP molecules are released as the triose phosphates are converted into pyruvate.

Products: 2 pyruvate, 2 (NADH+ H⁺) and 2 ATP (per glucose molecule)

(source Bio ninja)

Anaerobic respiration in bacteria and Fungi, also known as fermentation, occurs in the cytoplasm where pyruvate from glycolysis is converted into ethanol and carbon dioxide with the bio product of NAD⁺. The NAD⁺ is used in glycolysis while the Ethanol is excreted as the cell cannot use its energy.

Aerobic respiration in protozoa takes place in the mitochondria and is initiated by pyruvate, it can be split into three steps:

1. Link reaction:
   - Pyruvate is transported to the mitochondrial matrix.
   - In a reaction where NAD⁺ is oxidised to produce NADH+H⁺ and the pyruvate loses a carbon in the form of CO₂.
   - The other two remaining carbons are formed with Coenzyme A to form acetyl CoA

2. The Krebs cycle:
   - In the mitochondrial matrix acetyl CoA combines with a 4 carbon compound to create a 6 carbon compound.
   - The 6 carbon compound is broken down into the original 4 carbon compound by a series of reactions, which result in the formation of 2 CO₂ molecules, 1 ATP, 3 (NADH+ H⁺) and 1 FADH₂.

3. The electron transport chain:
• (NADH+ \( H^+ \)) and FADH\(_2\) provide the electron transport chain, a chain of membrane proteins, in the inner mitochondrial membrane with electrons.
• Electrons pass along the chain losing energy at each step, which is used to pump \( H^+ \) ions from the inner mitochondrial membrane to the intermembrane space, creating a gradient
• Hydrogen Ions return to the matrix through the channel protein ATP synthase, which produces ATP molecules via chemiosmosis.
• Oxygen is the final electron acceptor in the chain, it reacts electrons with \( H^+ \) ions to form water.

(Source Bio Ninja)

After looking at the two processes of respiration I noticed that both start with glycolysis and more specifically a 6 carbon sugar. Glucose seemed to be a perfect option to use in experimentation as it is available at the school lab and its amount can easily be varied by varying mass.

Having selected the material and pathway to study, I can develop my research question too “How does varying the concentration of glucose affect the rate of respiration in soil and could this help in the search for extra-terrestrial life?”

In order to answer the research question I had to set up an experiment.

The Design
The first step was to collect my soil sample. I chose a clay soil type from my back garden, . It is important that I use the same soil type for each experiment. Then I had to collect all the equipment and sugar I would need to conduct my experiment. Fortunately, my school were willing and able to provide the hardware, I used Logger pro for data collection.
I that by increasing the availability of glucose the rate of respiration will increase, as the availability of glucose for glycolysis will be greater allowing the process of both aerobic and anaerobic to take place more quickly in the micro-organism in the soil.

The apparatus used is listed below:

- 4 carbon dioxide probes, includes 4 probe boxes
- Computer with measuring software (logger pro)
- Distilled water
- Glucose (crystal form)
- Sucrose (crystal form)
- Fructose (crystal form)
- Clay soil (50g per probe)
- Spatula
- Pipette

I conducted an initial experiment to make sure I could collect meaningful data from my soil sample and also to establish the experimental routine and check the data collection system. I also had to decide on sugar concentrations. I started with a test experiment in order to see how adding a 4g per litre sugar solution (mixed with distilled water) affected the Carbon dioxide production in comparison to only adding distilled water (control). I found a significant increase in the Carbon dioxide recorded within the probe flask. The amount of carbon dioxide produced over time represents the rate of respiration of the soil.

Based on this initial trial, I decided to focus on three different sugar concentrations: 4, 8 and 12 grams per litre.

Since there are a lot of variables that can influence the results they were controlled as follows:
<table>
<thead>
<tr>
<th>Variable</th>
<th>1. light intensity</th>
<th>2. temperature</th>
<th>3. carbon dioxide concentration</th>
<th>4. Probe Used</th>
</tr>
</thead>
<tbody>
<tr>
<td>control</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1. I will keep the experiment at the same physical location in the laboratory with the blinds closed, to maintain the same light exposure for each test.

