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Amylase from House Cockroaches (*Periplaneta americana*) as Medium for Ethyl Alcohol Extraction from Common Agricultural Wastes

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ABSTRACT

Bioconversion of starch into ethanol or ethyl alcohol involves two-steps. The first step is saccharification which converted starch into sugar using enzymes. The second step includes two phases started with fermentation where sugar is converted into ethanol and ends with distillation where pure ethanol is collected as ethyl alcohol. Although there are over 150 amylolytic yeast species, the amylase from cockroaches was extracted and used as medium enzyme to produce ethyl alcohol from common agricultural wastes.

Of the five sample wastes, potato (Solanum tuberosum) peelings, mango (Mangifera indica) seeds, bandera espaňola (Canna indica L), cassava (Manihot esculenta) peelings and banana (Musa acuminate) peelings, the potato peelings yielded the highest amount of ethyl alcohol. Moreover, the yielded alcohol from agricultural wastes was cloudy, with sharp odor, flammable and in liquid form which made it physically comparable to commercial ethyl alcohol. The enzyme amylase in cockroaches have the shortest period of breaking sugar molecules and separation of carbon dioxide and ethanol processes compared to enzyme in yeast and bacteria

Keywords: Canna indica L, Distillation, Ethyl Alcohol, Extraction, Fermentation

INTRODUCTION

Insects are commonly found in almost all households and cockroaches is one among them. Insects are the most widespread of all animals. Some insects are friends of human but great majority are harmful to man simply because they destroy crops and spread diseases like cockroaches [1, 2]. Commonly, people have thought of cockroaches as poisonous and dangerous insect but researches proved they are not though this insect can play good role in initiating diseases. They smell awful and multiply fasts [3]. Cockroaches, because of their numbers and their property to easily multiply, are an important part of our ecosystem. They can eat a wide range of substance including other cockroaches [4]. Because of the variety of bacteria and protozoa in their gut, they play an important role in the rapid decomposition of forest litter and animal fecal matter including agricultural wastes [1].

Household wastes which are present almost anytime and are being disposed of in various manners like recycling, reusing and decomposing which make these wastes useful for other purposes. Some dumped these wastes which attracts various types of insects and thus a potent source of diseases. If household wastes are to be treated, they can be a possible source of ethyl alcohol. Ethyl alcohol or ethanol to some has been made since ancient times by the fermentation of sugars. In the most common way, fermentation process of ethyl alcohol lasts for about a month [5]. Natural starch from potato (*Solanum tuberosum*) [6] peelings, mango (*Mangifera indica*) [6] seeds, bandera española (*Canna indica L*) [6], cassava (*Manihot esculenta*) [6] peelings, and banana (*Musa acuminate*) [6] peelings can be raw materials. The yeast which can be enzyme, zymase and amylase changes the simple sugar into ethanol and carbon dioxide. Naturally made, amylase is present in cockroaches [7]. Thus, this study took the liberty of investigating on the effectiveness of amylase from the subject insect as medium to produce ethyl alcohol.

METHODOLOGY

Procedure and Materials

Collection and Preparation of Raw Materials

One hundred fifty grams (150grams) each of banana peelings, bandera española plant, potato peelings, mango seeds and cassava peelings were collected, sundried, grinded, and mixed each sample with 100ml water. Each mixture was called substrate. Each sample was secured individually in a flask and was prepared in triplicates.

Materials

One hundred (100) grams of cockroaches were acquired (without the stomach, wings and other extremities), and grinded by sand to obtain 50 ml amylase. Sample yeast and bacteria were also prepared. Five sets of amylase, yeast and bacteria were prepared. Filtration, Extraction, and Saccharification Process

The amylase and yeast were filtered. The filtrates were poured in each of the flasks containing substrates and set aside. The same was done with bacteria.

Fermentation and Distillation Processes

The three sets of substrates cockroach amylase, yeast and bacteria were subjected to 2-week fermentation period and distillation was done after to obtain ethyl alcohol. Each enzyme has 5 substrates.

RESULTS AND DISCUSSION

Table 1 shows the amount of ethyl alcohol produced from various agricultural wastes. With similar amount of grinded sample wastes, water as the dilute substance, amylase as enzyme and similar period of fermentation, varying amount of alcohol were produced. As revealed in the table, potato peeling yielded the highest amount of ethyl alcohol which was 9.0 ml followed by cassava peeling with 7.0 ml while bandera española yielded the least amount of 3.5 ml. While it is true that the sample agricultural wastes have carbohydrate content in their chemical composition, the role of amylase in cockroaches was to extract this vital component in the formation and production of ethyl alcohol. Fact is, potato is a good source of carbohydrate thus potato peelings yielded the highest amount of ethyl alcohol.

