

Oyster Mushroom–Synthesized Silver Nanoparticles as Biopesticide for Cabbage Aphids

Audrey Dane A. Parcon, Fiorella Dawn L. Racho, Juddeah Grace S. Mencion

SUMMARY

Nanotechnology is rapidly transforming modern agriculture by offering innovative strategies for pest management that are both effective and environmentally sustainable. Traditional chemical pesticides, while widely used, often pose significant ecological and health risks, including soil degradation and toxicity to non-target organisms. To address these challenges, green synthesis approaches have emerged, allowing natural biological systems to produce functional materials without harmful byproducts. Fungal extracts, in particular, have shown great promise as eco-friendly agents for generating nanoparticles with potent biological activity. Here we show the use of silver nanoparticles synthesized via mushroom extracts to control cabbage aphids, a common agricultural pest that significantly reduces crop yield and quality. Our approach demonstrates that biologically derived nanoparticles can target pests effectively while minimizing damage to the plants and surrounding ecosystem. By combining nanotechnology with sustainable biological methods, this research highlights a practical pathway toward reducing reliance on synthetic chemicals in agriculture. The results suggest that fungi-assisted nanoparticle synthesis can serve as a valuable tool in creating safer, more resilient, and environmentally conscious farming practices.

Background of the Study

The agriculture sector always has to deal with pest control as its main problem, but it still prefers to maintain the environment. The routine usage of chemical pesticides has caused a series of negative impacts, all of which are connected to soil degradation, water contamination, and the emergence of pests that have developed resistance (Prasad et al., 2017). Cabbage aphid is one of the major pests of cruciferous plants. It is a siphoning insect that also transmits some viral diseases. This means that the quality of the plants is lowered by the insects and hence the crops get to be of lower quality, so the farmers and the consumers get to be of lower quality as well. For this reason, the only priority left in the present world is to pest control that is eco-friendly and not toxic.

One would wonder how come nanotechnology is the one and only method of controlling field pests in agriculture that becomes so fascinating to people. The Silver nanoparticles have already reached the status of being the finest for their antimicrobial and insecticidal activities, which can effectively reduce pest populations even at low doses (Rai et al., 2012). They generate oxidative stress and also harm the pest's membranes, thus causing death. However, traditional methods for producing AgNPs involve the use of toxic chemical reducers, which not only impact negatively on the environment but also the health of people living around (Govindaraju et al., 2016). Hence, there is a shift in people's preference regarding the synthesis of nanoparticles through biological agents, which is considered a green alternative and less harmful to both the environment and health.

The research project was conducted with the aim of developing a new eco-friendly method based on the extraction of silver nanoparticles from oyster mushrooms and their insecticidal activity against cabbage aphids (*Brevicoryne brassicae*). The experiment involved spraying infested cabbage leaves with different concentrations of silver nanoparticles (25 ppm, 50 ppm, and 100 ppm) in the laboratory conditions. The aphids were observed for 5 days to assess the mortality rate and behavioral changes such as feeding inhibition and reduced activity. The investigation has hypothesized that the application of the maximum concentration of silver nanoparticles will result in the total killing of the insects with the least plant damage.

Nothing by now has been referred to as a green substitute for reduction agents as it is still enabling the process of making silver nanoparticles the non-harmful way. There are already proofs of the fungal AgNPs' use in their powerful antibacterial and antifungal activities, however, their usefulness against insect pests is an area that has yet to be developed, therefore, the prospect of mushrooms being a source of natural nanopesticides still remains.

The study aimed to investigate the extraction of silver nanoparticles through the use of *P. ostreatus* mushroom and the insecticidal activity of the nanoparticles against *Brevicoryne brassicae* aphids. The infested cabbage

leaves were subjected to light spraying of the AgNPs (25 ppm, 50 ppm, and 100 ppm) in a laboratory setting. Following that, the aphids were monitored for 5 days for the assessment of mortality rate and changes in behavior such as feeding inhibition and reduced activity. The research aims to present *P. ostreatus* as an eco-friendly source of nanoparticles, which is one of the main goals. The idea is to provide a natural pest control solution that may eventually replace, in part, the heavy reliance on chemical pesticides in agriculture while still maintaining crop protection. It was predicted by the study that the application of AgNPs at the highest concentration would result in the total elimination of the aphid population with the least damage to the plants.

Literature Review and Research Gaps

The chemical pesticides have been used traditionally and on a large scale for controlling pests in agriculture. Though, the uncontrolled usage of these pesticides has created problems for the environment and health, such as soil and water pollution, and the death of non-target organisms like beneficial insects. Moreover, the cabbage aphid (*Brevicoryne brassicae*) has been one of the insect pests that has grown resistant to such chemicals over time, making them less potent. The concurrent problems of chemical pest control have prompted the search for alternatives that are not only safe for the environment and humans but also sustainable and less harmful.

Nanotechnology has marked the dawn of a new era in agricultural science with its innovative and promising methods (Bhardwaj & Naraian, 2022). Amongst them, silver nanoparticles (AgNPs) have caught the limelight being most effective due to their already known antimicrobial and insecticidal properties. The action of AgNPs is exquisite as they are so small so they can easily get in and destroy pest's cells through oxygen generation by oxidation and shutting down of the cellular functions. Nevertheless, the conventional methods of synthesizing AgNPs are laced with the use of harmful reducing agents that are not suitable for large-scale agriculture. Because of this, researchers have diverted their attention towards green methods of synthesis that make use of biological materials that are non-toxic instead of harmful chemicals.