2. Heat sources will be kept away from the experiment which will be performed kept at room temperature.

3. The concentration of carbon dioxide in the room will be left at a standard level. Note: this is hard to control.

4. I will use the same probes for each test, in order to minimize error.
<table>
<thead>
<tr>
<th>Dependent</th>
<th>Sugar concentration or sugar type</th>
<th>Sugar solutions will be applied at 4, 8 and 12 grams per litre. Or Use three sugars, glucose, sucrose and fructose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Independent</td>
<td>Carbon dioxide present in the probe chamber. This will be my measure of respiration.</td>
<td>Use a CO₂ probe inserted in the probe chamber to monitor the change in carbon dioxide over 12 hours. Data collected on a computer using Logger Pro software.</td>
</tr>
</tbody>
</table>

See method in appendix, Figure 10.

It’s important to note that although most factors have been fully controlled it is impossible to control some minor factors since the probes are left at the back of a class room, if people open the blinds or breath over the probes light intensity and carbon dioxide concentration may be slightly affected. However since the probe will be recording for 12 hours hopefully any small anomalies won’t be significant.
Results

**Investigating the relationship between concentration of Glucose in a solution added to the soil and Carbon dioxide production in soil**

The graph shows a simple trend in an increase of carbon dioxide present in the probe container as the concentration of glucose increases. There is a plateau point at 9930 PPM (±5) which both 12 and 8 grams per litre concentrations reached, which represents the maximum limit of detection of the probe. The lowest concentration of glucose used, 4 grams per litre (g/L) did not reach this value as the rate of increase in carbon dioxide present was not high enough to reach the plateau value instead ending at 8520 PPM. This suggests that the lowest concentration produced the lowest rate of respiration. In addition it is notable that there was a greater increase in rate of respiration between 4 and 8 g/L than between 8 and 12 g/L which might suggest that any further increase in concentration above 12 g/L would produce an even lower increase in rate. This could suggest that there is a maximum concentration after which any increase would not evoke a further change in the rate of respiration.
Trial 2 shows a similar trend to trial 1 whereby the amount of carbon dioxide produced increased with increasing glucose concentration. 12 g/L plateaus just before 10 hours at around 9900 PPM (±50) while 8 g/L reached this plateau just after 15 hours. 4g/L did not reach a plateau and instead reached its maximum point after 15 hours of around 7600 (±50) then the carbon dioxide production began to fall.
The third trial showed the same concentration dependent effect on CO₂ production. Note that the first two graphs show CO₂ production over 24 hours while the last one does not. This occurred as I changed my method to reduce the recording time to 12 hours. I did this as the maximum values were recorded within 10-15 hours in the first experiments and the lower concentration was beginning to record reduced CO₂ values after 16 hours. It also allowed me to add more trials to my experiment as it reduced the required time.

In addition to the graphs I calculated rate of respiration (considering all CO₂ production came from the soil) by using the following formula.

\[
\text{Rate of respiration: } \frac{\text{Maximum CO}_2 \text{ level} - \text{Starting CO}_2 \text{ level}}{\text{Time to reach max point (h)}}
\]

<table>
<thead>
<tr>
<th>concentration</th>
<th>Rate of respiration (PPM/hour) (±100 ppm):</th>
</tr>
</thead>
<tbody>
<tr>
<td>12 g/L</td>
<td>953</td>
</tr>
<tr>
<td>8 g/L</td>
<td>605</td>
</tr>
<tr>
<td>4 g/L</td>
<td>406</td>
</tr>
</tbody>
</table>
The higher the concentration of glucose in the solution the higher was the rate of respiration, this is due to the use of glucose in glycolysis the first stage of respiration which occurs in both anaerobic and aerobic respiration and therefore impacts all the micro-organisms in the soil. Since glucose is the starting molecule of the whole anaerobic respiration process the higher the concentration the more glucose is available to be broken down and turned into pyruvate hence firstly increasing the rate at which carbon dioxide is produced from the link reaction and secondly increasing the production of pyruvate produced which will in turn increase the rate of the links reaction and therefore the whole aerobic respiration cycle, which causes the increase in CO\textsubscript{2} production.

