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Lab report Amylase 2011-02-01 Aim: The aim of this investigation is to detect the enzyme activity of amylase in saliva. Introduction: Enzymes are protein catalysts. This means they are chains of amino acids with a particular shape that allows them to interact with a specific molecule called a substrate to bring about a chemical reaction. When the reaction is complete, the enzyme is unchanged and able to interact with another substrate molecule. Many enzymes are present in your digestive system. Each digestive enzyme breaks down a specific type of macromolecule.

For example, pepsin breaks down protein and lactase breaks down lactose, a disaccharide. Digestion begins in your mouth with saliva which contains an enzyme called amylase. Amylase works to break down complex carbohydrates. Through hydrolysis, amylase breaks down the polysaccharide, AKA starch, into the monosaccharide, glucose. In this experiment, Iodine solution will be used to test for the presence of polysaccharides in test tubes. The basic principle of this iodine test is that when an iodine solution (i. e. queous solution of potassium iodide) comes in contact in starch, the solution turns blue black in color. In the presence of iodine, amylose in starch forms a deep blue color. [pic] [pic] Amylase , the enzymeStarch, the substrate [pic] Maltose (a disaccharide), the product Hypothesis: In this investigation we will study the effect of the enzyme salivary amylase on starch. Salivary amylase is an enzyme that catalyzes the reaction that break starch down into maltose, which is a disaccharide.

Iodine will react with the coiled molecules and turn deep blue when added to a solution. A solution that remains a yellowish-brown color is a negative test for starch, whereas one that turns deep blue is a positive test for starch. In

this experiment we will use two kinds of amylase: microbial(Termamyl)l and salivary Materials: 24 hole plate jar of starch a tube of amylase Termamyl (Group A) / water (group C) / Group B will need saliva 2 Pasteur pipettes Mixing stick cup with a prepared solution of iodine cup of water to rinse pipette Procedure: 1.

Prepare a watch, preferably with a stopwatch function. 2. Add a Pasteur pipette with 1 ml of iodine solution to all wells of the plate with 24 holes. 3. To the first hole with a clean Pasteur pipette, add one drop of starch, stir. Observe the color of the mixture. 4. Group A: Add to starch 3 ml of enzyme solution Termamyl 5. Thoroughly mix the starch with the enzyme and immediately add one drop to another hole. Start the stopwatch or note the exact time. Group B: Add to the starch a few milliliters of saliva (volume about 1 teaspoon) 6.

After one minute take a sample of starch and add one drop to the third hole, stirring its contents with the pipette tip. Stir starch all the time. 7. Action described in paragraph. 6 repeat every minute, each time noting table color change in the hole and the consistency of starch. It is recommended to rinse the clean water after each collecting pipettes starch, because it would be starch may affect the credibility of the result. 8. You can complete the experience, when a solution of iodine will stop changing color. Data Collection & Processing Table 1. Iodine test for presence of starch Group A | No. of hole | Time [min] | Color of the solution | | 1 | control | deep blue | | 2 | 0 | deep blue | | 3 | 1 | deep blue | 4 | 2 | deep blue | | 5 | 3 | deep blue | | 6 | 4 | deep blue | | 7 | 5 | purple | | 8 | 6 | purple | | 9 | 7 | purple | | 10 | 8 |

purple | | 11 | 9 | Less purple | | 12 | 10 | More purple than previous | | 13 | 11 | yellowish-brown | | 14 | 12 | A little deep blue | | 15 | 13 | yellow | | 16 | 14 | yellow | | 17 | 15 | yellow | | 18 | 16 | yellow | | 19 | 17 | yellow | | 20 | 18 | yellow | | 21 | 19 | yellow | | 22 | 20 | yellow | | 23 | 21 | yellow | | 24 | 22 | yellow | Table 1. shows the changing of the color of a solution of iodine. The test has been conducted for 30 minutes by adding a 1 drop of solution of Termamyl & starch in every minute. In the first sample only starch was added to the iodine. [pic] Pic. 1. 1 Iodine test for presence of starch Results: In first 6 samples the presence of a starch was fair and clear which is indicated by a deep-blue color but from 7 sample the presence of a starch decreases (color changes)as an amylase started to react with a starch. From 15 sample the color of a solution is yellow which means that there is no starch anymore. The enzyme amylase breaks down the starch. Table 1. 2 Iodine test for presence of starch Group B | No. f hole | Time [min] | Color of the solution | | 1 | control | deep blue | | 2 | 0 | deep blue | | 3 | 1 | Less deep blue than in previous | | 4 | 2 | Less deep blue than in 2 | | 5 | 3 | deep purple | | 6 | 4 | deeper purple | | 7 5 | Less deep purple | | 8 | 6 | purple | | 9 | 7 | Deeper purple | | 10 | 8 | purple | | 11 | 9 | deeper purple | | 12 | 10 | Pale purple | | 13 | 11 | Pale purple | | 14 | 12 | Less pale purple | | 15 | 13 | Less pale purple | | 16 | 14 | pink | | 17 | 15 | Pale pink | | 18 | 16 | Pale pink | | 19 | 17 | Less pale pink | | 20 | 18 | Less pale pink | | 21 | 19 | Pale orange | | 22 | 20 | yellow | | 23 | 21 | yellow | | 24 | 22 | yellow | Table 1. 2 presents the data of the changing the color of the solution of an iodine. In this case instead of adding Termamyl we used human saliva. Results: Effects are almost the same in those 2 tests. But in the Group B the results are more clear. The changing of the colors are very visible what we can see on the

picture 1. 2. Moreover we can deduce that in the Group B it took more time to break down the starch.

But on the other hand we can clearly see all of the steps, because colors are changing in some order, they are becoming brighter but in Group A we observe the rapid transition between colors. [pic] Pic. 1. 2 Conclusion In our experiment we investigated the enzyme activity of amylase by doing a Iodine test on samples with iodine and starch solution. We proved that both kind of amylase break down starch by doing an experiment. We used two kind of amylase to conduct this laboratory. First one was a microbial amylase (Termamyl) and the second one was amylase of human saliva. The results were proper with the hypothesis . Iodine reacted with the starch giving the deep blue color. A solution that remained yellow indicates the absence of starcz, whereas one that turned into deep blue indicates presence of starch.

In A &B groups the results was correct whereas we are able to deduce that human salivary enzymes were more effective in starch hydrolysis than microbial amylase enzymes. This is indicated by the fact that in the picture 1. 2 we can clearly see the steps of hydrolising the starch. Evaluation Although the results were successful there were several assumptions that could be improved. First of all in the Group A in the picture 1. 1 we can observe that 14 sample has deep blue color. This is due to the fact that we added to much of amylase so the concentration was raised therefore the rate of reaction was increased. Moreover in Group B the results were so clear due to the fact that person who supplied saliva had been chewing a gum.

It improved the results because the ph of saliva was changed by chewing a gum. As we have already known the optimum ph for enzymes is 7. Also we should measure the temperature in a class because the temperature also affect the rate of reaction and it could also affect the experiment. However thanks to the prepared experiments and besides the errors I could finish my experiments and collect the relevant data to prove the enzyme activity of amylase. Bibliography: 1. Clegg C. J Biology for the IB Diploma 2. http://en. wikipedia. org/wiki/Enzyme date: 01. 02. 2011 3. http://en. wikipedia. org/wiki/Amylase date: 01. 02. 2011 4. http://en. wikipedia. org/wiki/Iodine_test date: 01. 02. 2011