

Phytofabrication green synthesis of silver nanoparticle from *Pontederia crassipes* with antimicrobial and antioxidant potential

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Abstract

Pontederia crassipes (water hyacinth) has been identified as a significant contributor to substantial economic and ecological detriment in tropical aquatic systems and environments. The formidable species is nearly impossible to eliminate, characterized by rapid reproduction that facilitates swift and extensive proliferation, potentially suffocating entire rivers and aquatic ecosystems. Nevertheless, a wide array of bioactive secondary metabolites is present in *P. crassipes*, demonstrating a broad spectrum of antibacterial activities. Water hyacinth leaf extract comprises alkaloids, flavonoids, phenols, glutathione, terpenoids, and saponins, all of which exhibit significant antibacterial properties. This study sought to evaluate the antibacterial efficacy of Water Hyacinth leaf extract, the active phytoconstituents of *P. crassipes*, and the biosynthesis of novel Water Hyacinth AgNPs mediated by phenolic compounds, utilizing antibacterial lyophilized hydro-ethanolic extracts from *P. crassipes* as both bio-reduction and capping agents. *Pontederia crassipes* exhibits significant anti-oxidant action. Water hyacinth is a fast-proliferating aquatic plant species. Specifically, it adversely affects the aquatic ecosystem and biological system. Water hyacinth, however, is abundant in cellulose, a biodegradable substance.

Keyword: *Pontederia crassipes*; Water hyacinth; Antibacterial resistance; Biosynthesis; Silver nanoparticles; Antioxidant

1. Introduction

Pontederia crassipes (water hyacinth) has been single out as one of the major causes of enormous economic and ecological losses to tropical water systems and habitats. Almost impossible to eradicate, the ominous species is characterized by rapid multiplication, giving rapid and extensive spread that can choke entire rivers and water bodies. However, vastly diverse bioactive secondary metabolites are found in *P. crassipes*, which exhibit an expansive assortment of antibacterial properties used in traditional medicine. The rapid emergence of multidrug-resistant (MDR) pathogens to common drugs is a pandemic of global proportions. Global health institutions are calling for a coordinated, global action plan to develop new biomaterials to combat the rising threat by MDR strains, improve treatment outcomes, and save lives. This study proposes consumption of the water hyacinth lily species through a novel material biosynthesis approach not only to manage the aquatic infestation but to develop advanced multifunctional, anti-microbial silver nanoparticles through bio-reduction and capping of silver salts targeted at MDR microbes. Methodology: After screening for active phytoconstituents of *Crassipes* [1].

The Biosynthesis of Frightening forecasts by the UK government-commissioned "Review on Antimicrobial Resistance" forecast that AMR-related fatalities could account for up to 10 million yearly deaths before 2050 [2].

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The antibiotic resistance pandemic has been attributed by most researchers to a number of causes, including the development of antibiotic resistance genes among populations. The careless and inappropriate prescription of drugs abuse and sub-use, as well as the duality use of most antibiotics in humans and agriculture [3].

Based on the phytochemical analysis of water hyacinth leaf extract containing alkaloids, flavonoids, phenols, glutathione, terpenoids, and saponins. Water hyacinth leaf extract is known to show broad spectrum antimicrobial activity against gram-positive and gram-negative bacteria and fungi depending on pH, concentration, and duration of action [4,5].

Research shows that the content of flavonoids, terpenoids, and saponins acts as antibacterial against the activity of the **glycosyltransferase (GTF)** enzyme that plays a role in the formation of biofilms by changing the permeability of the cell membrane. So indirectly, this mechanism is expected to inhibit the attachment of bacteria to host cells [6,7].