The same applies for anaerobic respiration used by bacteria and fungi, more pyruvate will be produced due to higher glucose concentration causing more to be broken down into ethanol and carbon dioxide, hence increasing the CO\textsubscript{2} production (or the rate of respiration).

Due to the success of my first set of experiments I decided to further inquire into my research question. As of yet I have only investigated glucose. However to find the most effective way of increasing respiration in micro-organisms it's necessary to investigate other sugars as well. I chose two other common sugars, fructose and sucrose. I thought sucrose would be an interesting sugar to investigate as it is a disaccharide consisting of 50% glucose and 50% fructose, while fructose has a different structure to that of glucose. See figure 6.

I investigated the respiration of fructose and sucrose. Fructose enters glycolysis through 2 different pathways, either as Fructose-6-phosphate just before step 1 phosphorylation or as Fructose-1-phosphate before step 3, oxidation. See figure 8.

Disaccharide Sucrose is broken down into Fructose and glucose, by the enzyme invertase (in bacteria), which are used in glycolysis as mentioned above. In order to discover the effects of both sugars on respiration I need to set up another experiment.
**Further Research**

I reproduced my experiment for varying glucose concentrations however instead of using glucose I either used fructose or sucrose.

**Sucrose**

**The effect of varying sucrose concentration on carbon dioxide production in soil**

The overall trend in carbon dioxide production when sucrose concentration was varied is shown in the graph below, the other two trials can be seen in the appendix, see figure 1 and 2.

**Trial 2:**

![Sucrose Concentrations](image)

Note: no control was set for this experiment as the probe miss functioned.

I calculated the rate of reaction using the same formula used previously.

<table>
<thead>
<tr>
<th>concentration</th>
<th>Rate of respiration (PPM/hour) (±100 ppm):</th>
</tr>
</thead>
<tbody>
<tr>
<td>12 g/L</td>
<td>451</td>
</tr>
<tr>
<td>8 g/L</td>
<td>953</td>
</tr>
<tr>
<td>4 g/L</td>
<td>1040</td>
</tr>
</tbody>
</table>

For rates of reaction of other trials see appendix figure 11.
This experiment showed different results to those of glucose. All concentrations of sucrose stimulated carbon dioxide production, but there was a less clear and consistent concentration dependent effect.

**Fructose**

**The effect of varying fructose concentration on Carbon dioxide production of soil**

The following graph reflects similar results to the other two trials and therefore I will use it to discuss the general trend. See appendix for other trials, figure (3,4).

![Graph showing CO2 production over time with different fructose concentrations.](image)

Fructose does not follow the same pattern as glucose as its most effective concentration was 8g/L in all trials, with 4g/L and 12g/L showing roughly similar effects. This may be related to the fact that fructose follows a slightly different pathway than glucose when used in glycolysis, (see figure 8)

Trial 3:
Concentration Rate of respiration (PPM/hour) (±100 ppm)

<table>
<thead>
<tr>
<th>Concentration</th>
<th>Rate of respiration (PPM/hour) (±100 ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>12 g/L</td>
<td>378</td>
</tr>
<tr>
<td>8 g/L</td>
<td>536</td>
</tr>
<tr>
<td>4 g/L</td>
<td>353</td>
</tr>
<tr>
<td>control</td>
<td>41</td>
</tr>
</tbody>
</table>

Other rates of reaction are found in the appendix 11.

The next step in answering my research question is to find out which sugars are optimal at which concentrations, I will be doing so by following the same process as the previous experiment however instead of varying concentration I will vary sugar type.

**Varying Sugar Type**

**12g/l**

The graph shows a significant variation in respiration rates between the three sugars each in a solution of 12 g/L of water added to the soil. Glucose has the highest rate reaching its plateau value of around 9900 PPM after approximately 7 and a half hours whereas fructose and sucrose failed to reach the plateau values after 12 hours. Sucrose reaches 5699 PPM after 12 hours and fructose reached 9196 PPM after 12 hours. At 12 g/l glucose is the most effective.
sugar, this may be down to its more direct path in glycolysis compared to the two other sugars.