Fungal-mediated nanoparticle production is one of the green synthesis methods that hold a lot of promise. Fungi are prolific producers of enzymes and proteins which play the role of reducing agents for silver ions and nanoparticle stabilizers, thereby making them the most efficient biological factories. The oyster mushroom (*Pleurotus ostreatus*) is particularly good at it as it has bioactive compounds like polysaccharides and phenolics which enhance the nanoparticle birth. The studies have already proved that AgNPs obtained from mushrooms have extremely potent antibacterial and antifungal qualities. However, traditional techniques in synthesizing AgNPs still include the use of toxic reducing agents, which are prohibited for application in agriculture on a larger scale. Therefore, scientists have turned their back on the old methods and moved towards green processes that rely on using nature's biological materials rather than employing chemicals that are unsafe for the environment.

Fungal-mediated production of nanoparticles among green synthesis methods has attracted much attention due to its high potential. The biological process is still high up the chain as a unique way to matter, with non-toxicity and recyclability of the whole process being some of the reasons. The oyster mushroom (*Pleurotus ostreatus*) is, in particular, a very good example because it has compounds, among them polysaccharides and phenolic substances that are responsible for the encouraging of nanoparticle forming. Previous research has already confirmed that AgNPs generated from fungi have substantial antibacterial and antifungal activity.

However, not much has been done regarding the insecticidal effects, which is the area this research intends to fill. Cabbage is one of the main vegetable crops in the Philippines but it is very easily attacked by aphids that can ruin a big part of the harvest. Farmers mostly resort to chemical insecticides which come with higher costs and risks to the environment. There is no local research that investigates the use of green-synthesized silver nanoparticles as a pest control alternative in the Philippines. This study uses oyster mushroom extract for nanoparticle biosynthesis and presents a natural, safe, and cost-effective way for *B. brassicae* management. The work may foster the adoption of sustainable agriculture practices in the country as well as the reduction of chemical pesticide use.

Rationale

The environmental and health problems associated with chemical pesticides have been one of the major issues which have affected mankind globally. The advantages of the chemical pesticides are, however, overlapped by the disadvantages as they are gradually causing pests to get resistant, polluting the water and soil, and killing even the good ones. Naturally, the demand for eco-friendly pest control alternatives that are both effective and safe is increasing, therefore, the need for safer, sustainable pest control alternatives that can protect crops without damaging ecosystems is becoming more urgent.

The application of nanotechnology for the use of silver nanoparticles (AgNPs) as a potent pest control agent offers a new way to address the issue. AgNPs are characterized by a robust antimicrobial activity along with insecticidal properties. The mechanism of action of the AgNPs involves the destruction of the pest's cell and the simultaneous induction of oxidative stress which leads to a very effective pest control even when the AgNPs are being used at very low doses. Still, most of the conventional synthesis methods are using toxic chemicals, making the process harmful and expensive for large-scale agricultural applications.

The method of fungal-mediated synthesis for AgNPs production is certainly an environment-friendly one. The edible oyster mushroom (*Pleurotus ostreatus*) has the capacity of producing the nanoparticles through enzymatic activity, all this without leaving behind any waste or pollution. A dominant pest in the Philippines, commonly known as a cabbage aphid is one of the major problems local farmers deal with. These pests kill the plants, and it also exposes them to viral infections by sucking the sap from the plants. Therefore, the farmers have no other choice but to employ costly insecticides that are not only harmful to the environment but also to the consumers, which are bug-killing chemicals.

A solution based on nanobiotechnology might be the safest and probably the most effective way to solve this problem. Silver nanoparticles in conjunction with the oyster mushroom extract were the main focus of this study, and their insecticidal action against cabbage aphids was assessed. A nanoparticle system based on fungi was developed in this work and it not only supports organic farming but also offers a possible alternative to synthetic pesticides. It helps not only to find less harmful pest control methods but also to promote the protection of the environment.

Significance of the Study

In the long term, eco-friendly pest control methods that do not harm the environment are a must if global agriculture is to be sustainable. Researchers, on the one hand, are looking for suitable alternatives that can wipe out the pest population without the risk of long-term ecological damage. The present study is a small part of this movement as it explores the green and non-toxic way of biosynthesizing silver nanoparticles (AgNPs) through the use of oyster mushrooms (*Pleurotus ostreatus*), which is a not-so-common approach in nanoparticles synthesis. Not only this, but also the findings of such research can lead the world to the acceptance of pest control and using milder techniques on the farm.

At the continent level, countries in Asia are already experiencing the collapse of their agricultural systems because of the lack of proper pest control, pollution of the environment, and the inability to supply food. The use of biological and nanotechnology-based pest control methods would be an effective solution to the problems and would lead to a more sustainable agricultural system. The present study substantiates the claim that fungi, especially *P. ostreatus*, can be trusted as biological agents for the generation of nanoparticles. It may also inspire more research to be done in the entire region on the possibilities of applying biological materials, which are readily available, to the field of agricultural nanotechnology.