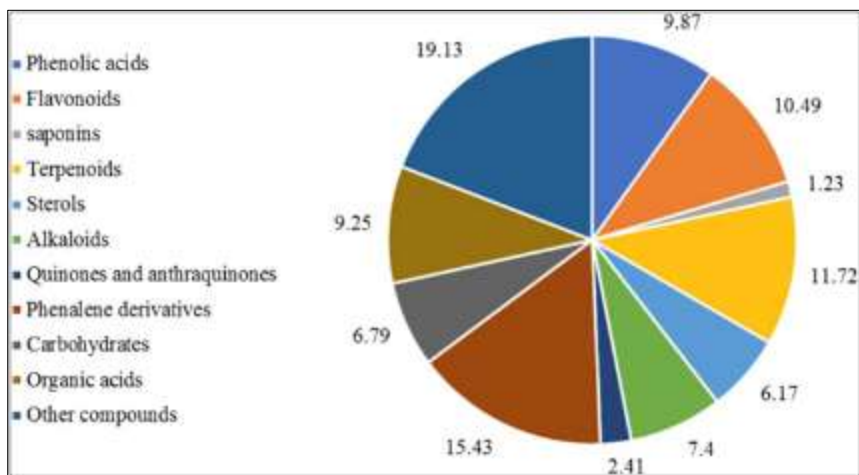


Figure 1 Total number of phytochemicals present in *P. crassipes*

1.1. Geographical Distribution

Water hyacinth is found across the tropical and subtropical regions; it originated from the Amazon Basin, but its entry into Africa, Asia, Australia, and North America was facilitated by human activities [8]. Along with the United States, 56 other countries have reported it as a noxious weed [9]. The geographical distribution also includes Indo-China and Japan with in the U.S. *P. crassipes* occurs throughout the southeast, north to Virginia, and west to Texas, as well as in California and Hawaii [10]. Seasonal escapes from cultivation are reported from New York, Kentucky, Tennessee, and Missouri, but populations apparently do not survive through winter [11]. In Mexico, more than 40,000 hectares of reservoirs, lakes, canals, and drains are infested with water hyacinth [12].

In West Africa, it was first reported in Cameroon between 1997 and 2000, and since then, the country's wetlands have become "home" for the weed [13]. In Asia, water hyacinth is widespread on freshwater wetlands of the Mekong Delta, especially in standing water (MWB/RSCP, 2006). It has been detected in the Sunderbans mangrove forest of Bangladesh [14]. It has caused a heavy situation in the wetlands of the Kaziranga National Park, India. Deeporbeel, a freshwater lake formed by the Brahmaputra River, is heavily infested with this weed [15]. The lake is considered one of the large and important riverine wetlands in the Brahmaputra valley of lower Assam, India. It has also caused many social, economic, and environmental problems in Southern China [16].



Figure 2 *Pontederia crassipes*

1.2. Origin

In 1823, the German naturalist C. Von Martius discovered the species while carrying out floral surveys in Brazil. He named it *Pontederia crassipes*. Solms included it in the *Eichhornia* genus, 60 years later, as had previously been described by Kuntz in 1829. However, a collector by the name of von Humbolt had already collected specimens from Colombia in 1801, together with the species *azurea* [17].

The reason for the world-wide distribution of this weed varies, but generally it has coincided with the plant's ornamental properties or as feed [18].