Note: Plateau occurs when the probe reaches its maximum recording value.

**8g/L**

Other rates of reaction are found in the appendix 11.

Rates of reaction are found in the appendix, figure 11.

This graph shows a similar trend to the 12 g/L graph as glucose produces the highest rate of respiration followed by sucrose and fructose. However this time the differences in the rate of respiration have fallen, glucose still plateaus after 8 hours. However Fructose plateaus after 9 hours followed by sucrose which plateaus after 10 hours all at the same PPM value of 9900. However it is important to note the lines are closer together than on the previous graph, this means the rates of reaction are getting closer to the same value. Therefore it seems as the
concentration falls the rates of reaction are getting closer together, sucrose is becoming more effective while glucose is falling for its rate of reaction.

This graph shows an opposite trend than the two previous graphs, sucrose produced the highest rate of respiration followed by fructose and glucose. Sucrose reached the plateau level of approximately 9900 PPM after 9 hours followed by fructose after 12 hours, however glucose did not reach the plateau level after the 12 hours. It finished at 8521 PPM.

Note: trial 2 can be viewed in the appendix, figure 5. As well as rates of reaction (figure 11)

These graphs show each sugars varying effectiveness at each concentration. Glucose is most effective at both 1.2 and 0.8 g/L however sucrose is the most effective at 0.4g/L. Fructose produces its highest rate of respiration at 0.8 g/L. The first graph clearly shows 12g/L glucose to be causing the highest rate of respiration followed by fructose and sucrose far behind, but at the 8g/L concentration the difference in the rates of respiration produced by three sugars was reduced. At the lowest concentration (4 g/L) sucrose produced the highest rate of respiration and glucose produced the lowest.

This trend occurs due to the use of glucose in both anaerobic and aerobic respiration of organisms in the soil, where the higher the glucose concentration the more can be used in glycolysis to be converted into pyruvate which will increase the overall rate of respiration.
Although my results do not show it, I would predict that at a higher concentration the rate of respiration will fall as more glucose is added due to saturation of the soil. When anymore glucose is added after that value it would just begin to act as an inhibitor to respiration.

Fructose seems to produce the highest rate at the middle concentration (0.8 g/L).

Sucrose is most effective at increasing respiration at the lowest concentration (0.4 g/L). Sucrose is a 50/50 mix of glucose and fructose and therefore I assume it is chemically more difficult to break down for use in glycolysis, therefore at the highest concentration (1.2 g/L) sucrose was least effective as the concentration was too high as all the sucrose could not be broken down quickly enough for use in glycolysis causing a lower rate of respiration. However at the lowest concentration the sugar was fully broken down and used in glycolysis causing the rate of respiration to be higher, in comparison to glucose which all gets used up quickly due to the low concentration and therefore causes a slower rate of respiration. I predict that sucrose will have an optimal concentration between 0.4 g/L and 0.8 g/L where there isn’t too much to sucrose to reduce rate due to the sugar being broken down and there being too little sugar so that it all get used up. This is a topic for possible further investigation.

**Conclusion**

Overall the experimental methods used produced results which showed trends despite variability in the results for some sugars. There are some factors which were impossible to control, this included controlling which organism are in the soil. Although a control experiment was used for each test each soil sample may have had different species or volume of organisms. This would have had some effect on the tests. In addition the reliability of data could have been improved by performing additional trials in order to generate average and standard deviation values. Nevertheless, the data do show clear trends e.g the effect of glucose was consistently concentration dependent.

In addition it’s important to note that each sugar greatly increased the rate of carbon dioxide production, used as a measure of respiration, when compared to soil alone. This shows the experiments were effective as it has allowed me to discover how to increase respiration in soil organisms. This method and approach to studying respiration could therefore be of use in the exploration of other planets and search for life. However the most effective sugar has not been clearly shown as the effectiveness of the sugar depends on the circumstances, for example at high concentrations glucose is the most effective but at lower concentrations sucrose is most
effective. This point may be an important discovery when considering how to use sugars to increase growth in extra terrestrial organisms.

