

Sources of Potential Fungi Generated Biogenic Nanoparticles for the Control of Diseases Transmitting Mosquitoes: A Review

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Abstract: Vector mosquitoes are diseases transmitting malaria, filarial, dengue, and Japanese encephalitis are an enormous burden to public people worldwide. There is no proper vaccine for those diseases; even though malaria is significant, challenges are still waiting for successful management. Recently, fungi and fungi-derived products control mosquito larvae, pupae, and adults better than compared to plants or other microorganisms. The using fungi such as *Aspergillus* sp., *Beauveria bassiana*, *Metarrhizium anisopliae*, *Verticillium lecanii* are more virulent for controlling mosquito vectors, *Aedes aegypti*, *Anopheles stephensi*, *Culex quinquefasciatus*. The synthesis of silver, gold, zinc, and copper nanoparticles from those fungi has been getting good biological sources for significant reduction of mosquito larval and pupal populations. Fungal-based nanoparticles are highly effective and biorational insecticide for the control of vector populations. In this review, we discussed various sources of fungi that can be synthesized from different nanoparticles to control disease-transmitting mosquito vectors.

Keywords: microorganism; fungi; nanoparticles; mosquito control; biorational insecticide.

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1. Introduction

Mosquitoes carrying pathogens or parasites are endemics in the human population worldwide [1-4]. The threatened mosquito species are an enormous burden to the public and transmit malaria, filarial, yellow fever, Japanese encephalitis, dengue, and zika virus [5-8]. Presently, no right vaccine is available for the treatment of malaria. In this scenario, all other mosquito-borne diseases are challenging issues for successful control and management. Recently, nanoparticles paid more attention to the prevention and control of mosquito-borne diseases.

In this above-said fact, recently, nanoparticles are considered a potential candidate for mosquito control to prevent mosquito-borne diseases [9]. Nanoparticles have reached a goal with the benefit of increasing their efficacy, controlled delivery to the target species, and also long-lasting ability. Using eco-friendly nanoparticles can help control the environmental

problem, perhaps using chemical insecticides as larvicides [9-15]. Currently, silver nanoparticles are widely used for mosquito vector control [16,17, 18].

Recently, fungal toxin or fungal-derived products based nanoparticles are effective in controlling aquatic mosquito larvae and pupae. Therefore, fungal nanoparticle syntheses are an effective bio-control weapon instead of using conventional insecticide[19,20]. Nonetheless, the nanoparticle dispersion must be purified to eliminate fungal residues and impurities, which can be achieved using simple filtration, membrane filtration, gel filtration, dialysis, and ultracentrifugation [21]. The size of the nanoparticles depends on the synthesis conditions such as fungus species, temperature, pH, and dispersion medium, as well as the presence of capping on the nanoparticles [22]. The color of the dispersion is also directly related to the surface plasmon resonance, which varies according to the size and absorbance of the nanoparticles [23]. The synthesis and reduction of biometal could boost the development of a non-toxic, clean, and green approach to the environment for the better production of nanoparticles [24, 25]. Various microbial synthesized nanoparticles are obtained from enzymes, peptides, proteins, electron shuttle quinones, and exopolysaccharides and their mechanism of synthesis by various bioreducing agents [26]. Microbes like bacteria, fungi, and yeast [27-29], are useful production of metal nanoparticles under normal room temperature. The fungi *Fusarium oxysporum* [30,31], *Aspergillus fumigates* [32], and *Verticillium* species [33] are used for the production of metal nanoparticles. According to Li et al., [43], 10 min synthesis of *Phytophthora infestans* fungus nanoparticles. Similarly, the larvicidal potential of fungus *Cochliobolus lunatus* against larvae of *A. aegypti* and *An. stephensi* were reported previously [35]. The fungus synthesized silver and gold nanoparticles are effectively control *A. aegypti* larvae [36,37]. The present review evaluates the larvicidal potential of synthesized varying metal nanoparticles using fungias, a better source for the control of mosquito larvae and pupae in the environment because of their fungal chitinolytic activity and nanoparticle reducing agents [38].