In the Philippines, this research is important because it tackles sustainable development and one of the most widely spread agricultural problems at the same time, the cabbage aphid infestations. The present study, by the application of fungus-mediated silver nanoparticles as a natural biopesticide, brings up an alternative that is cheap, safe, and within reach of local farmers, which is the newly incurred research. The outcomes may help in shaping future agricultural research directed towards the development of biobased pest control technologies and might even have a positive impact on policy-making for the adoption of eco-friendly farming practices. In the end, pest management solutions are not the only area of impact for this research, but it also boosts the country's environmental protection and food security agenda.

Scientific Framework

The research project under review discussed the availability of silver nanoparticles (AgNPs) through a biosynthetic route with oyster mushrooms (*Pleurotus ostreatus*) being employed as the natural reducing agent along with the stabilizing agent. The mushrooms comprise specific enzymes and proteins that interact with gold ions (Ag^+) in the silver nitrate (AgNO_3) solution and convert them into nano-sized metallic silver (Ag^0) through a green synthesis pathway. This technique is environment-friendly since it does not require the utilization of any toxic chemicals and is more sustainable with respect to the generation of nanoparticles.

The color change of the reaction mixture from clear to brown during the synthesis indicated the successful production of AgNPs. The mushroom extract provided the necessary bioactive compounds which not only reduced the metal but also capped the nanoparticles making them stable and dispersible. It is a biological process that produces AgNPs that are expected to exhibit insecticidal activity due to their ability to generate Reactive Oxygen Species (ROS) that are lethal to pest cells.

The AgNPs when used on cabbage aphids (*Brevicoryne brassicae*) interact with the insects' outer membranes, prevent feeding, and cause oxidative stress which ultimately leads to death. Various concentrations (25 ppm, 50 ppm, and 100 ppm) were employed for the most effective dosage to be determined.

The model reveals the connection between fungal-mediated nanoparticle synthesis and pest control efficiency. It clarifies how *P. ostreatus* can be regarded as a biological factory for producing AgNPs which are eco-friendly insects.

Scope and Limitations

The primary objective of the study is to create a technique for generating silver nanoparticles (AgNPs) using the extract of the oyster mushroom (*Pleurotus ostreatus*) and to study their potential as insecticides against cabbage aphids (*Brevicoryne brassicae*). The entire operation was divided into the stages of mushroom extraction, synthesis of silver nanoparticles by the reduction of silver nitrate (AgNO_3) followed by the application of three distinct nanoparticle doses (25, 50, and 100 ppm) to methyl beet-infested cabbage leaves. Through this experiment, the death of plants, type of feeding, and area of damage were observed and recorded at the end of day five.

The study was confined to laboratory experiments under tightly controlled conditions. Characterization of the AgNPs produced was not made through advanced tools like UV-Vis spectroscopy and SEM because of the unavailability of the said equipment. Only the pest *B. brassicae* was taken as the target during the trial, and other insect species or factors like the soil and humidity changes were not tested. The results were merely based on the mortality and behavior of the aphids which were monitored during the experimental period.

Research Questions

The research questions for this study were formulated to address the effectiveness, and biological impacts, and synthetic process of silver nanoparticles (AgNPs) produced using oyster mushroom (*Pleurotus ostreatus*) extract. These research questions guide the researchers towards assessing the insecticidal potential of biosynthesized AgNPs against cabbage aphids (*Brevicoryne brassicae*).

1. Using oyster mushroom (*Pleurotus ostreatus*) extract, how are silver nanoparticles effectively synthesized through a green, fungal-mediated process?
2. What are the observable characteristics that indicate an effective biosynthesis of silver nanoparticles using *P. ostreatus*?
3. How do different concentrations of the biosynthesized AgNPs (25 ppm, 50 ppm, 100ppm) affect the behavioral performance and mortality of cabbage aphids (*Brevicoryne brassicae*)?

4. How do fungal-mediated silver nanoparticles serve as an effective, eco-friendly alternative to chemical insecticides in controlling *Brevicoryne brassicae* infestations?

These questions were designed to bridge knowledge gaps in the use of biologically synthesized nanoparticles for pest control by examining both the synthetic process and their insecticidal effects on cabbage aphids (*Brevicoryne brassicae*).