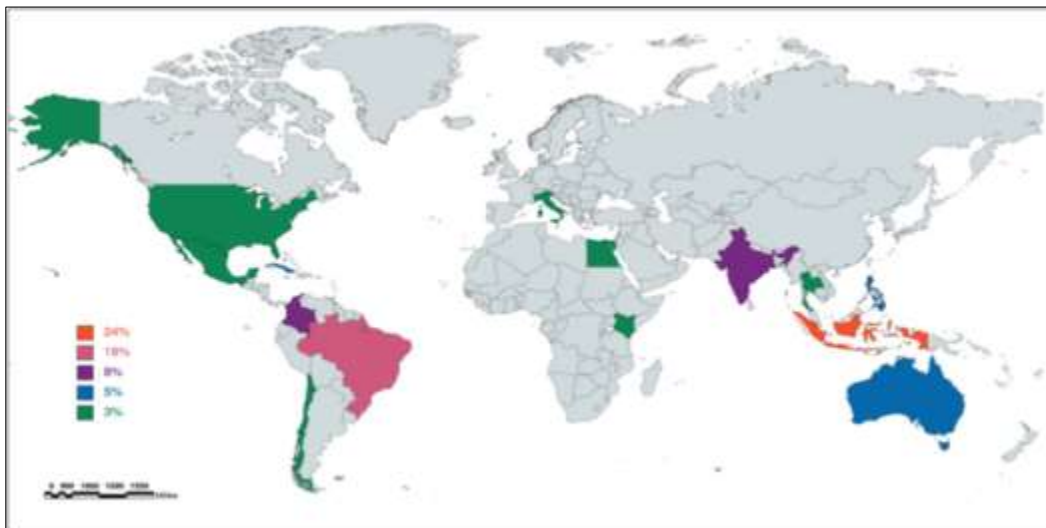


Figure 3 Main countries with studies concerning water hyacinth (*Pontederia crassipes*)

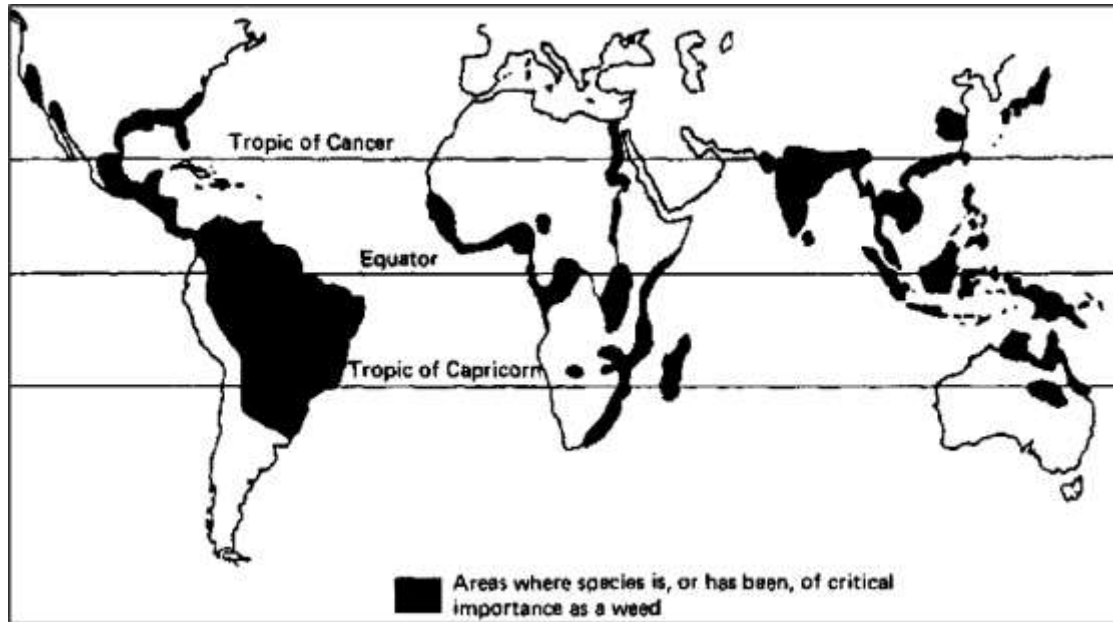


Figure 4 Geographic Distribution of Water Hyacinth weed throughout the world

1.3. Botanical study

1.3.1. The plant family: Pontederiaceae

Pontederiaceae family plants are a small flowering aquatic plant that belongs to the herbaceous monocotyledons. 6-9 genera and about 30-35 species are comprised of this family, which is mostly native to the New World Tropics. Besides their native range, many taxa had been spreading as weeds or ornaments [19].

This family is small where heterostylous aquatic plants are identified, and these are found in tropical and subtropical levels of waters [20].

1.3.2. Identification

The leaves are thick and dark and also flatten on top of the water like a mat. The purple flower is on top of its mat along with its reproductive organ, as the leaf is a floating **macrophyte**. The roots of water hyacinths are thick and dense, which many macro- and micro-invertebrates of small or juvenile fish use. This plant lives in a colony that is created by them [21].

