

Exploring the Role of Chitosan: A Natural Solution for Plant Disease and Insect Management

Abstract

Chitosan is an eco-friendly and multipurpose biopolymer that is obtained from the deacetylation of chitin. It is a polysaccharide with several uses. Chitosan is a useful option in phytopathology because of its biocompatibility, biodegradability, and bioactivity. Two naturally occurring substances that may be used in agriculture to manage plant diseases are chitin and chitosan. It was found that these compounds are toxic and that they prevent the growth and development of fungi. According to reports, they were effective against viruses, bacteria, and other pests. When host plants are exposed to microbial infections, fragments of chitin and chitosan have been shown to elicit activities that trigger a range of defence responses, such as the accumulation of phytoalexins, pathogen-related (PR) proteins and proteinase inhibitors, lignin synthesis, and callose formation. Thus, the goal of this study is to give the reader current knowledge about the use of chitosan formulations as pest and diseases control tools as well as information about their potential uses.

Keywords: chitosan formulations, chitosan nanoparticles, plant protection, biological control

1. Introduction

Agrochemical overuse harms the ecosystem, therefore a smart way to mitigate the problem is to release the chemicals under regulated conditions from biodegradable-particles. Alternative, biodegradable, and ecological materials have been sought for by research for a variety of scientific uses in recent decades. Although agriculture is essential for providing food, fibre, and a host of other necessities for human survival it faces a number of significant challenges that need to be resolved to ensure the sustainability and security of food supply worldwide. Due to the highly competitive context and the more stringent ecological parameters, it becomes the prime concern of the scientist, researchers, farmers, etc. to be conscious about quality and ecology. Scientists started looking for safe, natural pesticide substitutes that are both inexpensive and very effective,

in keeping with the idea of going back to nature and utilising science to increase plant efficiency and lower disease risk. In addition to these substitutes, biopolymer-derived particles are becoming a more common and perfect resource for sustainable agriculture. Natural polymers called biopolymers are produced by living organisms' cells which are made up of monomeric units that are covalently bound together to create bigger molecules, much like other polymers. They are numerous, diverse, and their byproducts are essential to life. Their intriguing characteristics make them more and more significant for various uses [1]. Proteomics and poly (amino acids), polysaccharides (cellulose, starch, and xanthan), chitosan, alginate, and carrageenan, and organic polyoxoesters [poly (hydroxyalkanoic acids), poly (malic acid), and cutin] are among the vast array of polymers that can be synthesised by living substances. Biopolymers have gained popularity in agriculture recently. Many studies on the effectiveness of biopolymers in agriculture have been conducted in this respect [2-3].

Chitosan, a linear copolymer composed of D-glucosamine and N-acetyl-D-glucosamine units connected by β -(1-4) glycosidic linkages, is widely employed in control plant disease and insects, improving plant properties, and keeping the biological balance between plants and beneficial microorganisms in the soil. Rouget developed chitosan in 1859 by heating chitin in an alkaline solution [4]. Hoppe-Seyer dubbed this substance "chitosan" a few years later, but its chemical makeup wasn't discovered until 1950 [5] (Fig. 1). Chitosan's antibacterial, antioxidant, and chelating qualities, in addition to being nontoxic and biocompatible, have made it popular. As a byproduct of living things, chitosan (CHT) is produced when chitin is deacetylated from the exoskeleton of crustaceans, including shrimp, shellfish, crabs, cuttlefish, squid pens, and crawfish [6]. In addition, alginate and carrageenan, which formed a naturally occurring anionic polysaccharide that was recovered from seaweeds, are two of the most often used and researched biopolymers [7]. Chitosan may also be made from fungal chitin [8]. Chitosan and its derivatives have a great deal of promise for use in agriculture. These compounds boost crop yields through physiological processes that include quickening the rate of germination and stimulating plant growth [9], boosting photosynthetic pigment, increasing nitrogen-fixing nodes in the leguminous family [10-11], decreasing water loss through transpiration to prevent drought conditions, lowering stress levels, and boosting the uptake of minerals and nutrients [12].

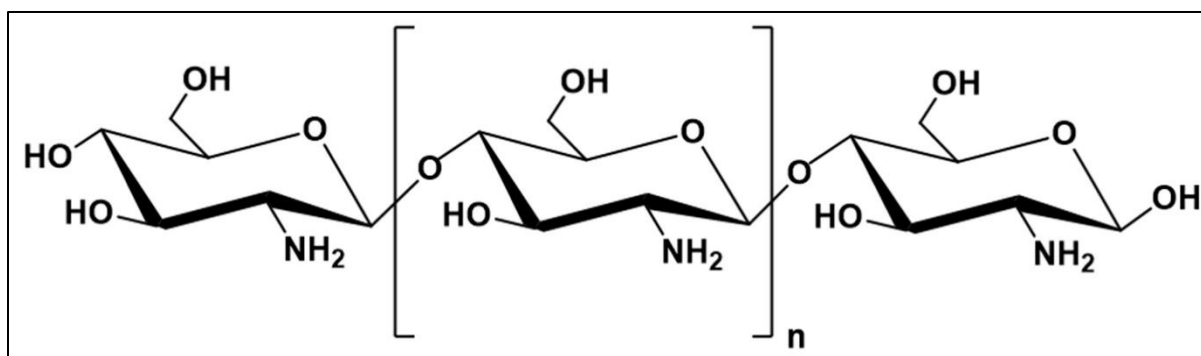


Fig. 1: Chemical structure of chitosan

Apart from growth and development of crop, chitosan has well-known antimicrobial, antioxidant, and antitumoral properties [13], and its application has spread to many sectors, including the pharmaceutical, medical and veterinary sectors. Furthermore, heavy metals including lead, uranium, copper, and mercury may be removed from soil and agricultural effluent by chitosan, allowing it to be utilised again for irrigation [14]. Following several ground breaking discoveries about the protective properties of chitin and chitosan in the early 1980s, a remarkable amount of research on the application of these molecules in agricultural settings has been published. This article provides an overview of chitosan-based derivatives used in plant protection as well as a summary of some of the applications of chitosan in agriculture.