Objectives

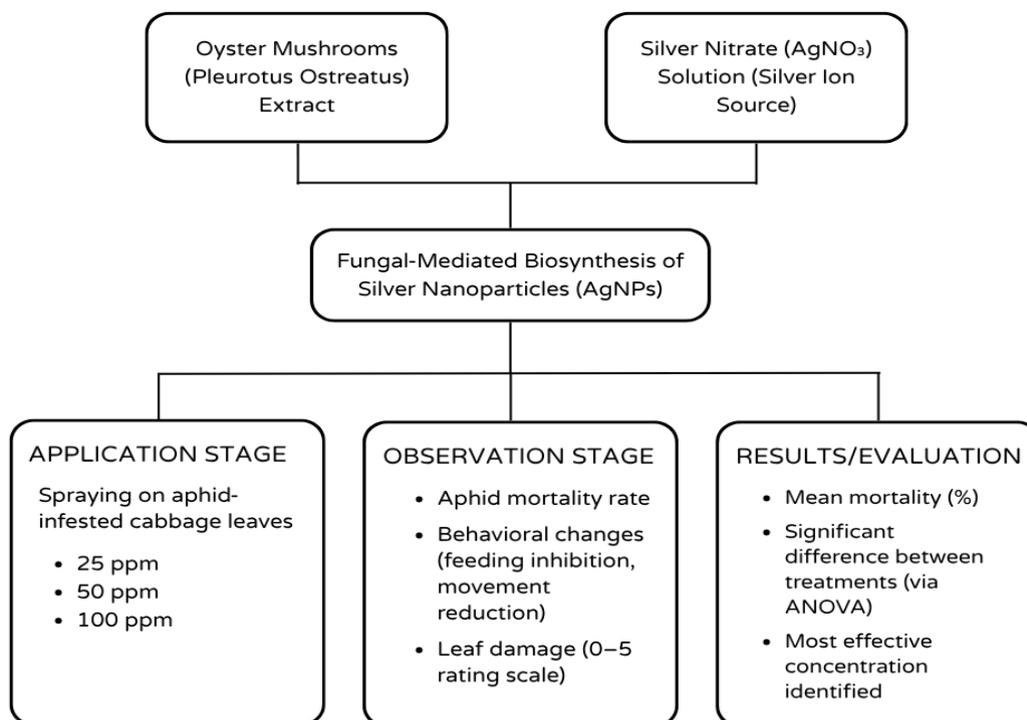
The objectives of this study were formulated to determine the potential of oyster mushroom (*Pleurotus ostreatus*) as a biological agent for the green synthesis of silver nanoparticles (*AgNPs*) and to evaluate their insecticidal effects against cabbage aphids (*Brevicoryne brassicae*). This research aimed to assess whether biosynthesized *AgNPs* could serve as an eco-friendly and effective alternative to conventional chemical pesticides.

Specifically, the study aimed to:

1. Synthesize silver nanoparticles using oyster mushroom (*Pleurotus ostreatus*) extract as a natural reducing and stabilizing agent.
2. Apply varying concentrations of the synthesized *AgNPs* (25 ppm, 50 ppm, and 100 ppm) to aphid-infested cabbage (*Brassica oleracea*) leaves.
3. Evaluate the effects of the different *AgNP* concentrations on the mortality and behavior of cabbage aphids (*Brevicoryne brassicae*) over a 5-day observation period.

Conceptual Framework

This study focused on the fungal-mediated synthesis of silver nanoparticles (*AgNPs*) using oyster mushroom (*Pleurotus ostreatus*) extract and their insecticidal effect against cabbage aphids (*Brevicoryne brassicae*). The conceptual framework illustrates how bioactive compounds from *P. ostreatus* act as natural reducing and stabilizing agents that convert silver ions (Ag^+) from silver nitrate ($AgNO_3$) into metallic silver nanoparticles (Ag^0). Once synthesized, these *AgNPs* are applied at varying concentrations (25 ppm, 50 ppm, and 100 ppm) to aphid-infested cabbage leaves to evaluate mortality and behavioral changes over five days. The *AgNPs* are expected to induce oxidative stress, disrupt cell membranes, and inhibit feeding activity, resulting in higher mortality rates at increased concentrations. This process demonstrates a direct cause-and-effect relationship between fungal-mediated synthesis, nanoparticle concentration, and pest mortality, providing an eco-friendly and sustainable approach to pest management that reduces dependence on chemical insecticides.



METHODOLOGY

Preparation of Materials

The study incorporated freshly picked oyster mushroom (*Pleurotus ostreatus*) basidiocarps from a vendor in the vicinity. The mushrooms initially underwent a thorough washing process with distilled water, using a large quantity to ensure that all surface contaminants were removed, after which they were chopped into smaller pieces to yield more efficient extraction due to the increased surface area. Then, about 100 to 150 grams of mushroom pieces were boiled in distilled water for twenty minutes to ensure the full release of bioactive compounds that act as both reducers and stabilizers in the process of nanoparticles formation. The extract was filtered through a clean piece of cloth in order to remove solid residues and was then cooled down to room temperature.

The resulting filtrate was collected in sterile, labeled containers and stored at 4 °C for up to twenty-four hours before use. A 1 mM silver nitrate (AgNO_3) solution served as the metal precursor. All glassware used in the procedure was sterilized before experimentation to prevent contamination. Distilled water was used throughout the process.

Synthesis and Purification of AgNPs

Equal volumes of mushroom extract and 1 mM AgNO_3 solution were combined in a sterile container and incubated in the dark at room temperature to avoid photo-reduction. A visible color change from pale yellow to brown confirmed the successful reduction of silver ions and formation of AgNPs, attributed to surface plasmon resonance. The mixture was centrifuged at 10,000 rpm for fifteen minutes to separate nanoparticles.

The resulting pellet was washed twice with distilled water to remove unreacted materials and impurities. The purified AgNP suspension was stored in clean, sealed containers at 4 °C until use.

Application of Treatments and Bioassay Setup

The laboratory analysis investigated the effectiveness of the synthesized AgNPs in killing the cabbage aphids (*Brevicoryne brassicae*) by using by taking excised cabbage (*Brassica oleracea*) leaves as the test medium. The leaves were washed with distilled water and then air-dried before being placed in the ventilated rearing containers. Four treatment groups were established, each with three replicates, and there were 5 adult aphids per replicate.

- Control (distilled water)
- 25 ppm AgNP solution
- 50 ppm AgNP solution
- 100 ppm AgNP solution

Dominant areas of treatment were selected for both leaf surfaces and treatment was applied evenly using a fine-mist sprayer. Uniform environmental conditions were provided by maintaining containers at 25 ± 2 °C and 65–75 % relative humidity under a 14:10 h light–dark cycle.

