Extraction of Custard Apple Seed Oil to Produce Natural Pesticide

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Abstract
Synthetic pesticide is commonly used by farmers and in houses to kill pests mainly mealy bugs. Synthetic pesticide functions well but without our knowledge it gives various negative effects to the environment and human health. In conjunction with this, the idea of extracting oil from custard apple seed to produce natural pesticide was executed. The objective of the research is to produce an organic natural pesticide from custard apple seed. Secondly, to determine the amount of methanol solvent used to extract custard apple seed powder. To produce natural pesticide, custard apple seed is required. Moreover, methanol solvent is used to extract the oil from the seed kernels. The extraction is carried out with various amount of methanol solvent along with constant temperature, it is 67℃. The product from the extraction is certain amount of oil and it is classified as natural pesticide. Furthermore, the extracted samples were tested on 9 different plants for 9 days by spraying the pesticide on affected part. The results show the best functioning pesticide is depends on the amount of methanol solvent used. From the research, the objectives have been achieved. The research concludes, the extracted oil from custard apple seed can be used as an organic pesticide.

Keyword: Apple seed oil, natural pesticide, extraction

1.0 Introduction

The growing population demands a greater amount of crop yield which results in the increased usage of pesticides. The population growth means more food is being produced and the usage of chemical is more likely. Like other organisms, humans compete with other species for food and shelter and struggle to protect themselves from predators. For a stable and healthy population to survive, the use of pesticides must be applied to crops for greater efficiency. Insecticides are used to kill insects which transmit diseases. A pesticide is any substance or mixture of substances that prevents, destroys, repels, or mitigates any pest. A pest is an animal or plant that can injure the environment or the health of populations in that environment. Besides, crop protection products are also known as pesticides. Pesticides are classified into two groups which are synthetic and natural pesticides. Chemical or biological substances used to control unwanted pests that can harm a food, health, or environment. Pesticides are one of the vital tools that help farmers grow healthy crops, protecting the food supply against yield losses and damage caused by the weeds and insects. The crop yields as well as the quality of the food will drop due to the lack of usage of pesticide. This would result in shortage of food supply and a sudden increase in food prices (Popp, J., Pető, K., & Nagy, J., 2013).

Natural pesticides can also be called organic or biopesticide. These products are derived from the ground, plants or even animals. The purpose of organic pesticides is to cultivate an organic garden. Organic pesticides are being

promoted heavily in recent years due to its benefits and risk as well. The benefits are they are friendlier to the environment. In other words, go green where we could reduce the carbon footprint. Next, natural pesticides are healthier for all living organisms because humans intend to consume the vegetables and fruits that are sprayed. The quality of soil can be improved by using natural pesticide. This results in better overall plant growth and mean less chances for contamination. Pest can never develop resistance and this pesticide will never cause pollution. It is also relatively cheaper than synthetic pesticide. On the other hand, the risks of using natural pesticides are it would delay the process by taking longer to work. Simultaneously, some products will be less effective. Many companies offer natural chemicals however it is harder to find natural pesticide. Therefore, natural pesticides are safer and eco friendlier to be used (Damalas & Eleftherohorinos, 2011).

Custard apple or sugar apple is one of the important minor fruit crops belonging to the family of Annonaceae. It can be grown in any tropical countries except temperate regions. (R. Maruthadurai & V. Karupaiah, 2014) Custard apple plant grows well in hot climates and adjusts in any kind of soil. The seeds of the custard apple germinate in 3 weeks between the temperature 18 ◦C and 25 ◦C. The custard apple seeds have good antifungal properties. The custard apple seed oil is an insecticide, inhibiting the activity of insect feeding and repellent to pests such as aphids, caterpillars, white mealy bugs, grasshoppers and plant hoppers. (Garud, A., Garud, N., & Tailang, M., 2015).