In conclusion, to answer my research question, understanding of respiration can be used in the search for extra-terrestrial life by potentially increasing growth of micro organism by the introduction of sugars. My research would suggest that Glucose and sucrose were the two most effective sugars to be taken to other planets in order increase growth of extra terrestrial life, assuming life is found in the form of micro-organisms I have been investigating. Glucose was most effective in short periods of time at high concentrations which is useful for more urgent growth of organisms, however it is used up quickly therefore may not be the best sugar for sustained growth of extra-terrestrial life. Sucrose is more effective over a longer time period, as at a higher concentration (12 g/l) it produced the slowest rate of respiration therefore I used up less quickly then glucose and potentially more effective for longer growth projects. Fructose was the least effective sugar and I would not recommend taking it on a project aimed to grow extra-terrestrial life as it produced the slowest rate of respiration in most of the tests.

Therefore I would recommended Sucrose and glucose to be taken on board space ships to other planets for the project of growing extra-terrestrial life. I would recommend sucrose for longer journeys as it is broken down more slowly than glucose therefore at the same concentration sucrose will last longer than glucose, which could be important when considering how much sugar can be carried by a space craft. In addition the amount of water which can be taken is also limited therefore a longer lasting sugar would be more effective as lest water would have to be used meaning less fuel would be required and the space craft would weigh less, which would reduce the cost of the project. However for shorter trips, where growth is need quickly glucose would be recommended as at higher concentration it produces the highest rate of respiration (see figure 11) over a shorter period of time.
Bibliography

Appendices

Results: (see excel sheet)

Figure 1: Varying sucrose concentration

![Graph showing varying sucrose concentration](image_url)
Figure 2: Varying sucrose concentration

Trial 3

CO2 Production (ppm)

Time (h)

0 5 10

0 2000 4000 6000 8000 10000

control
4 g/l
8 g/l
12 g/l
Figure 3: Varying Fructose Concentration

![Fructose Concentrations](image)

Figure 4: Varying fructose concentration

![Trial 2](image)

Figure 5: Varying sugar type 4 g/l
Figure 6: Structure of Sugars

```
glucose

\[
\begin{align*}
  & \text{CH}_2\text{OH} \\
  & \text{H} \\
  & \text{OH} \\
  & \text{H} \\
  & \text{H} \\
  & \text{H} \\
\end{align*}
\]

fructose

\[
\begin{align*}
  & \text{HOC}-\text{CH}_2\text{OH} \\
  & \text{OH} \\
  & \text{H} \\
  & \text{H} \\
  & \text{OH} \\
  & \text{H} \\
\end{align*}
\]

sucrose

```

Figure 7: Glycolysis

```

Glucose

\[
\text{Phosphorylation} \quad 2 \text{ ATP} \\
\rightarrow \quad 2 \text{ (ADP + Pi)}
\]

Hexose Biphosphate

\[
\text{Lysis} \quad 2 \text{ x Tissue Phosphate}
\]

Oxidation

\[
\text{ATP Formation} \quad 2 \text{ NAD} \\
\rightarrow \quad 2 \text{ (NADH + H)} \\
\rightarrow \quad 4 \text{ (ADP + Pi)} \\
\rightarrow \quad 4 \text{ ATP}
\]

\[
\text{2 x Pyruvate}
\]

```

4 g/l (Trial 2)

![Graph showing CO₂ production over time with different sugars: control, sucrose, fructose, glucose.](image)
Figure 8: Metabolism of common monosaccharides and some biochemical reactions
Figure 9: Making solutions

4 g/l: 0.04 g of sugar, mixed with 10 ml of distilled water

8 g/l: 0.08 g of sugar mixed with 10 ml of distilled water

12 g/l: 0.12 g of sugar mixed with 10 ml of distilled water

Measured out mass in either a glass flask or a small plastic container.