The use of conventional insecticides have disadvantages; toxic chemical generate form the waste leads to an environmental hazard. Recently increasing green approaches for nanoparticle synthesis have more attention than conventional methods [39,40]. The desirable size and morphology form bacteria, yeast, and fungi have been done [41]. Clean and non-toxic nanoparticle production has environmentally acceptable [42]. Fungi have higher quantities of extracellular proteins found, which contribute to nanoparticles become stable [43-45]. The good biomass production of fungi are more advantages [46] than other sources like plants; it is more suitable for mass production and syntheses [47,48].

Recently, physical and chemical methods of nanoparticles synthesis from *Fusarium oxysporum* [49], *Aspergillus fumigates*, *Verticillium* species, *Chrysosporium tropicum* [50,51], *Cochliobolus lunatus* [52], and *Puccinia graminis* [53]. Highly stable AgNPs are significant control of mosquito larval and pupae. The size of nanoparticles up to 100nm and size, distribution, and morphology were considered [54]. The synthesized and control of nanoparticles from microorganisms are potential for eliminating vector and vector-borne diseases [55,56]. Since the past, plants, fungi, bacteria, and viruses have utilized low cost, energy-efficient, and non-toxic to the environment [57].

2. Synthesis of Fungus Mediated Silver Nanoparticles

Fungal species produced bioactive proteins, which are used right from agriculture to the pharmaceutical industry [58-60]. The microbial bioactive compounds have been developed

and are alternative to the conventional insecticide [61]. The fungal metabolites have highly toxic to the aquatic mosquito and are low in other organisms. The nanoparticles are synthesized various from silver, gold, zinc, copper; palladium has paid greater attention because of its physical, chemical, optical, electromagnetic, and mechanical properties [62,63]. Fungi produced known 6400 bioactive protein [64], and its cultivation is large scale production [65,66,67]. Fungi-produced mycelia contain a large amount of enzymes and proteins [68,69]. There were two types of synthesis nanoparticle from fungi, intracellular and extracellular. Intracellular synthesis, metal precursor added to the mycelia and internalized with biomass. The time needed for the required synthesis of nanoparticles and then followed by centrifugation, filtration through disrupting to get a release of nanoparticles [70-72].

Extracellular synthesis, metal precursor added to the fungal filtrate contain only fungal protein and enzyme, the result of the formation of nanoparticles [73-76]. The better syntheses of nanoparticles from the fungi are simple, and membrane filtration, gel filtration, or ultra centrifugations are used to synthesize nanoparticles [77-79] better. The fungal secondary metabolites could be the control of mosquito larvae and pupae. The fungal synthesized nanoparticles are effective and virulent for the alternative for the synthetic insecticide and pollution-free environment [80].

2.1. Synthesis of metal nanoparticles.

Silver nanoparticles vary from other inorganic chemicals because silver is a widely used metal due to its physical and chemical properties [81]. Silver nanoparticles are highly stable, have high thermal, electrical conductivity, chemical stability, catalytic activity. AgNPs properties rank are topper list [82,83,84]. AgNPs require Ag precursor, reducing, stabilizing agents. Silver is a reducing agent for many microorganisms as well as plant sources [85,86]. Similarly, other nanoparticles like zinc size range 50-120nm. The range indicates extracellularly sulfate reduced to sulfide due to sulfate reductase enzyme. Pbs nanoparticles by *Torulopsis* when exposed to aqueous Pb ions [87]. Fungi, *Aspergillus* synthesized lead nanoparticles have been better bioremediation and management [88]. The gold nanoparticles synthesized from fungi are recently reviewed, and their resistance and oxidation good be reliable; yellow and red color referred to large and small gold nanoparticles. The respecting varying morphology could be stabilizing substances like ascorbic acid and citrate [89]. Stabilization can achieve by polyvinyl alcohol [90].