2.0 Problem statement

The environmental impact of pesticides is often greater than what is intended by those who use them. Over 98% of sprayed insecticides and 95% of herbicides reach a destination other than their target species, including non-target species, air, water, bottom sediments, and food (Steve, 2012). Though there can be benefits using pesticides, inappropriate use can counter productively increase pest resistance and kill the natural enemies of pests. Pesticides can contaminate unintended land and water when they are sprayed aerially or allowed to run off fields. Furthermore, synthetic pesticide is dangerous to human health and environment. Consuming foods from chemical pesticide sprayed plantations will give adverse health effects by disrupting our hormonal growth and leads to cancer (Damalas & Eleftherohorinos, 2011). Environmentally, it effects our food chain besides creating and worsening land pollutions. Therefore, we aim to develop natural pesticide from custard apple seed oil to help reduce and stop this poisoning to species, organisms, risk of human health and environment from further prolong disaster.

3.0 Objective of study

Objectives of this study are firstly, to produce an organic natural pesticide from custard apple seed and secondly to determine the amount of methanol solvent used to extract custard apple seed powder.

4.0 Literature review

Insects, fungi and plants are considered beneficial until they endanger humans' health and compete food in daily life, at which time they become pests. Three millennia have passed since Homer mentioned 'pest averting sulphur' and more than a century since dinitro-o-crosel became the very first synthetic organic insecticide.

Many theories have been proposed followed by suitable experiments to find out the advantages, disadvantages, benefits and uses of pesticides- both botanical and synthetic. In a world where productivity is given priority, one cannot effort to damage agriculture's fruit when the collateral damages can be controlled. Pesticides that are generally known as chemical compounds are used to kill pests (bugs), including insects, rodents, fungi and weeds. Considering cultivation, scientists classified these pesticides orderly according to their functions, level of chemical compounds used, locations and such.

Proteomics is an emerging discipline in pesticide toxicology, pesticide metabolism and mechanism of pesticide were accepted in numerous areas of pesticide research with wider understanding of pesticide resistance, mechanism of toxic action, mode of action and biodegradation of pesticides has aided the discovery of new effective and safe pesticides with identification if bio-makers' protein (Su-Wei Qi and Qing X. Li, 2010).

Pesticide is a common term that characterizes several classes of insecticides, herbicides, fungicides, rodenticides, wood preservatives, garden chemical sand household disinfectants that are used to either to kill or protect from pests. These pesticides differ in their physical, chemical and identical properties from one class to other. Therefore, it is worthy to classify them based on their properties and study under their respective groups.

Synthetic pesticides are manmade chemicals, and do not occur in nature. They are categorized into various classes depending on the needs. Presently, there are three most popular method of pesticides classification suggested by Drum. These three popular methods of pesticides classes comprise:

(i) Classification based on the mode of entry,
(ii) Classification based on pesticide function and the pest organism they kill,
(iii) Classification based on the chemical composition of the pesticide.

Toxicity of a pesticide is defined as the ability to cause injury or illness. Toxicity of a specific pesticide is decided by applying test animals to varying dosages of the active ingredient and each of its formulated products. Ague toxicity of a pesticide attribute towards the chemical's ability to cause any injury to a person or animal from a simple exposure, usually for brief duration. Exposure is focused on eyes, oral or mouth, dermal or skin and inhalation or lungs. Acute toxicity is obtained simply by examining the dermal, oral, inhalation toxicity of test animals. Besides, both the eye and skin irritation are also examined. The toxicity of a pesticide is measured in LD50 which is defined as the concentration of a toxicant needed to kill 50% of test animal population (Hamm, J., 2017). The value is based on its single dosage and are measured

using milligram per kg of weight (mg/kg). The higher the LD50 value the greater threat is it to human and animal.

The chronic toxicity of a pesticide is decided by exposing the test animals to extended exposure to the active substances. Any harmful effects that happen from small doses repeated over a period are defined chronic effects. Some of the identified effects from vulnerability to specific pesticides including neurotoxic effects (nerve disorders), blood disorders, production of tumours, and birth defects. The chronic toxicity of a pesticide is harder to determine through laboratory analysis than acute toxicity. Products are classified on the foundation of their related acute toxicity (their LD50 or LC50 values) (Eric S. Lorenz, 2017).