The taxonomy, morphology, anatomy, and ecology of water hyacinth *Pontederia crassipes* are described in Table 1.

Table 1 Scientific Classification of Water Hyacinth [22]

Scientific Classification	
Kingdom	Plantae
Phylum	Spermatophyte
Subphylum	Angiospermae
Class	Monocotyledonae
Order	Pontederiales
Family	Pontederiaceae
Genus	Eichhornia
Species	Eichhornia crassipes

1.4. Morphology and Characteristics of Water Hyacinth

It is a widely established attractive aquatic plant having circular to oval-shaped leaves with malleable covered petioles and carrying aesthetic lilac to blue in color flowers [23]. The fully grown plant comprises long, pendant roots, rhizomes, stolons, leaves, inflorescences, and fruit. The presence of air-filled sacs in the leaves and stems enables them to float on the surface of the water [24].

The prevalence of water hyacinth in aquatic environments that are nutrient-rich can create thick grass mats surrounding a vast area of water. The large, dense, lustrous, and ovoid leaves of the plant can grow above the top of the water body as high as 1 m. The plant possesses lengthy, bulbous, and spongy stalks [25].

The plant can grow to a height of up to 1 m, with an average height of about 40–60 cm. The plant's inflorescence can support 6–10 flowers similar to the lily, with each flower being 4–7 cm in diameter [26]. The plant's ability to float also enables it to grow in harsh conditions such as moist sediments for long periods [27].

1.5. Taxonomy

Water hyacinth, an indigenous monocot aquatic plant of Brazil and Ecuador region of South America, is associated with the group of plants called lilies [28]. It is commonly known as the floating/common water hyacinth. It belongs to the genus, which comprises seven species: *natans*, *E. heterosperma*, *E. crassipes*, *E. azurea*, *E. diversifolia*, *E. paniculata*, and *E. paradoxa*. Among the seven species, only two species are prolific in Africa, namely, *E. natans* and *E. crassipes*, the latter being abundant in Nigeria. *Pontederia crassipes* is distinguishable from other similar aquatic plants by its lustrous leaves, the partial lump of its petioles, and its distinct purple flower [29].

