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Green Synthesis of Copper Nanoparticles using *Ananas comosus* Waste and Their Antimicrobial Activity: A Review

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Abstract

A. comosus (*A. comosus*) is widely cultivated in Malaysia and abundant of plant waste was created from the harvesting process. Up until today, exploration has focused on the optimum utilisation of *A. comosus* plant waste was minimally reported. This review extensively revealed the waste extract of *A. comosus* phytochemical compounds that can potentially act as reducing and stabilising agents for copper nanoparticles (CuNPs) via green synthesis methods. This review also provided information on the antimicrobial activity of the synthesised CuNPs. The information gathered in this review will contribute towards further development of copper nanoparticles synthesised using a green approach.

Keywords: *A. comosus*, antimicrobial, Copper nanoparticles, Waste, Green synthesis

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1. Introduction

Due to the fact that waste frequently results in microbial deterioration and major environmental issues, it is a developing concern and exemplifies the significant issue of disposing of *A. comosus* waste. If not handled properly, it will further contribute to environmental degradation. The waste produced by the *A. comosus* processing business is enormous [1]. Idealising *A. comosus* waste through further processing until it is transformed into lucrative products using environmentally friendly techniques is both a challenge and an opportunity [1]. In addition, the green synthesis of nanoparticles is now a focus of research in order to get vital goods and also to substantially minimise or abolish the waste products created [2]. Orange peel extract was one of the waste products used in the synthesis of silver nanoparticles [3]. The average size of the silver nanoparticles (AgNPs) detected in transmission electron images were determined to be roughly 15 nm in another study that utilised dry grass as a waste material [4]. Additionally, onion peel has also been used to create metallic nanoparticles like zinc oxide nanoparticles (ZnONPs) [5]. Since the study of the green synthesis method is increasing widely due to its wide applications and the significant advantages of the technique used, the degree of effectiveness of nanoparticles mediated by *A. comosus* waste extract can be tested for various microorganisms. The significance of this research is that it is able to overcome the abundance of *A. comosus* waste in Malaysia by using it as a renewable source. In addition, this study carries out the latest research design on the green synthesis of nanoparticles from *A. comosus* waste extract that has a big potential as a good antimicrobial agent against common pathogenic bacteria and fungi in plants and crops in Malaysia, respectively.

2. *Ananas comosus*

A. comosus is a member of the Bromeliaceae family, a big and varied family with around 2000 species [6]. It has a short stem and thin, rigid leaves that develop into fruits that range in size from medium to enormous. After mangos and bananas, it is the third most widely grown tropical fruit and the largest producer. Pineapple features a majestic crown of spiky, blue-green leaves and a broad, cylindrical form. Its skin is crusty and green, brown, or yellow. The fibrous flesh of *A. comosus* has a bright, tropical taste that blends the tastes of sweet and sour and is yellow in colour [1]. *A. comosus* is one of the fruit varieties with the most potential for growth in Malaysia's domestic and international markets, where 70% of the pineapple that was produced was eaten as fresh fruit [7]. It constitutes one of the most widely farmed fruits, yielding approximately 352,000 tonnes annually, with approximately 52000 tonnes traded each year and a substantial number of postharvest losses observed [8]. It also seems more reliable as Malaysia increases the size of its pineapple plantations in 2018 [9].

Over 17,601 hectares of *A. comosus* have been planted in Malaysia since about 2018. Johor, which represents 57% of all pineapple crops (8,934.53 hectares), is the state with the greatest pineapple crops, trailed by Sabah (988 hectares) and Sarawak (972.48 hectares) [10]. This encouraging rise demonstrates the potential for fresh pineapple on the global market [7]. *A. comosus* is a subtropical fruit that is indigenous to Thailand, the Philippines, China, Brazil, and India [6]. They are native to warm areas in the Americas and are the leading producers of *A. comosus* [7]. The global production of tropical fruits, particularly *A. comosus* has been seeing a notable increase. This growth can be attributed to the attractive market prospects that exist on a global scale, driven by the strong demand for tropical fruits and the growing knowledge of their nutritional benefits. The countries of the Philippines, Costa Rica, Brazil, Indonesia, and China collectively contributed to a global production of 28 million metric tonnes of the aforementioned fruit [11].