5.0 Materials and methods

5.1 Extraction of oil from custard apple seed

As a first step, custard apple seeds were washed and sundried. The custard apple seeds which is free from moisture is then crushed and grounded into fine powders. Mortar and pestle were used to ground the seeds. Then, the grounded seeds were transferred into a crucible using a spatula.

Next, an empty crucible was weighed on a weighing balance. An approximated level of grounded kernel seeds was transferred into that empty crucible using spatula. This process was repeated until it reaches the desired value it is 3g.

Later, estimated 30 ml of methanol was poured into a 100 ml beaker. It was adjusted so that there will be an accurate level of 30 ml methanol. Followingly, the 30 ml methanol was transferred into a round bottom flask which is the bottom part of the soxhlet using a funnel.

Then, some cotton wool was placed into the soxhlet followed by the grounded kernel seeds. Later, distilled water was added to the water bath. On the other hand, the completely fixed soxhlet was clamped to the retort stand. It was set up partially immersing at the water bath. The water bath then was turned on and the temperature was set for about 65℃ for 12 hours of extraction. Water from the tap was turned on so that there is inflow and outflow of water at the vertical condenser from the tube connected at the side.

Upon completion of 12 hours, the extracted oil is filtered out using a filter paper. The filter paper was kept on a filter funnel and the oil transferred to a small beaker. Then, pH value for the oil were taken to determine the pH value for the sample. Next, the oil was transferred to a spray bottle and labelled as A.

These steps were continued for the rest 8 samples. But the extraction time differs. Sample B was extracted for 8 hours while sample C was extracted for 4 hours with 30 ml of methanol. Besides, for sample D, it was extracted for 12 hours, sample E for 8 hours and sample F for 4 hours with 40 ml of methanol solvent. Meanwhile, sample G was extracted for 12 hours, sample H for 8 hours and sample I for 4 hours with 50 ml of methanol.

5.2 Planning for testing

For each sample, pH value was tested to determine the acidity. Besides, to identify the objective, testing is carried out on plants. Since there are 9 samples from extraction, thus 9 plants are chosen to conduct the testing. The plants were chosen according to flowering and non-flowering plant. The plants are labelled from A to I representing the samples extracted. The testing was conducted with 3 days gap in between.

Figure 2: Plants for testing

6.0 Discussion

6.1 Results of the samples

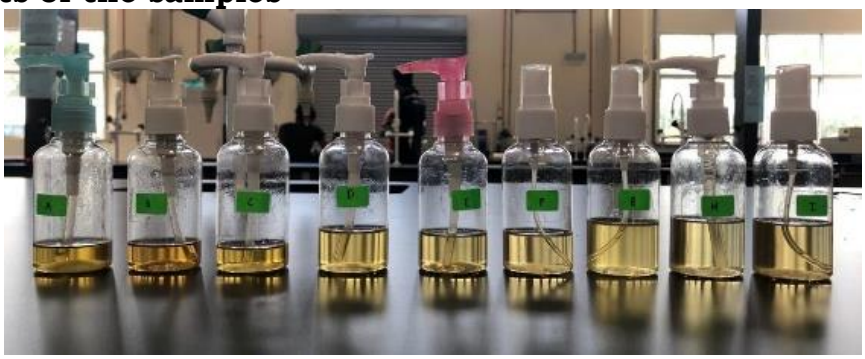

Figure 3: Extracted samples

According to the figure 3 above, the amount of the samples differs depend on the amount of solvent used. Samples A, B and C are extracted using 30 ml of Methanol solvent. Meanwhile, samples D, E and F are extracted with 40 ml of Methanol solvent. Samples G, H and I are extracted with 50 ml of Methanol solvent. From this, the more the amount of methanol, the greater the volume of extracted samples.