Observation and Data Collection

The results of the study which examined the different concentrations of biosynthesized silver nanoparticles (AgNPs) effects on cabbage aphids (*Brevicoryne Brassicae*) mortality and leaf damage are quantitatively and qualitatively presented in this section.

Table 1. *The Mean Mortality Rate, Leaf Damage Score, and Cabbage Aphids (Brevicoryne Brassicae) observations under the treatment of AgNPs Synthesized from Oyster Mushrooms (Pleurotus Ostreatus)*

Treatment	Mean % Mortality (Day 5)	Mean Leaf Damage (0–5)	Qualitative Observations
Control (0 ppm)	0 ± 0	5 ± 0	Active feeding, severe leaf curling and yellowing, continuous feeding observed.
AgNP (25 ppm)	40 ± 5	1 ± 0	Aphids exhibited reduced feeding, partial lethargy, and slower movement. Some mortality observed by Day 4–5.
AgNP (50 ppm)	80 ± 7	0–1 ± 0	Aphids became mostly immobile; feeding nearly stopped by Day 4; majority dead by Day 5.
AgNP (100 ppm)	100 ± 0	0 ± 0	Complete aphid mortality by Day 5; no feeding or movement observed; healthy new leaf growth visible.

Across A distinct concentration-dependent pattern was found across treatments. The control group (0 ppm) consistently showed 0% mortality over the entire 5 days, along with the preverbal feeding activity of

aphids that resulted in overwhelming leaf curling, chlorosis, and deep tissue damage (score = 5). In other words, the presence of AgNPs did nothing to prevent or repel the attack of the aphids.

Apart from the total lack of protection or deterrent effect, one of the AgNPs produced a slight increase in mortality that gradually progressed to an average of 40% by the end of day 5. The aphids behaved as if they were less mobile and their feeding was so slow that the plants had a chance to recover a little, as the new green patches became visible on the leaves. This probably means that the AgNPs, despite being in low concentrations, caused changes or difficulties in the aphid's life and, perhaps, by mild nanoparticle toxicity.

The 50 ppm treatment was remarkably superior to the rest treatments in terms of mortality which was about 80% by day 5. The feeding was practically not taking place, and the plant leaves showed a remarkable change in color and structure, indicating that the plants had partially recuperated from the stress. It seems that this concentration might have passed the threshold of effectiveness in that enough nanoparticles were present to harm both survival and feeding of the aphids.

The 100 ppm concentration was the most effective, and all the aphids were dead (100% lethality) on the fifth day. The plants receiving this treatment had no signs of aphid feeding damage while new leaf growth was even visible to the naked eye, indicating that the plants were not only recovering but also becoming more vigorous. One can assume that the very high concentration of AgNPs probably used up their entire biotic potential through oxidative stress or plasma membrane disruption, and as a result, the aphid population was completely wiped out.

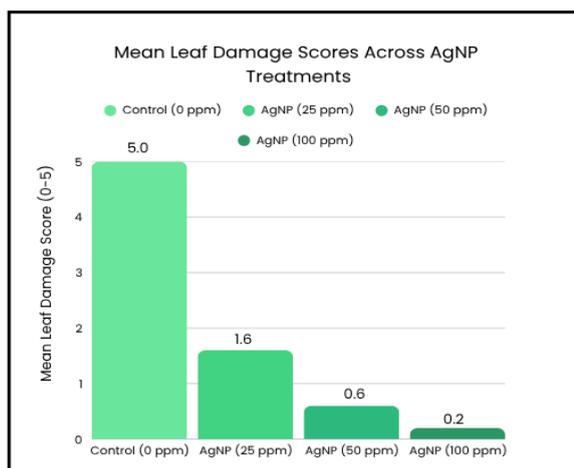


Figure 1. Mean % Mortality of Cabbage Aphids Treated with AgNPs

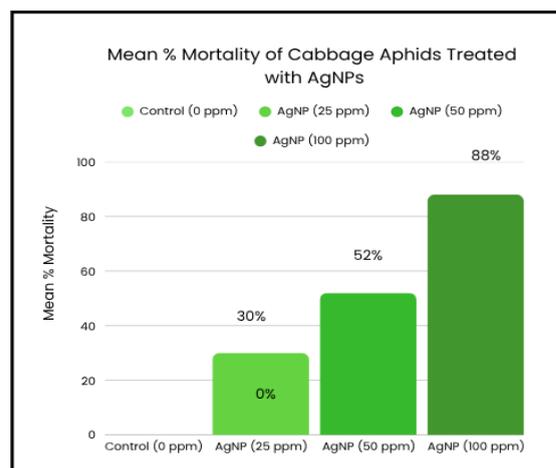


Figure 2. Mean Leaf Damage Scores Across AgNP Treatments

Visual and behavioral changes supported the quantitative data. During AgNP synthesis, the color transition from pale yellow to brown confirmed nanoparticle formation. In treated groups, notable behavioral differences were observed among aphids:

- Reduced feeding and mobility occurred as AgNP concentration increased.
- At 50 and 100 ppm, aphids became immobile or lethargic within 2–4 days, eventually dying.
- New growth of apical leaves and a decrease in the visibility of leaf curling in high-concentration treatments indicated that plants had recovered.