3. Sustainability of *Ananas comosus*

The global yield of *A. comosus* is 25.4 million metric tonnes. Consequently, the waste grows in relation as pineapple output rises. *A. comosus* primarily has its waste in the form of a crown, peel, stem, and core [1], as shown in **Figures 1 and 2**. Pineapple wastes can harm the environment if they are not properly treated [12]. Due to its high moisture and sugar content as well as its susceptibility to microbial deterioration, this waste disposal can be troublesome and release harmful gases including hydrogen (H_2), carbon dioxide (CO_2), and methane (CH_4) [13]. This may have occurred as a result of how agricultural wastes affect the amount of time that crop residue has to degrade aerobically, while the cellulose found in natural substrates is persistent in the environment and continues to be a contaminant [14]. Many studies have found that this organic solid waste is rich with bioactive chemicals with a variety of uses [15]. Utilising *A. comosus* waste into profitable merchandise was already acknowledged as it offers the most environmentally friendly technique and is also attributed to its functional components and characteristics [16].

4. Nanotechnology and nanoparticles

Nanotechnology and nanoparticles are a growing area of nanotechnology where nanoscale particles become more sensitive when compared to their original counterparts [17]. It is concerned with the production method and handling of particles with a size range of 1 to 100 nm [18]. In addition to many other characteristics like size, shape, surface charge, chemical structure, surface area, and coagulation properties of different nanoscale materials, the catalytic potential, electrical conductivity, optical sensitivity, magnetic behaviour, or biological reactivity are used to characterise the chemical, physical, and biological properties of nanomaterials [19].

5. Role of plants in green synthesis of nanoparticles

Given the non-pathogenic nature and extensive investigation into several routes, plants are thought to be more suited for green nanoparticle generation [18]. The generation of clean and ecologically acceptable nanoparticles employing plants is known as green synthesis and applies the widely established green chemistry idea [20]. Plants are recognised as low-maintenance, cost-effective chemical factories of nature and have kinetics that are significantly greater than those of other biosynthetic methods that seem to be comparable to the chemical phytochemicals that different plant sections such as fruit, leaves, stems, and roots generate. They have now been extensively exploited for the green production of nanoparticles [20]. Nanoparticle (NP) production from plants is undoubtedly a simple process. A metal salt is created using plant extract, and the formation of nanoparticles could be completed in a salt solution within a short period of time, depending on the nature and properties of the plant extracts [17].

The plant extract functions as reducing and stabilising agents for green synthesis due to the presence of various secondary metabolites, including phenolic acid, flavonoids, alkaloids, and terpenoids, which continuously participate in a redox reaction to create environmentally acceptable and unique metallic nanosized particles [21]. In addition, it has proven possible to create a wide range of metal nanoparticles utilising various plants [18]. Even though extremely small amounts of heavy metals are harmful at really trace levels, plants have shown the opportunity of detoxifying heavy metals as well as aggregating them, by which the environmental pollution problem may be solved [20]. The ability of plants to survive in situations with high metal concentrations has improved where they can modify the chemistry of the harmful metals, reducing or eliminating their toxicity [18]. Many greener nanoparticles, including cobalt, copper, silver, gold, palladium, platinum, zinc oxide, and magnetite, are effectively synthesised using plants [21]. These nanoparticles exhibit special optical, thermal, magnetic, physical, chemical, and electrical characteristics that have a wide range of uses in a diversity of human-interest sectors [18].

6. Green synthesis of nanoparticles (NPs) using *Ananas comosus* waste extract and their antimicrobial activity

One of the *A. comosus* waste which is leaves result in the formation of silver nanoparticles (AgNPs) from its extract [22]. The characterization illuminates the existence of benzophenones, anthocyanins phenol, flavonoid groups, amide, amine, ethers, alcohols and polyphenols present in the *A. comosus* leaf extract. The FTIR analysis from this study reveals that phenolic compounds are involved in the reduction of Au^{3+} to Au^0 and the stabilisation processes [22]. The existence of the functional groups of OH (hydroxyl) and C=O (I amide) plays a role in the reduction process and results in the formation of AgNPs [23]. Other than leaves, the other waste part of the *A. comosus* also contributed to the stabilisation of metallic nanoparticles such as peel extract, fruit extract, and waste pericarp to synthesise silver nanoparticles [2], gold nanoparticles, and zinc oxide nanoparticles, respectively [24-25]. Many parts of *A. comosus* waste has been successfully used for nanoparticle green synthesis for antimicrobial testing. The study of *A. comosus* peel extracts for the synthesis of silver nanoparticles have been reported for their antibacterial activities [2]. The usage of *A. comosus* leaf extracts for the synthesis of gold nanoparticles [22] and silver gold nanoparticles have also been reported for their antibacterial activity. In addition, zinc oxide nanoparticles from the peel extract of *A. comosus* shows antifungal activity, while copper nanoparticles are synthesised from the fruit extract of *A. comosus* shows both antibacterial and antifungal activity [25-26]. **Table 1** summarises the overall green synthesised nanoparticle studies using *A. comosus* waste part extracts and their antimicrobial activity.