The protein and enzyme on the surface of the fungi could be trapped because of gold ions [91]. The varying sources of gold nanoparticles have different properties. AuCl or AuCl₃ follows both intra and extracellular pathways that require one electron to give gold nanoparticles, whereas, AuCl₃ requires giving three electrons for occur reduction in three steps [92]. Gold nanoparticles from the fungus, *Cyclindrocladium floridanum* were demonstrated by Narayanan and Sakthivel [93]. Mukherjee [94] reported that *Verticillium sp.* Synthesized gold nanoparticles. Soni and Prakash [88] reported for green synthesis of gold nanoparticles from *Aspergillus niger*. The color changes occur its absorption at 530nm (Table 1). They also reported gold nanoparticles are toxic to *An. stephensi*, *Cx. quinquefasciatus* and *A. aegypti* larvae (Table 2).

Nanoparticles from copper are relatively a novel approach and using microorganism for the nanoparticle synthesis are a better achievement of nanobiology field [95]. The nanoparticles are living in the cell organelles, and their shape and size depend on the biological reduction of metal ions [96]. *Fusarium oxysporum* synthesize copper nanoparticles range from <https://nanobioletters.com/>

93-115nm at ambient temperature [97,98]. Copper nanoparticles synthesized from *Aspergillus* species extracellularly [99]. Copper has a spread spectrum of biocidal activities, and have several studies done with copper had remarkable antibacterial activity at the nanoscale level [100]. Copper has suitable biomedical activity, and it seems to be essential for living organisms [101].

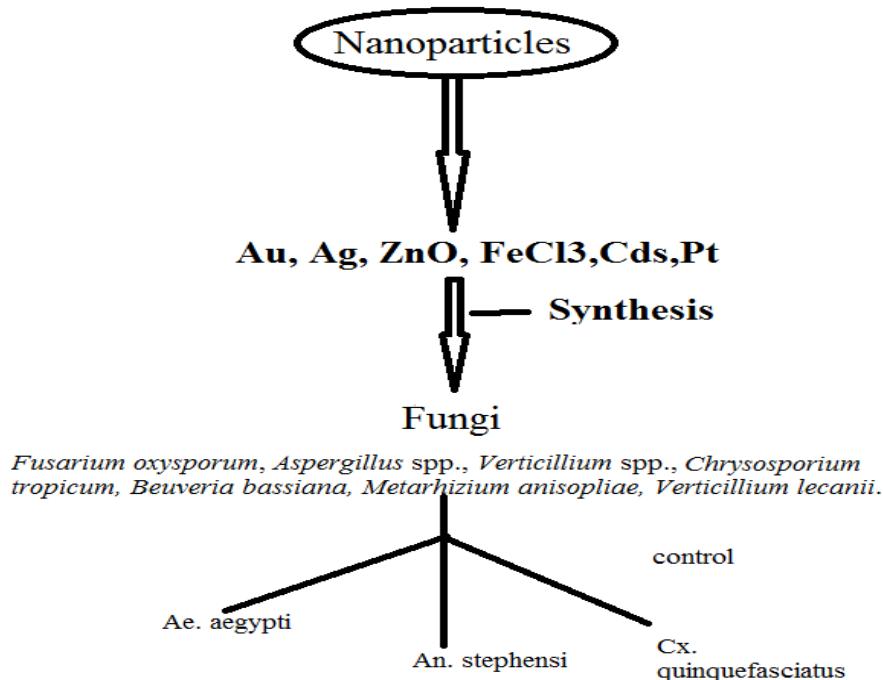


Figure 1. Various metal nanoparticles synthesis from fungi against vectors of mosquitoes.

3. Fungus Nanoparticles for the Control of Mosquito Vectors

3.1. *Aspergillus* spp.