1.6. Medicinal uses

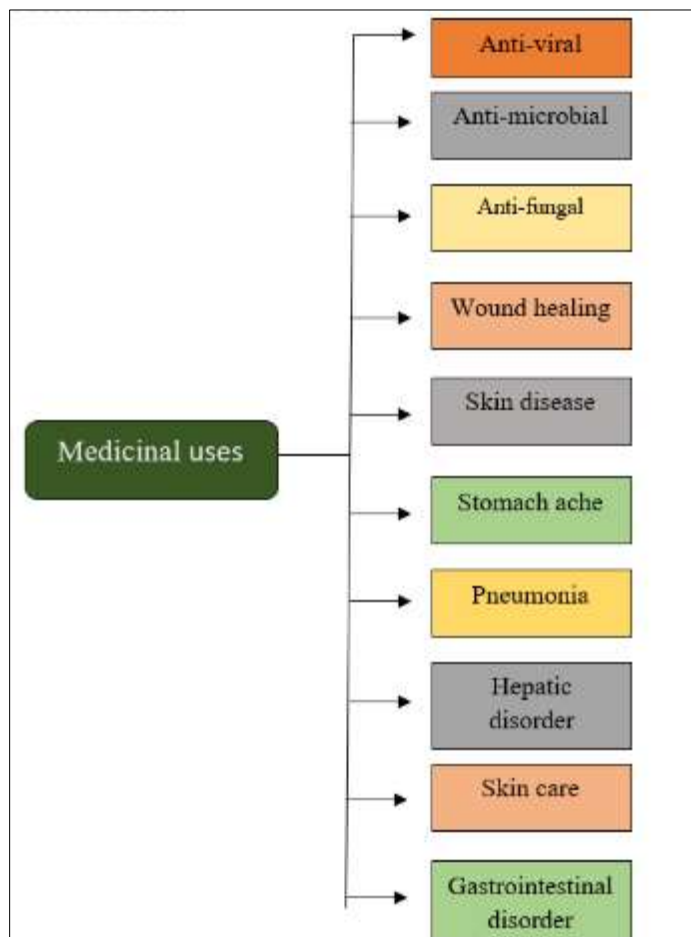


Figure 5 Medicinal Uses of Pontederia Crassipes

- **Antiviral:** Used to treat influenza viruses
- **Antimicrobial:** Extracts of the plant have been shown to be effective against bacteria and fungi
- **Antifungal:** The plant contains bioactive compounds that have antifungal properties
- **Wound healing:** In Chhattisgarh, India, fresh juice from the plant is used to treat wounds
- **Skin disease:** In Tamil Nadu, India, flower oil is applied to treat skin diseases
- **Stomach ache:** In Assam, India, flowers and roots are used to treat stomach aches
- **Pneumonia:** In Assam, India, roots are used to treat pneumonia Hepatic disorders: In Bangladesh, roots and flowers are used to treat hepatic disorders
- **Skin care:** In Nigeria, the plant is used for skin care
- **Gastrointestinal disorders:** Traditionally used to treat diarrhea, intestinal worms, digestive disorders, and flatulence

2. Methods and materials

2.1. Plant materials preparation

Pontederia crassipes plants were collected from Lake and plants were authenticated taxonomically by the national herbarium file. Then plants were washed under running water Leaves were detached from the stems and tubers. Then leaves were solar dried naturally for 20 days in the shade. The dried leaves were ground into a powder using a coffee grinder. Then powder was stored in a tightly closed jar awaiting both hydro and ethanoic solvent extraction.

2.2. *Pontederia crassipes* leaf hydro-ethanolic extract preparation

In a 2000 ml round bottom flask 300 g of *Pontederia crassipes* dried leaf powder added to 900 ml of a 30:70 hydro-ethanolic solution keep for cold extract for 3 days with daily 5 minute shakings twice a day. Then obtained extract was filtered using filter paper into a suitable conical flask through a Buchner funnel Evaporated under low pressure followed by lyophilization under 120 Pa pressure and a temperature of -20°C . The lyophilized extract crystals were kept in a refrigerator at 4°C .

2.3. Biosynthesis of Silver Nanoparticles:

100 ml of a 1 mM solution of silver nitrate was prepared using distilled water, 20 ml of *Pontederia crassipes* extract liquor was added drop wise into the 100 ml of 1 mM aqueous solution of AgNO_3 Heated to between $60\text{--}80^{\circ}\text{C}$ for 1 hour in an Erlenmeyer flask. The reaction was monitored at pH 1, 3, 7, and 10, and optimization was achieved by adjustments with 0.1 N HCl and 0.1 N NaOH accordingly to nanoparticles were observed from 0 to 2 hours at 15 minute intervals and overnight for optimal synthesis of AgNPs Crystals was formed. Next day, synthesis of silver nanoparticles was monitored at temperatures of 25, 50, and 100°C AgNO_3 was measured at different concentrations from 0.50 to 3.0 mM AgNPs solution was centrifuged for 30 minutes at 8,000 rpm Precipitate was washed with ethanol or Recrystallization

3. Pharmacological activity

3.1. Anti-microbial activity

Microorganisms considered separate from human beings but profound beneficial influence as part of our lives. Employed in the manufacturing of dairy products, certain foods, the minimum processing of certain medicines and therapeutic agents, the manufacturing of certain chemicals, and in numerous other ways. Despite the established useful function and potentially valuable activities of microorganisms, these microscopic organisms of life may be best known as agents of food spoilage, herpes, Legionnaires disease, influenza, jaundice, typhoid, dermatomycoses, dysentery, and malaria. Many extracts of *Pontederia crassipes* demonstrated anti-bacterial activities.