7. Green synthesis of copper nanoparticles (CuNPs) using plant extract and their antimicrobial activity

Green synthesis of copper nanoparticles (CuNPs) using plant extract and their antimicrobial activity: given the ease of synthesis, low cost, and potential for utilisation, metal nanoparticles stand out among the other types of nanoparticles. Due to its numerous benefits, copper nanoparticle synthesis is of tremendous interest [6]. In addition to being reasonably safe for people, soluble copper compounds have also been proven to have strong antimicrobial action against a variety of pathogens, including bacteria and fungus, where they seem to kill by producing reactive hydroxyl radicals that can destroy cells permanently by oxidising proteins, cleaving DNA and RNA, and damaging membranes from lipid peroxidation [27]. Besides, copper is the most utilised material around the world as it is more affordable than silver and gold, has a high conductivity, and has significant uses in electrical, optical, catalytic, biomedical, and antifungal or antibacterial processes. The high surface-to-volume ratio and strong reactivity of copper nanoparticles result in immediate interaction with other particles [6]. The big concern with copper nanoparticles, nevertheless, is the instant aggregation that results from surface oxidation. Hence, by choosing a good stabiliser for the copper nanoparticle capping, we can get around such issues [6]. The phytocomponents included in aqueous fresh pineapple extract, such as flavonoids, alkaloids, and terpenoids, could account for the reduction and capping of copper ions to copper nanoparticles [6].

The steps are reduction, nucleation, and crystal growth. Reduction occurs because the carbonyl and hydroxyl groups have a higher redox potential than other groups and become the electron source that transfers to metal, which leads to the beginning of crystal nanoparticle formation (nucleation). During the process of crystal formation, the carbonyl and hydroxyl groups will stabilise the nanoparticle crystals formed [28]. In addition, copper nanoparticles perform and behave more effectively than bulk copper particles due to their distinct features. Copper nanoparticles are discovering new applications in agriculture because of their antimicrobial properties and are effective as antibacterial agents against a variety of pathogenic bacteria, including *Escherichia coli, Salmonella, Staphylococcus*, and *Streptococcus mutans*, according to several studies. According to reports, harmful fungi, including *Botrytis cinerea*, *Colletotrichum acutatum*, *Verticillium dahliae*, *Verticillium albo-atrum*, and *Botrytis fabae,* also have some antifungal activity when exposed to copper nanoparticles [29]. Several studies have made use of the CuNPs synthesised from various plants for phytopathogenic bacteria and fungi. These show that CuNPs also have a big potential for synthesis using *A. comosus* extract. **Table 2** shows the characteristics and antimicrobial evaluations of CuNPs using green synthesis from various plants. Different pathogenic bacteria and fungi result in various zones of inhibition. The increase in the concentrations or volume of nanoparticles from *A. comosus* waste extract causes an increase in the zone of inhibition. However, every pathogenic bacterium depends on its own minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC), which means that they react to the minimum antimicrobial concentration that will prevent their proliferation and the least amount of an antibiotic substance necessary to eradicate a certain microorganism, respectively. Meanwhile, every fungus depends on its own minimum fungicidal concentration (MFC), which means the lowest concentration of monoterpenes, resulting in the death of 99.9% of the inoculum. Among all the types of nanoparticles mediated by *A. comosus* waste extract, copper nanoparticles seem to show the best antimicrobial activity. Furthermore, the synthesis of copper nanoparticles also needs to be optimised in terms of the different compositions of copper sulphate to extract, temperatures, pH, and the formation of copper nanoparticles at different intervals of time. The characterization of copper nanoparticles was determined by using UV-visible absorption spectroscopy, powder X-ray diffraction (PXRD), Fourier transform infrared (FTIR), scanning electron microscopy (SEM) [6], and transmission electron microscope (TEM) analysis [30]. The characterization was conducted to analyse certain aspects of nanoparticle mediation. **Table 3** shows the functions of the characterization techniques upon studying nanoparticle properties, while **Table 4** shows a few outcomes for the characterization of nanoparticles using *A. comosus* extract.

Table 1: Green synthesized nanoparticles studies using *A. comosus* plant parts extracts and their antimicrobial activity.