Secondly, the colour intensity is different. Samples A, B and C is in woody brown colour whereas samples D, E and F are in brownish yellow colour while samples G, H and I are in light brown colour. The amount of methanol solvent affects the colour intensity of the extracted samples.

The third physical properties of the extracted samples are it was in liquid form. This enhances the sample even more because oil type fluid will cover the cuticle layer of the leave and will act as wax. Thus, it will affect photosynthesis process. However liquid type sample will not prevent the plant from carrying out photosynthesis. The chemical property for the samples is, it was irritant and has quite strong smell.

Table 1: Properties of samples

Solvent used	Sample	Volume of methanol	Time taken for extraction	Colour
METHANOL	A	30 ml	12 hours	Dark Yellowish-brown
	B		8 hours	Dark Yellowish-brown
	C		4 hours	Dark Yellowish-brown
METHANOL	D	40 ml	12 hours	Yellowish-brown
	E		8 hours	Yellowish-brown
	F		4 hours	Yellowish-brown
METHANOL	G	50 ml	12 hours	Light Yellowish-brown
	H		8 hours	Light Yellowish-brown
	I		4 hours	Light Yellowish-brown

6.2 Results from pH meter

Once completed the experiment, the pH values were taken for each sample to determine the acidity. According to research, the ideal pH for pesticide is from 5.5 to 6.5 (Gentili, R., 2018). This pH is weak acid in terms where there is no negative effect towards the plant. However, the pest will be killed. In this experiment, all the samples are categorized as weak acid. The pH test conducted for this samples achieves the target of other pesticides in market.

Table 2: pH Values of Each Sample

Samples	pH
A	6.20
B	6.38
C	6.39
D	6.22
E	6.31
F	6.36
G	6.45
H	6.31
I	6.34

6.3 Testing on the plants

9 plants were chosen to test the 9 extracted samples

Figure 4: Spraying the samples

The results were obtained after 3 days from the testing. Results for each plant after 3 testing is shown.

Figure 5: Testing 1, 2 and 3 for Plant A

Figure 6: Testing 1, 2 and 3 for Plant B

Figure 7: Testing 1,2 and 3 for Plant C

Figure 8: Testing 1,2 and 3 for Plant D

Figure 9: Testing 1,2 and 3 for Plant E

Figure 10: Testing 1,2 and 3 for Plant F

Figure 11: Testing 1,2 and 3 for Plant G

Figure 12: Testing 1,2 and 3 for Plant H

Figure 13: Testing 1, 2 and 3 for Plant I

In this experiment, the extracted pesticide has successfully reduced the number of pests in the 9 days of testing towards the plants. Throughout the testing, sample D functioned well as there are some obvious visible changes on the leaf. The leaf has been captured from day one till the third testing and has been attached. There is reduction in the number of pests and at the same time, the black spots have decreased. This can be seen as the leaf has a clearer layer after the testing. On the other hand, the extracted pesticide does not harm the plant by making the leaf to turn yellowish and wilt. Besides, the plant could carry out photosynthesis as usual as the pesticide is in liquid form. Mostly, much pest could not be found on plants in our home since the plants been cleared from weeds and it has been taken care well. Testing could not be done in any plantation due to the outbreaking pandemic.

7.0 Conclusion

Based on the results, the objective of this project has been achieved. Natural pesticide is produced from custard apple seed and determined the amount of methanol solvent used to extract the custard apple seed powder on various temperature. The more the volume of methanol solvent, the greater the value of pH for 12 hours extraction. Meanwhile the pH values were low for 8

hours and 4 hours extraction. The amount of methanol solvent used for extraction does not affect the function of pesticide as all the samples reduced pests. The pesticide was in liquid form where it enables the photosynthesis process. If the pesticide produced happened to be in oil form, it would have acts as wax on the cuticle layer of the leaf and will block the leaf from trapping sunlight to produce food. We can conclude that, the natural pesticide produced from custard apple seed proves itself efficient, advantageous, cheap safety to handle. This raw material will be very cheap which minimizes the total cost of processing along with solvent recovery.

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