| Agricultural | Weight of | Volume of | Volume of | Volume of | Volume of | Length of | Volume of |
|----------------|-----------|-----------|------------|-----------|---------------|--------------|---------------|
| Waste | Grinded | Water | Gelatinous | Amylase | Sample and | Fermentation | Alcohol After |
| | Materials | Added | Substrate | Enzyme | Enzyme Before | Period | Distillation |
| | | | Formed | | Distillation | | |
| | | | | | | | |
| | | | | | | | |
| Banana Peeling | 150 g | 100 ml | 250 ml | 50 ml | 300 ml | 2 weeks | 6.5 ml |
| | | | | | | | |
| Cassava | 150 g | 100 ml | 250 ml | 50 ml | 300 ml | 2 weeks | 7.0 ml |
| Peeling | | | | | | | |
| Potato Peeling | 150 g | 100 ml | 250 ml | 50 ml | 300 ml | 2 weeks | 9.0 ml |
| | | | | | | | |
| Bandera | 150 g | 100 ml | 250 ml | 50 ml | 300 ml | 2 weeks | 3.5 ml |
| Espaňola | | | | | | | |
| | | | | | | | |
| Mango Seed | 150 g | 100 ml | 250 ml | 50 ml | 300 ml | 2 weeks | 5.0 ml |
| | | | | | | | |

Table-1: Ethyl Alcohol Produced from Different Agricultural Wastes

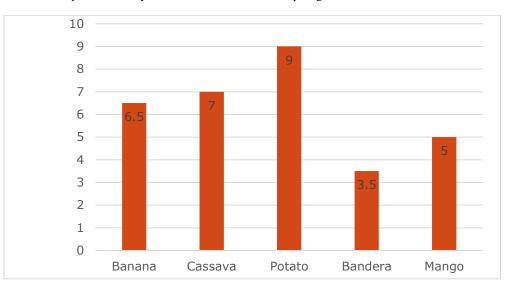


Figure 1 described the comparison of the yielded alcohols of the five sample agricultural wastes.

Figure-1: comparison of the yielded alcohols of the five sample agricultural wastes

Table 2 shows the comparison of the physical characteristics of commercial ethyl alcohol and ethyl alcohol produced from agricultural wastes using enzyme amylase in cockroaches as an aggregate. As seen in the table, both kinds of alcohol were in liquid form. The commercial ethyl alcohol was colorless while the produced ethyl alcohol from agricultural waste was cloudy. In terms of odor, both samples have sharp odor and were both flammable when subjected to a source of heat. The cloudy color of the produced alcohol was due to the manual process of production.

| Physical Characteristics | Commercial Ethyl Alcohol | Ethyl Alcohol from Agricultural Wastes |
|--------------------------|--------------------------|--|
| State | Liquid | Liquid |
| Color | Colorless | Cloudy |
| Odor | Sharp | Sharp |
| Flammability | Flammable | Flammable |

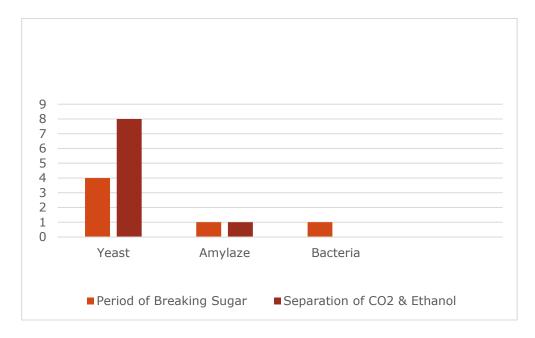
Table-2: Comparison of Physical Characteristics of Commercial Ethyl Alcohol and Ethyl Alcohol Produced from Agricultural Wastes

On Table 3, the enzyme yeast took four weeks in breaking the carbohydrate molecules and the separation of CO_2 to form ethanol took 8 weeks. The enzyme amylase from cockroaches took only 1 week in breaking carbohydrate molecules while the period of separation of CO_2 and ethanol required 2 weeks. Lastly, the enzyme in bacteria took 1 week period of breaking sugar molecules and found not capable of separating CO_2 and ethanol.

| Derivatives of Enzyme | Period of Breaking Sugar Molecules | Period of Separation of CO ₂ and Ethanol | |
|-------------------------------|------------------------------------|---|--|
| Enzyme in Yeast | 4 weeks | 8 weeks | |
| Enzyme Amylase in Cockroaches | 1 week | 2 weeks | |
| Enzyme in Bacteria | 1 week | Not capable | |

Table-3: Comparison of Period of Enzymatic Reaction in the Production of Ethyl Alcohol

Figure 2 shows the graphical presentation of the period of enzymatic reactions of the three kinds of enzymes used in the experiment. As shown, the enzyme amylase from cockroaches gave the fastest period of enzymatic reaction and the best period in separating carbon dioxide and ethanol. This means that in order to produce more amount of ethyl alcohol within a shorter period of time, the enzyme amylase from cockroaches can do the task.





CONCLUSION

As observed, the enzyme amylase from cockroaches accelerates the chemical reaction of the process in producing ethyl alcohol. While it was true that all the sample agricultural wastes have carbohydrate components, the potato peelings yielded the highest amount of ethyl alcohol while bandera española yielded the least amount, considering other variables constant. Moreover, the yielded alcohol from agricultural wastes was cloudy, with sharp odor, flammable and in liquid form which made it physically comparable to commercial ethyl alcohol. Finally, the enzyme amylase in cockroaches was found to have the shortest period of

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breaking sugar molecules and separation of carbon dioxide and ethanol processes compared to enzyme in yeast and bacteria, thus utilizing amylase in cockroaches in ethyl alcohol production can save time and effort which are both important in the busy activities of a chemist.

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