***A. comosus* waste parts**	**Nanoparticles**	**Antibacterial activity**	**Antifungal activity**	**Range of inhibition zone (mm)**	**References**
Peel	Silver	*E. faecium* *L. monocytogenes* *B. cereus* *S. aureus*	-	8.78-10.31	[2]
Leaves	Gold	*E. coli* *B. subtilis*	-	3-16	[22]
Leaves	Silver gold	*S. aureus* *E. coli*	-	7-8.18	[26]
Peel	Zinc Oxide	-	*Aspergillus niger* *Collectotrichum alatae*	6.2-8.2	[25]
Fruit	Copper	*S. aureus* *E. coli*	*Trichoderma viride* *Aspergillus niger*	6-14	[6]

Table 2: The characteristics and antimicrobial evaluations of CuNPs using green synthesis from various plants.

Type of plants	**Size and shape of copper nanoparticles (nm)**	**Antibacterial activity**	**Antifungal activity**	**References**
Persea americana	42–90 (Spherical)	*E. coli* *Streptococcus sp* *Klebsiella sp* *Rhizobacterium*	*Aspergillus flavus* *Aspergillus fumigates* *Fusarium oxysporum*	[33]
C. vitiginea	10−20 (Spherical)	*E. coli* *Enterococcus sp* *Proteus sp* *Klebsiella sp*	-	[34]
Gardenia jasminoides	0.2089 (Spherical)	*S. aureus* *E. coli*	-	[35]
Hagenia abyssinica (Brace) JF. Gmel.	34.76 (spherical, hexagonal, triangular, cylindrical, irregularly)	*E. coli* *P. aeruginosa* *S. aureus* *B. subtilis*	-	[36]
Celastrus paniculatus	2−10 (Spherical)	-	*Fusarium oxysporum*	[30]
Eucalyptus	10-130	-	*Colletotrichum capsici*	[37]
Mint leaves	23-39	-	*Colletotrichum capsici*	[37]

Table 3: The functions of the characterization techniques upon studying nanoparticles properties.

Techniques	Functions
X-Ray Diffraction (XRD)	Crystal structure Composition Crystallize grain size.
UV Visible Absorption Spectrometer	Optical properties Size Concentration Agglomeration state Hints on nanoparticles shape
Fourier Transform Infrared (FTIR)	Surface composition Ligand binding
Scanning Electron Microscopy (SEM)	Morphology Dispersion of nanoparticles in cells and other matrices or supports. Precision in lateral dimensions of nanoparticles Quick examination Elemental composition
Transmission Electron Microscope (TEM)	Nanoparticles size Size mono-dispersity Shape Aggregation state Detect and localize or quantify nanoparticles in matrices. Study growth kinetics

Source: [38]

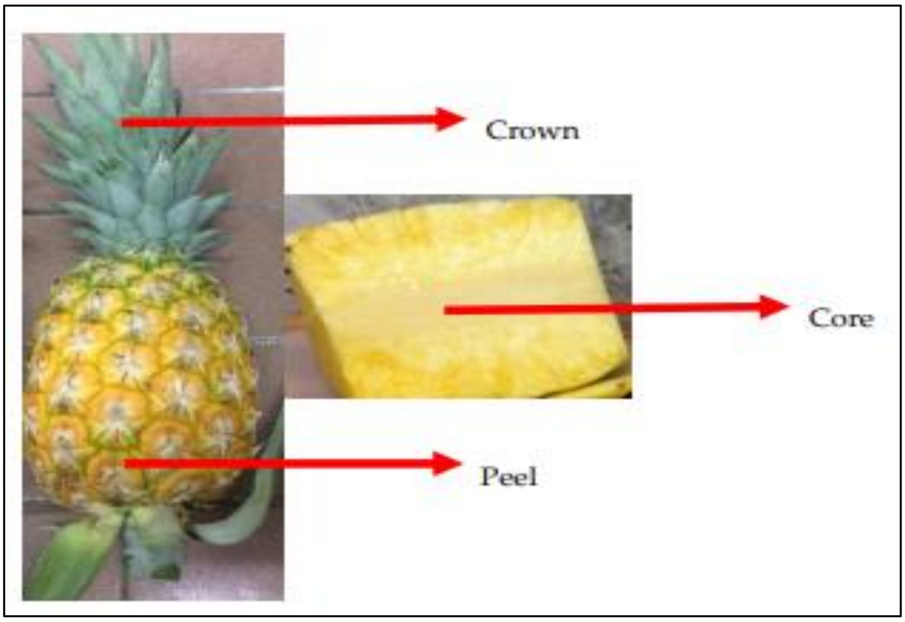

Figure 1: The crown, core and peel of *A. comosus* [31].