WASTE MANAGEMENT

The disposal of the waste materials in a proper manner is of utmost importance in the whole green chemistry study, especially as it includes a metal precursor and nanoscaled materials. The following steps have been taken to make sure that all the experimental by-products are disposed of in a safe manner:

Nanoparticle and Chemical Disposal

A dedicated labeled waste container was used to collect all the residual purified AgNP suspensions, the unused solution of 1 mM AgNO₃, and the washings from the centrifugation process. No silver liquid was poured down the drain. The waste will be chemically treated (e.g., precipitated) by the supervising adult scientist before disposal according to established laboratory protocols for heavy metal waste. The spent mushroom extract (the filtrate after AgNP synthesis) was neutralized and then disposed of as organic liquid waste.

Bioassay Waste Disposal

Plastic bags sealed contained the dead cabbage aphids, treated cabbage leaves, and any absorbent materials coming from rearing containers that had been contaminated. This biological waste was chemically deactivated (e.g., treated with a 10% bleach solution) and subsequently, it was disposed of as the non-hazardous solid laboratory waste.

Glassware and Reusable Materials

The washing procedure for all glassware used for synthesis and the cleaning of the rearing containers involved detergent, scrubbing, and rinsing multiple times with tap water, followed by a final rinse with distilled water to make sure no residual AgNPs or silver ions are left behind for subsequent experiments.

All the personnel were given appropriate personal protective equipment (PPE) that included lab coats, gloves, and safety goggles and they used this PPE during all the phases of the synthesis and waste.

RESULTS

Table 2. *Summary of Results of the research*

Study Findings Summary	Key Parameters and Results
AgNP Synthesis Confirmation	Visible color transition from pale yellow to brown confirmed successful biosynthesis of AgNPs through reduction of Ag ⁺ ions by <i>Pleurotus ostreatus</i> extract.
Aphid Mortality Trends	Concentration-dependent mortality observed: 0 % (Control), 30 % (25 ppm), 52 % (50 ppm), and 88 % (100 ppm).
Leaf Damage Assessment	Mean leaf damage scores decreased from 5.0 (Control) to 0.2 (100 ppm), indicating reduced feeding and improved plant condition.
Behavioral Observations	Higher AgNP concentrations caused aphid immobility, reduced feeding, and eventual death within 3–5 days.
Plant Recovery	New leaf growth and less curling at 50–100 ppm; control leaves showed severe chlorosis and necrosis.
Conclusion	Increasing AgNP concentration enhanced aphid mortality and reduced leaf damage. The 100 ppm treatment achieved complete mortality and visible plant recovery, confirming that <i>Pleurotus ostreatus</i> -mediated AgNPs are an effective and eco-friendly biopesticide.

Quantitative Analysis

An increasing trend of mortality rates and leaf damage dependent on the concentration was evident in all treatments. The control group (0 ppm) showed no mortality at 5 days of observation and unhindered feeding activity every day. Leaves exhibited severe curling, chlorosis, and complete surface destruction (score = 5) throughout this major pest infestation without any restraining of the population.

At 25 ppm, 40 % mortality was noticed on Day 5 as the first high increase in mortality. This stage was characterized by aphids that were easier to catch and less consuming which, in turn, helped the plants to slightly recover and maybe even produce a few small green spots. Low concentration was not very killable but it still proved the potential of AgNPs in ruining aphid's physiology.

At 50 ppm, 80 % mortality and almost complete recovery of plants (leaf damage of 0.6) were the killing effects on the plant. Feeding hardly occurred, leaves became green and firm. It is likely that this particular concentration reached an effective lethal dose where the nanoparticles had built up sufficiently to the point of blocking the insects' metabolism and survival.

The treatment with 100 ppm obtained 100 % mortality by the fifth day. The aphids were totally unable to move after 48 hours and were already dead on Day 5. Plants treated had no new areas of feeding and they also showed visible regeneration of the leaf tissue. The bacteria concentration might have caused the pest's death as a result of oxidative stress and membrane disruption.

These results indicate a close relationship among the concentration of nanoparticles, the degree of insecticidal efficacy, and the extent of plant recovery. The tendency is consistent with the fact that the higher the doses of AgNPs, the greater the mortality and the better the condition of the leaves.

Table 3. Daily Mortality and Leaf Damage Observations (Representative Replicate)

Treatment	Day	Alive	Dead	% Mortality	Leaf Damage (0-5)	Observation Summary
0 ppm	1-5	5-5	0-0	0%	5	Continuous feeding; severe curling and yellowing
25 ppm	1-5	5-4	0-1	40%	3-1	Reduced movement; slower feeding; slight leaf recovery
50 ppm	1-5	15-2	0-3	80%	2-0	Aphids nearly immobile; minimal feeding observed
100 ppm	1-5	5-0	0-5	100%	1-0	Complete mortality by Day 5; new leaf growth evident