Entomopathogenic fungi were found to be effective against mosquito larvae. Fungi contain enzymes and proteins as reducing agents for the synthesis of nanoparticles; they can be used to synthesize metal nanoparticles. Fungi have a huge amount of protein than comparatively less in bacteria; therefore, converting metal salts to metal nanoparticles is very easy and fast [102]. The extracellular synthesis of silver nanoparticles by *Aspergillus* species, *A. fumigatus*, *A. clavatus*, *A. niger*, and *A. flavus* (Fig. 1) and its nitrate reductase activity was reported by Kamiar Zomorodian [103]. In addition, primary fungal metabolites produce low molecular weight, often biologically active compounds known as secondary metabolites are usually produced by common biosynthetic pathways, often related to morphological development [104].

3.2. *Aspergillus niger*.

A. niger is a keratinophilic, filamentous ascomycete and group of saprophytic molds. Generally, conidia can grow up to 6-47 and Ph 1.4-9.8. This fungus is used to produce citric acids, amylases, lipases, cellulases, xylanases, and proteases to remove heavy metal ions from wastewaters [105]. *A. niger* contains anthraquinone compounds responsible for reducing and capping agents. *A. niger* has better larvicidal and pupicidal activity [106]. *A. niger* nanoparticles are respective larvicidal activity and for environmentally safer side for maintaining ecosystem [107]. The enzyme reductase from *A. niger* reduced silver ions to

AgNPs [108]. *A. niger* also produced extracellular lipase and purified fungal filtrate, which has a lethal effect on *An. stephensi*, *Cx. quinquefasciatus* and *A. aegypti*. Moreover, the presence of mycotoxin "Ochratoxin" in *A. niger* can be a fast metabolite for controlling adult mosquitoes.

3.3. *Aspergillus flavus*.

Fungus, *A. flavus* is a sporophyte haploid filamentous found globally and abundant in the temperate region. It's grown at 37°C with distinct spores and yellow color; a long stalk supporting vesicle is found [109]. *A. flavus* synthesis of nanoparticles either extracellular or intracellular by the mycelia extracts [110]. The extracellular synthesis of *A. flavus* was also reported [111]. The green methods of silver nanoparticles are characterized; AgNPs prepared silver nitrate precursor, reducing agent, and stabilizer. Nanoparticles formed initially by 33kDa protein and followed by a protein (cysteine and free amino acid groups) electrostatic attraction can stabilize the nanoparticle as forming a capping agent [112].

3.4. *Aspergillus fumigatus*.

A. fumigatus rapid biosynthesis have formed silver nanoparticles [113,114] by potential extracellular synthesis. AgNPs using cell-free filtrate are rapid and suitable for large-scale mass production [115]. The most efficient species of *A. fumigatus* is the highest nitrate reductase activity and produced a higher amount of nanoparticles, which is higher monodispersed than in other species [116]. The synthesized and characterized zinc oxide and zinc sulfate nanoparticles using *A. fumigatus* [117]. Shahzad et al.[118] synthesis of nanoparticles using Aspergillus fumigatus BTCB10, obtaining a size of 322.8 nm at 25°C and increasing size as the temperature was increased, reaching 1073.45 nm at 55°C. Nanoparticles are attributed to the aggregation of nanoparticles at a higher temperature.

3.5. *Aspergillus terreus*.

A. terreus is widespread, and it's found all around the world, mostly cultivated in forest soil [119]. It plays an important role in nanoparticles formation and its reducing and capping agent of fungi synthesized AgNPs. The coenzyme NADH is widespread and found that the organism is involved in the redox reaction and used to reduce agents. The reductase enzyme released by *A. terreus* might be a synthesis of nanoparticles. NADH acts as an electron carrier; silver ions obtained electrons through NADH. NADH is a key factor in synthesizing AgNPs from the fungi *A. terreus* [120].

3.6. *Metarhizium anisopliae*.