In east Africa, it is used for the preparation of crude medicine for treating numerous kinds of virulent diseases related to bacterial infections. In fact, the presence of saponins in the leaves makes that a good candidate, with notable biopotency as a microbial agent. Revealed the potency of *P. crassipes* leaf extract in combating staphylococcal infections, against methicillin-resistant *S. aureus* found in cattle and coagulase-negative staphylococci in rabbits. The inhibition of the growth and division of bacteria could be due to the saponin glycosides and their aglycones. *P. crassipes* displayed antibacterial activities against *S. faecalis*, *E. coli*, and *S. aureus*.

Meanwhile, developments of *Aniger*, *A flavus*, and *C albicans* were repressed by the plant through crude extract or its fractions. The water extract of the leaves demonstrated anti-microbial activity as well against *Bordetella bronchiseptica*,

Proteus vulgaris, and *Salmonella typhi*. In addition to the anti-bacterial activities of silver nanoparticles, synthesized biologically from the extract of *E. crassipes*, were checked against selected gram positive and gram negative bacteria, investigate the anti-microbial activity of hydro alcoholic and ethanolic extracts on *E. coli*, *S. epidermidis*, *P. aeruginosa*, and *B. subtilis*. The antifungal effect of the shoots and leaves of the ethanol extracts were evaluated against two fungi *A. fumigatus*, *M. ruber*, employing the disk diffusion method.

Pontederia crassipes inhibit synthesis of nucleic acids of both gram positive and negative bacteria play a role in intercalation of hydrogen bonding with stacking of nucleic acid bases which cause inhibitory action on **DNA** and **RNA** synthesis. Silver ions bind with the **30S** ribosome subunit, deactivate the ribosome complex, and stop protein synthesis. The synthesis of immature precursor proteins involved in the cell membrane formation through the effect of silver nanoparticles on ribosomes, transcription, and translation, which subsequently means cell death.

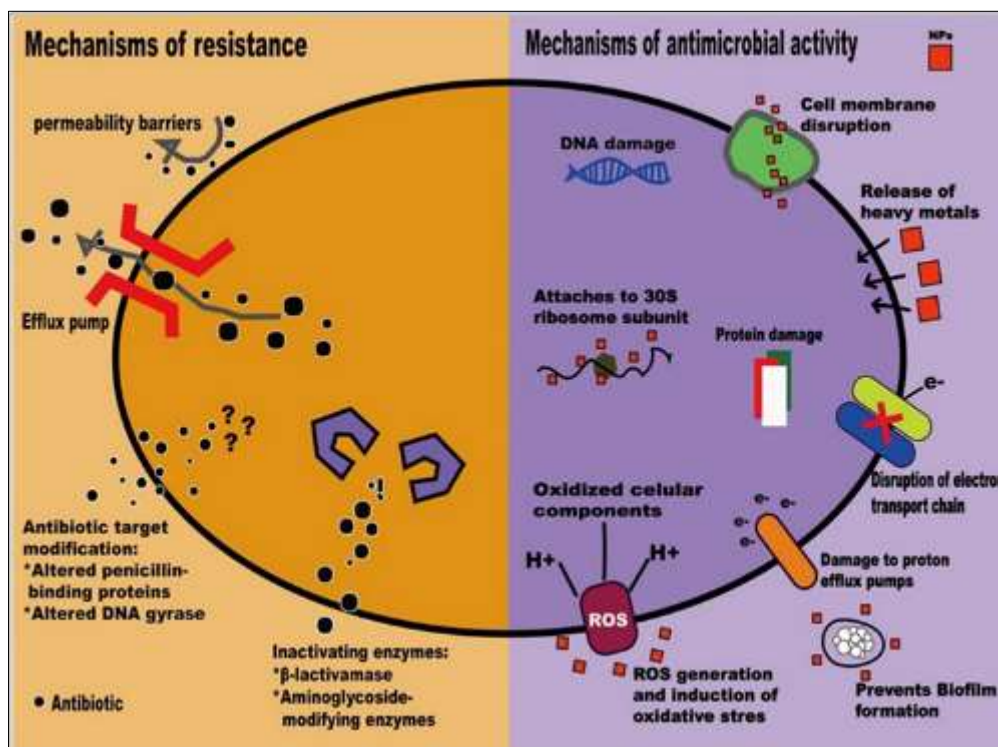


Figure 6 Mechanism action of antimicrobial activity

3.2. Anti-oxidant activity:

Antioxidants are compounds that protect cells against the damaging effects of reactive oxygen species (ROS), such as singlet oxygen, superoxide, peroxy radicals, and peroxy nitrite.