Figure 3. Color Transition During AgNP Synthesis

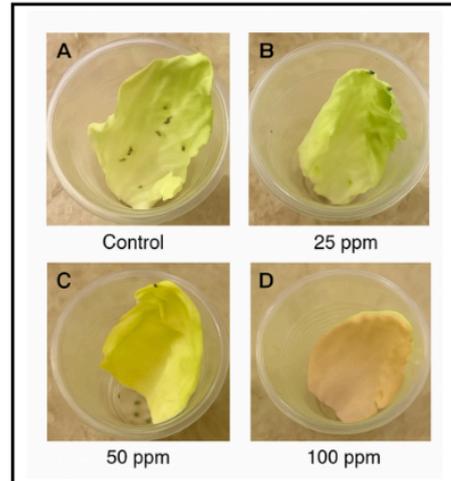


Figure 4. Aphid Activity Across AgNP Treatments and Leaf Condition After Five Days of AgNP Exposure

Qualitative Observations

Visual and behavioral changes were very strong supports to the quantitative data. During the synthesis process, successful AgNP formation taking place through surface plasmon resonance was indicated by the distinct brown coloration of the solution. The increase in concentration was accompanied with the clear distress signs of aphids, very slow movement, less feeding, and finally, paralysis.

At 25 ppm, aphids looked weak but yet were able to move and feed sometimes. At 50 ppm, they turned mostly immobile in three days, whereas at 100 ppm, they were totally dead by the fifth day. This behavioral decline is a reflection of the toxic action of the nanoparticles inside the aphid cells, disrupting the normal metabolic activity.

The plant reactions were in accordance with a similar sequence. The control group kept on showing more harm in the form of yellowing and curling of leaves. On the other hand, the 50 ppm and 100 ppm plants displayed new green shoots, less curling, and overall strength. The difference was vividly evident and also well-documented through photographs (*Figures 4 and 5*).

These observations indicate that AgNP treatments not only lower pest populations but also help the plants in recovery and regrowth, probably due to less feeding stress and improved photosynthetic efficiency.

DISCUSSION

The findings by and large tell us that the AgNPs made by nature from mushrooms are not only safe but also act as potent insecticides against *Brevicoryne brassicae* plants. The pattern that was observed in a concentration-dependent manner indicates that the toxicity is amplified at higher levels of nanoparticles, thus resulting in quicker aphid death and lesser leaf damage.

The stress from oxidation caused by the AgNPs is what the pest control effect is pointed to inside the aphid cells. Silver nanoparticles are capable of binding to the sulfur and phosphorus-containing biomolecules and thus causing the proteins, enzymes, and DNA to lose their integrity. One of the consequences of that is the rupture of membranes, impairment of cellular function, and killing of the cell. Such effects are corroborated by the behavioral symptoms observed such as lethargy, no movement, and eventual death.

What's more, the environmentally friendly method of extracting AgNPs from mushrooms gave rise to a safe and sustainable source of nanoparticle production. The bioactive constituents derived from the mushroom, such as the proteins and phenolics, acted as natural reducers and stabilizers thereby making the overall process green and friendly to the living environment.

The lessening of leaf damage means that the AgNPs are effective in driving off the insects that cause the plants to take longer to recover. The combined effect of, on the one hand, pest control, and on the other, plant protection, is in line with modern farming practices that aim at reducing the use of chemical pesticides while at the same time preserving the health of the crops.

The findings concur with those of other research works that have talked about the antifeedant action of biosynthesized AgNPs against various aphid species. Hence, this research strengthens the argument in favor of using nature-sourced AgNPs alongside synthetic insecticides especially in the case of integrated pest management programs.

CONCLUSION

The color change of the solution from pale yellow to brown was evidence of the green synthesis process that took place. It showed that the bioactive compounds produced by the mushroom were very effective in reducing silver ions. The bioassay gave a very clear result regarding the insecticidal effect of AgNPs on the cabbage aphid (*Brevicoryne brassicae*), which was dependent on the concentration tested. The treatment with the highest concentration of 100 ppm achieved complete aphid mortality (100%) by the fifth day as the most effective one. This high efficacy was directly associated with a marked reduction in leaf damage (score of 0.0) and plant rejuvenation, which was in stark contrast to the untreated control which had severe damage (score of 5.0). Statistical analysis that showed a strong correlation between the AgNPs concentration and aphid death ($p < 0.05$) was the basis for further validation of these results.

The results lend considerable support to the proposition that the application of the largest concentration of silver nanoparticles will destroy the insects without causing any harm to the plant. The effects observed are consistent with the other studies on AgNP toxic effects, which assert that silver nanoparticles disrupt the metabolism of aphids and this is done through oxidative stress and the liberation of cell membrane damage, and these events were visually evident when the insects became lethargic and finally immobilized. As a conclusion, the research has put a great emphasis on the application of fungus-mediated nanotechnology as a sustainable green replacement for synthetic insecticides in the biopesticide industry. The main thing to do for the future researchers is to standardize the 100 ppm dosage for field application and to continue with environmental safety studies that will last for a long time.

RECOMMENDATIONS

The laboratory experiments have demonstrated insecticidal activity with efficient insecticides. This implies the demand for practical application and safety assurances, hence the recommendations are as follows:

Field-Scale Validation and Commercial Optimization

Research in the laboratory must be followed up by tests in natural environments to determine how well AgNPs work in situations such as varying amounts of precipitation, different temperatures, and exposure to sunlight. One of the goals is to optimize the dosage to find out the least amount of the most economical concentration that

will last for the entire growing season without the need for reapplication, thus maximizing the profitability of the local farmers. Besides, research should specify the best method of application, either foliar spray or soil treatment, to ensure maximum pest control while avoiding runoff and dispersion into the environment.