M. anisopliae is found in soil-borne fungal pathogen of terrestrial insects, offers an environmentally friendly alternative to chemicals for the control of mosquitoes. *M. anisopliae* is widely used to control larval and adult stages of *Aedes*, *Anopheles*, and *Culex* mosquitoes species [121,122,123]. *M. anisopliae* strain has been developed to control a wide variety of terrestrial arthropods, including pests of agro-forests crops and vectors of the human pathogen [124,125]. Larval and pupae of *Cx. pipiens* were treated with *M. anisopliae* mediated AgNPs at different times and different concentrations. The larvae of *Cx. pipiens* show 100% mortality to the prepared AgNPs after/h, whereas the pupae of *Cx. pipiens* were less liable to the novel

AgNPs. Synthesis nanoparticles from *M. anisopliae* can be used as a greener method for a safe environment for vector control [126].

Table 1. Synthesized metal nanoparticles of their varying size and shape fabricated from fungal species.

Fungi	Nanoparticles	Size (nm)	Shape	References
<i>A. clavatus</i>	Au	24.4 ± 11	Triangular, spherical, and hexagonal	[91]
<i>A. flavus</i>	Ag	8.92	spherical	[107]
<i>A. fumigates</i>	ZnO	1.2 - 6.8	Spherical and hexagonal	[136]
<i>A. oryzae</i>	FeCl ₃	10-24.6	Spherical	[137]
<i>C. albicans</i>	Au	5	Monodispersed spherical	[40]
<i>C. glabrata</i>	CdS	20Å, 29Å	Hexamer	[138]
<i>F. oxysporum</i>	Pt	70-180	Rectangular, triangular, spherical, and aggregates	[139]
<i>P. brevicompactum</i>	Au	10 - 60	Spherical, triangular and hexagonal	[140]
<i>P. jadinii</i>	Au	<100	Spherical	[33]
Verticillium sp.	Au	20 ± 8	Spherical	[94]
<i>T. asperellum</i>	Ag	13-18	Nanocrystalline	[141]
<i>T. koningii</i>	Au	30-40	Small spheres to polygons	[142]
<i>P. chrysosporium</i>	Au	10 -100	Spherical	[91]
<i>C. tropicum</i>	Ag	20-50	Spherical	[143]
<i>C. keratinophilum</i>	Ag	24-51	Spherical	[143]
<i>F. oxysporum</i>	Ag	20-40	Spherical	[143]
<i>A. niger</i>	Ag	20-70	Spherical	[143]
<i>V. lecanii</i>	Ag	20-50	Spherical	[143]
<i>M. anisopliae</i>	Ag	190-110	Spherical	[127]
<i>P. verucosum</i>	Ag	3-24	Spherical	[135]

Table 2. Various nanoparticles synthesized from fungus against the vector of mosquitoes.

Fungi	Mosquito sp.	Nanoparticles	Size (nm)	References
<i>C. tropicum</i>	<i>A. aegypti</i>	Ag, Au	2-15 & 20-50	[95,109,51]
<i>C. Lunatus</i>	<i>A. aegypti</i> & <i>An. stephensi</i>	Ag	3-21	[136]
<i>C. tropicum</i>	<i>C. quinquefasciatus</i> & <i>An. stephensi</i>	Au	20 -50 & 2-15	[95,109,51]
<i>A. flavus</i>	<i>C. quinquefasciatus</i>	Ag	2-10	[137]
<i>M. anisopliae</i>	<i>An. culicifacies</i>	Ag	20-24	[138]
<i>B. bassiana</i>	<i>A. aegypti</i>	Ag	20	[139]
<i>C. tropicum</i>	<i>C. quinquefasciatus</i> & <i>An. stephensi</i>	Ag & Au	20 -50 & 2-15	[95,109,51]
<i>A. niger</i>	<i>C. quinquefasciatus</i>	Ag	20 - 70	[140]
<i>P. verucosum</i>	<i>C. quinquefasciatus</i>	Ag	3-24	[135]
<i>C. keratinophilum</i>	<i>C. quinquefasciatus</i> & <i>An. stephensi</i>	Ag	24-51	[143]
<i>F. oxysporum</i>	<i>C. quinquefasciatus</i> & <i>An. stephensi</i>	Ag	20-40	[143]
<i>V. lecanii</i>	<i>C. quinquefasciatus</i> & <i>An. stephensi</i>	Ag	20-50	[143]

3.7. *Beauveria bassiana*.