The anti-oxidative activity of *P. crassipes* is connected with the structure of the molecule. The presence of conjugated double bonds and the occurrence of functional groups in the rings. An imbalance between antioxidants and ROS results in oxidative stress, leading to cellular damage.

Oxidative stress has been linked to cancer, aging, atherosclerosis, ischemic injury, inflammation, and neurodegenerative diseases (Parkinson's and Alzheimer's).

E. Crassipes may help provide protection against these diseases by contributing, along with antioxidant vitamins and enzymes, to the total antioxidant defense system of the human body. *E. Crassipes* reduces the production of and quenches reactive oxygen species through suppression of singlet oxygen, inhibition of enzymes that generate ROS, chelating ions of transition metals, which may catalyze ROS production, and quenching cascades of free-radical reactions in lipid peroxidation.

The antioxidant activity of *E. Crassipes* is connected with the structure of the molecule, the presence of conjugated double bonds, and the occurrence of functional groups in the rings. An imbalance between antioxidants and ROS results in oxidative stress, leading to cellular damage.

E. crassipes induces substantial antioxidant activities, and it is confirmed to be a great source of natural antioxidants. The plant is a source of many compounds with radical scavenging activity, such as phenolic acids, sterols, terpenoids, and metabolites with high antioxidant activity.

Ethanol extracts from the leaves exerted robust Fe⁺ chelating activity. Meanwhile, the ethanolic extract of the flowers with a high content of phenolic compounds exhibited a substantial reducing power and radical scavenging activity. In addition, the antioxidant properties of the methanolic crude extract of the whole plant and its isolated compounds, the alkaloids and terpenoids derivatives, were studied using the 2,2-diphenyl-1-picrylhydrazyl radical (DPPH) scavenging activity. As a result, the crude extract showed good anti-oxidant activity.

The high antioxidant potential of the methanolic crude extract could be explained by virtue of the synergistic activities of all bioactive compounds. *E. Crassipes* extracts have shown encouraging anti-aging effects, as determined through DNA damage inhibition and DPPH radical scavenging assays. There was a pronounced increase in the DNA damage inhibition and DPPH radical scavenging ability with an increase in the concentration of ethyl acetate extracts of the plant.

Recently, the antioxidant properties of the leaf protein hydrolysates indicated excellent antioxidant activities, especially the two peptides that have shown high radical scavenging activities.

However, quercetin 7-methyl ether (30) isolated from the whole plant exhibited weak antioxidant activities using the DPPH method

4. Conclusion

Based on the results of research that has been done, it can be concluded that the water hyacinth leaf extract can inhibit bacterial growth in plaques of patients with gingivitis with a minimum concentration of extract.

Some medicinal uses of water hyacinth have been reported. Further research may be needed in this regard to explore the possibility of drug formulation from this weed. Considering its use as feed for animals, toxicity studies have to be further investigated to ensure that the plant is safe for use, whether as food for animals or as treatment of ailments.

The antimicrobial effect of the water hyacinth AgNPs was derived from the chemical and green synthesis way and has been reported in this research work. The green synthesis yielded sound antimicrobial effects comparable to those of pure silver nanoparticles. However, the recycled water hyacinth AgNPs from e-waste did not exhibit good antimicrobial outcomes due to the larger particle size resulting from the lower pH of the AgNO₃ solution. Proper nanosized Ag particles with good antimicrobial results can be obtained by controlling the pH within the range of 7 to 12.

Compliance with ethical standards

Acknowledgment

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Disclosure of conflict of interest

No conflict of interest to be disclosed.

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