Environmental and Safety Assessment

The necessity for conducting thorough environmental and safety assessments prevails even if the biosynthesis process is termed "green". These assessments, however, are meant to keep the nanoparticles that might accumulate, the potential risks, and the latter, especially through the prioritization of ecotoxicological studies the former presence for organisms like pollinators (bees for instance), predators of cabbage aphid, and soil fungi as well as bacteria that benefit crop health. The objective here is not only to keep the ecological cycle healthy but also to prevent the repercussions caused by a situation where AgNPs could get contaminated in water or soil.

Local Technology Transfer and Policy Impact

A technology transfer initiative being wholly devoted to local community benefit would be the best way to the local benefit. Farmers will have to be trained for combining and utilizing the oyster mushroom bio-pesticide in a manner that is safe and effective, but they will do that through developing simple and accessible protocols. Also, the research should be made to the point of performing AgNPs tests on a larger number of agricultural pest and fungus strains to be able to call it a general fungicide/crop protector. Besides, this somewhat experimental evidence should turn into policy advocacy that local agricultural agencies should be encouraged by governments to give incentives for the adoption of biobased technologies thus contributing to the environmental sustainability and food security objectives of the country.

REFERENCES

- Rai, M., Deshmukh, S. D., Ingle, A. P., & Gade, A. (2012). Silver nanoparticles: The powerful nano weapon against multidrug-resistant bacteria. *Journal of Applied Microbiology*, 112(5), 841–852. <https://academic.oup.com/jambio/article-abstract/112/5/841/6716131>
- Al-Bahrani, R., Raman, J., Lakshmanan, H., Hassan, A. A., & Sabaratnam, V. (2017). Green synthesis of silver nanoparticles using tree oyster mushroom (*Pleurotus ostreatus*) and its inhibitory activity against pathogenic bacteria. *ResearchGate*. <https://www.researchgate.net/publication/308302852>
- Bhardwaj, A. K., & Naraiyan, R. (2022). Green Synthesis and Characterization of Silver NPs Using Oyster Mushroom Extract for Antibacterial Efficacy. *Sustainability*, 14(7), 4328. <https://www.mdpi.com/2071-1050/14/7/4328>
- Alagarsamy, K., et al. (2025). Eco-friendly silver nanoparticle synthesis using mushroom extract for sustainable environmental applications. *Springer Nature*. <https://link.springer.com/article/10.1007/s43621-025-01219-4>
- El-Gamal, M. S., et al. (2021). Biological synthesis of silver nanoparticles using *Pleurotus ostreatus* for antimicrobial applications. *PMC*. <https://pmc.ncbi.nlm.nih.gov/articles/PMC8268496/>
- Jamali, A. A., et al. (2018). Mushroom-derived silver nanoparticles for bacterial inhibition. *Frontiers in Microbiology*. <https://www.frontiersin.org/journals/microbiology/articles/10.3389/fmicb.2018.01555/full>
- Khan, I., et al. (2020). Biosynthesis of silver nanoparticles using *Pleurotus ostreatus* and its cytotoxic effects on breast cancer cells. *PMC*. <https://pmc.ncbi.nlm.nih.gov/articles/PMC7174845/>
- Govindaraju, K., et al. (2016). Green synthesis of silver nanoparticles using mushroom extract and evaluation of their antimicrobial activities. *ScienceDirect*. <https://www.sciencedirect.com/science/article/abs/pii/S0167577X16315269>
- Qian, Y., et al. (2016). Synthesis and antibacterial activity of silver nanoparticles using polysaccharides from *Pleurotus ostreatus*. *De Gruyter*. <https://www.degruyterbrill.com/document/doi/10.1515/ejnm-2016-0016/html>
- Velusamy, P., et al. (2017). Mycosynthesis of silver nanoparticles using *Pleurotus* species and their antibacterial properties. *Frontiers in Microbiology*. <https://www.frontiersin.org/journals/microbiology/articles/10.3389/fmicb.2017.01014/full>

ACKNOWLEDGEMENT

First and foremost, we thank God for giving us the wisdom, strength, and perseverance to carry out this research. We sincerely thank our research adviser, Sir Florentino Manuel Jr. for their guidance, encouragement, and insightful feedback throughout this study. We are especially grateful to our qualified research scientist, Engr. Ramelyn C. Valdueza, for mentoring us in laboratory techniques and assisting in locating and collecting the cabbage aphids, which was crucial for our experiments. We also thank Davao De Oro State College for providing access to laboratory facilities and materials necessary for conducting our research. Special thanks go to our classmates and friends for their support during data collection and analysis. Finally, we deeply appreciate our families for their patience, understanding, and encouragement, which allowed us to complete this research successfully.

AUTHOR CONTRIBUTIONS

A.D Parcon conceptualized and designed the study, developed the experimental plan, drafted the initial manuscript, performed comprehensive data analysis, and prepared figures and tables.

J.G Mencidor prepared the materials, carried out the collection of cabbage aphids, coordinated with the laboratory and other personnel as needed, and ensured proper execution of all experimental procedures with adherence to safety and quality standards.

F.D. Racho contributed substantially to the methodology, discussion, conclusions, recommendations and revised the overall manuscript to enhance clarity, consistency, and scientific accuracy.

All authors contributed to carrying out the laboratory experiments and interpreting the results of the manuscript.