Entomopathogenic fungi *B. bassiana* belong to the order Hypocreales (Ascomycota), which have a worldwide distribution and are naturally found in soil flora [127]. *B. bassiana* is a cosmopolitan that can inhabit a wide range of environments, including soil, insects, and plants. The fungus can live as a saprophyte in the soil, an endophyte in plants, or an entomopathogen affecting a wide range of arthropods [128]. Mycosynthesis of AgNPs using *B. bassiana* and its mosquitocidal properties against different larval instars were tested *Aedes aegypti*, *Anopheles stephensi*, and *Culex quinquefasciatus*. *B. bassiana* mediated silver nanoparticles are comparatively rapid and reliable cost. Antibacterial therapy in modern medicine *B. bassiana* synthesized silver nanoparticles would be more appropriate for environmentally safer insecticide for controlling mosquitoes (Table 2) [129].

3.8. *Verticillium lecanii*.

V. lecanii is one of the most common and important entomophagous Hyphomycetes fungi that occurred on coccids, aphids, thrips, Diptera, Homoptera, Hymenoptera, Lepidoptera, mites, and in all the climatic regions. Other important substrates for *V. lecanii* are rusts and other fungi. It has also been isolated from oak leaf litter, ash and birch, tea leaves, barley seed, baker's yeast, beet seed, and bursting corn kernels [130,131]. *V. lecanii* can grow on both living and dead materials [132]. It can produce conidia on solid media; in contrast, *V. lecanii* assumes a semi-yeast morphology in liquid media [133]. *V. lecanii* synthesized NPs were formed fairly uniform with spherical shape. Investigated the effect of *V. lecanii* synthesized AgNPs and AuNPs against the larvae and pupae of *A. stephensi*, *C. quinquefasciatus*, and *A. aegypti*. The larvae and pupae were found highly susceptible to the synthesized AgNPs than the AuNPs. The larvae of *C. quinquefasciatus* and *A. aegypti* were found to be more susceptible to the AgNPs, and AuNPs synthesized using *V. lecanii* compared with the larvae of *An. stephensi* (Table 2). This approach suggests that this rapid synthesis of fungus nanoparticles would be useful for developing a biological process for mosquito control [134].

4. Conclusions

Green synthesis of nanoparticles using fungi is an environmentally benign and renewable source for an effective reducing agent that could be utilized for a clean, non-toxic, and environmentally acceptable metal nanoparticle production. Many species of fungi have been utilized in nanotechnology for nanoparticle production, including *Fusarium oxysporum*, *Aspergillus* spp., *Verticillium* spp., and *Chrysosporium tropicum*, *Beuveria bassiana*, *Metarhizium anisopliae*, *Verticillium lecanii*, etc. The fungus nanoparticles formed are highly stable and significant for mosquito larvicide. Nanoparticles, generally considered as particles with a size of up to 100 nm, exhibit completely new or improved properties compared to the larger particles of the bulk material that they are composed of based on specific characteristics such as size, distribution, and morphology. This present review demonstrated the fungus nanoparticles potential mosquito control agent for rapid synthesis, is cost-effective, and could be reliable sources. Synthesis of nanoparticles using microorganisms can potentially eliminate vector-borne disease problems by making the nanoparticles more bio-compatible.

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Conflicts of Interest

The authors declare no conflict of interest.

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