Figure 10: Method
1. Gather equipment

2. Weigh out 45 g of soil and place in probe box.

3. Place a glass 10ml flask onto the scales, zero the scales, and add exactly 0.04g of sugar. Use a spatula to help with accuracy.

4. Measure out 10 ml of distilled water using a measuring cylinder; add it to the glass flask. Place a stopper on the top and shake to help the sugar dissolve.

5. Pour out the solution into the probe box. Label the probe box with the concentration and sugar name.

6. Place the carbon dioxide probe in the top of the box, and connect to the computer (see diagram) It is important that the probe is pushed tightly into the flask to prevent contamination of the experiment.

7. Open logger pro on the computer and once the probe is registered calibrate it. (by pressing the red button on the side of the probe using a pipette)

8. Repeat steps 2-7 for 0.08g and 0.12g of the same sugar, remember to also keep one probe for a control test. Or when varying the sugar type repeat steps 2-7 using the different sugars but still keeping a control of only distilled water and using the same concentration. (Note: pour the solutions into the soil at roughly the same time for the 4 different concentrations or sugar types)

9. 4 probes can be set up per computer, once all have been set up change the timer duration on logger pro to record 60 data points per hour for the experiment duration of 12 hours.

10. Press collect and leave for the given time. Make sure not to change the conditions of the experiment which might affect the rate of respiration for example temperature, light intensity, carbon dioxide concentration.

11. When recording data, note that carbon dioxide production is considered to come solely from the soil, therefore it is used to calculate rates of respiration.

Figure 11: Rates of reaction
rates of reaction (PPM/ hour)
(±100 ppm):

<table>
<thead>
<tr>
<th>Glucose</th>
<th>control</th>
<th>4 g/l</th>
<th>8 g/l</th>
<th>12 g/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>trial 1</td>
<td>104.0904</td>
<td>406.9233</td>
<td>605.517</td>
<td>953.9046</td>
</tr>
<tr>
<td>trial 2</td>
<td></td>
<td>680.8882</td>
<td>1174.187</td>
<td>1287.657</td>
</tr>
<tr>
<td>trial 3</td>
<td>52.57937</td>
<td>387.299</td>
<td>414.5045</td>
<td>522.0035</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>Fructose</th>
<th>control</th>
<th>4 g/l</th>
<th>8 g/l</th>
<th>12 g/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>trial 1</td>
<td></td>
<td>797.1548</td>
<td>1062.691</td>
<td>730.9791</td>
</tr>
<tr>
<td>trial 2</td>
<td>26.94343</td>
<td>407.6758</td>
<td>632.0373</td>
<td>510.9839</td>
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<tr>
<td>trial 3</td>
<td>41.71245</td>
<td>353.8411</td>
<td>536.8056</td>
<td>378.0436</td>
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</table>

<table>
<thead>
<tr>
<th>Sucrose</th>
<th>control</th>
<th>4 g/l</th>
<th>8 g/l</th>
<th>12 g/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>trial 1</td>
<td>128.2751</td>
<td>579.6703</td>
<td>1191.388</td>
<td>682.8013</td>
</tr>
<tr>
<td>trial 2</td>
<td></td>
<td>1040.733</td>
<td>953.7692</td>
<td>451.9537</td>
</tr>
<tr>
<td>trial 3</td>
<td>69.43554</td>
<td>550.8865</td>
<td>558.7963</td>
<td>710.9407</td>
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<table>
<thead>
<tr>
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<th>Sucrose</th>
<th>Glucose</th>
<th>Fructose</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
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<td>197.2286</td>
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<td>460.4281</td>
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<td>380.1753</td>
<td>10.35943</td>
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<table>
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<tr>
<th>8 g/l</th>
<th>Sucrose</th>
<th>Glucose</th>
<th>Fructose</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
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<table>
<thead>
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<th>12 g/l</th>
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<th>Fructose</th>
<th>Control</th>
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</thead>
<tbody>
<tr>
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<td>440.4202</td>
<td>1308.219</td>
<td>730.9791</td>
<td>93.44348</td>
</tr>
</tbody>
</table>

why do you want 30% to?  
(you data are +100 ppm ?)