Table 4: The characterization of nanoparticles using *A. comosus* extract.

Part of *A.comosus*	Type of nanoparticles	XRD	UV-Vis	FTIR	SEM	TEM	Ref.
Pulp	Silver nanoparticles	Crystalline	Optical absorbance at 200-800 nm	-	-	Size: 12 nm Shape: spherical	[39]
Peel	Silver nanoparticles	-	Absorption at 420 nm	-	Size: 200 nm Shape: spherical/ cube		[40]
Fruit	Copper nanoparticles	Crystalline	Absorption at 576 nm	Aqueous fresh pineapple extract: Phenolic compound, amine, alkyne Synthesized copper nanoparticles: Amide, phenolic compound, amine, alkyne, polyphenols, alkaloids and terpenoids	Size: 30- 50 nm	-	[6]
Leaves	Gold nanoparticles		Absorption at 520 nm	Benzophenones Anthocyanins Flavonoid groups Amide Amine Ethers Alcohols Polyphenols		Size: 20-30 nm Shape: Spherical, triangular, pyramidal	[22]

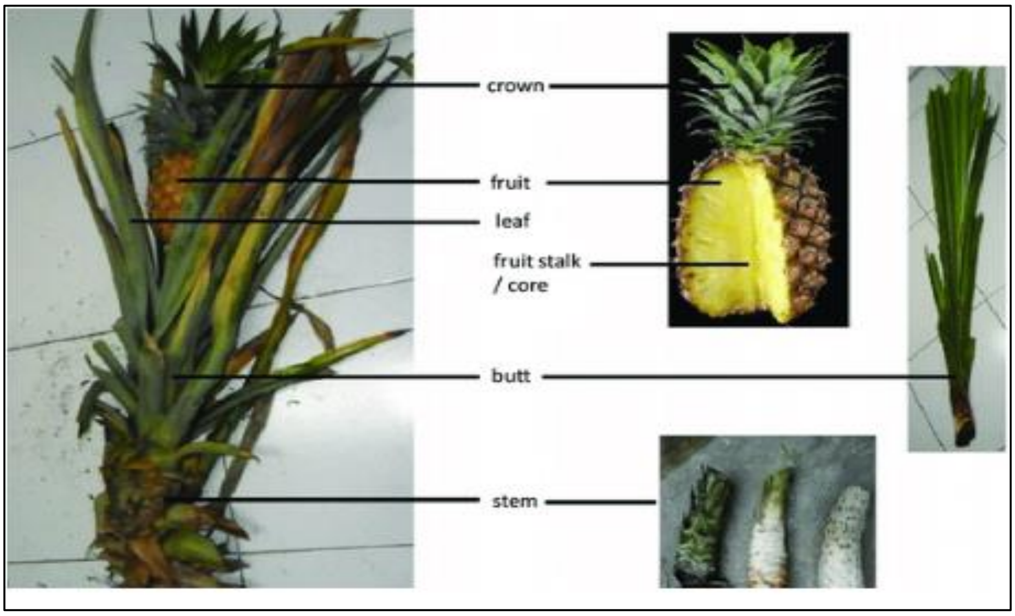

Figure 2: The overall macroscopic visualization of *A. comosus* [32].

8. Conclusion

Since a lot of nanoparticles mediate from *A. comosus* waste extract shows an excellent zone of inhibition, the researchers can enhance their research by focusing on natural remedy to treat antimicrobial activity rather than utilisation of chemical base pesticides in plantation sector which have a lot of disadvantages and can destroy our ecosystem. The great amount and types of bioactive compound in *A. comosus* waste extract is a big support to the stabilization of nanoparticles which the production can be optimized through study of experimental procedure. Thus, the abundance of plant waste not only limited to *A. comosus* can be used as a good alternative of renewable sources to enhance the antimicrobial sectors as it is eco-friendly and economical. This can also ensure the sustainability of plantation sectors in Malaysia and worldwide with the improvement of its management in every aspect.

9. Future direction

Enhancement in nanotechnology, specifically application in plantations and agrotechnology, plays a vital role nowadays since it is one of the biggest sectors worldwide. The development of applications can be extended to other sectors such as the environment, energy sources, medicine, and material science, which mainly comprise natural products. Thus, this is a great novelty in improving the safety of future use.

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