2. Mode of action of chitosan (CHT)

Plants have been shown to respond biologically to chitosan in a variety of ways, depending on the molecule's structure and quantity as well as the species and stage of development of the plant. Chitosan's exact mode of action is yet unknown. Chitosan may directly influence gene expression through chromatin interaction to induce a defence response in plants [15-17]. It has been demonstrated that CHT promotes plant development, plant biocontrol, and increases resistance to both biotic and abiotic stress. The involvement of nitric oxide and hydrogen peroxide in CHT signalling is suggested by the stimulating impact of various enzyme activities on the detoxification of reactive oxygen species. Its antibacterial activity is mediated by a variety of pathways, including chelation of trace metal elements and interactions with histone proteins and plasma membrane phospholipids. Other mechanisms for the antimicrobial effect of chitosan includes agglutination [18], disruption of bacterial cell membrane with leakage of intracellular substances [19], inhibition of toxin production and microbial growth [20], and disruption of cell wall integrity

and alteration of intracellular ultrastructure [21]. The effects of chitosan on plant growth, development, and production have not been well studied; nonetheless, it is primarily thought that chitosan stimulates plants' defences against microbes like bacteria [22]. According to Lizarraga-Paulin et al. [23], chitosan is a biopolymer that possesses antiviral, antibacterial, and antifungal characteristics. Plants that are treated with chitosan have less transpiration [24]. Chitosan is essential to plants' defence mechanisms. Applying CHT foliarly promotes crop development and aids in obtaining a sustainable yield [25]. This compound's ability to establish physical barriers around infection penetration sites and stop them from spreading to healthy tissues is another benefit of its biopolymer characteristics. Chitosan and its bioactive derivatives have the ability to activate H⁺-ATPase, depolarize biological membranes, and cause additional series of events. It has also been reported to induce programmed cell death (PCD).

3. Chitosan (CHT) antimicrobial properties against phytopathogens

Depending on the kind of chitosan (native or modified), its degree of polymerization, the host, the chemical and/or nutritional content of the substrates, and the ambient circumstances, chitosan demonstrates a range of antibacterial properties [26]. It has been observed in certain investigations that oligomeric chitosans (pentamers and heptamers) had superior antifungal activity compared to bigger units [27]. In many instances, the antibacterial activity rose in tandem with the chitosan molecular weight [28], and it appeared to work more quickly on algae and fungus than on bacteria [29].

Chitosan can interfere with electronegative charges on the microbial cell's exterior because of its polycationic nature [30]. In gram-positive bacteria, teichoic acids, lipopolysaccharides, and fungal cell membranes, there is an external electrostatic interaction between the positive amino glucosamine groups (NH₃⁺) of chitosan and the negative charge on the cell surface. This interaction causes changes in cell permeability, leakage of intracellular electrolytes and proteinaceous constituents, and cell death [31]. Repetitive amino groups on the polymer structure's backbone may be the cause of chitosan's antibacterial qualities, according to Divya et al. [32]. Due to its ability to chelate metals and essential nutrients, chitosan has been shown to starve microorganisms and hinder their growth [33]. This is because the amine groups inside the chitosan molecules regulate the intake of metal cations through chelation. Because the amine groups are not protonated and the electron pair on nitrogen in the amine group is accessible for donation to

metal ions, chitosan's metal-binding ability rises at high pH [34]. Because of a polymer coating on the cell surface that prevents the cell from accessing nutrients, the high molecular weight of chitosan may reduce the permeability of the cell membrane [35].

3.1 Chitosan's against viruses

It has been demonstrated that chitosan increases the host's hypersensitive response to infection and prevents viruses and viroids from spreading throughout the plant systemically [27]. Depending on the molecular weight of chitosan, different viral infections were suppressed to varying degrees [28]. Potato virus X, tobacco mosaic and necrosis viruses, alfalfa mosaic virus, peanut stunt virus, and cucumber mosaic virus have all been linked to similar findings [26].

3.2 Chitosan's against bacteria

Numerous types of bacteria are inhibited from growing by chitosan. Different species have minimum growth-inhibiting doses ranging from 10 to 1,000 ppm. It has been demonstrated that quaternary ammonium salts of chitosan, such as N,N,N-trimethylchitosan, N-propyl-N,N-dimethylchitosan, and N-furfuryl-N,N-dimethylchitosan, are efficient at preventing *Escherichia coli* from growing and developing [36], particularly in acidic environments. Likewise, it has been demonstrated that a number of chitin and chitosan derivatives suppress *E. Coli*, *Staphylococcus aureus* [37], several *Bacillus species*.

3.3 Chitosan's against fungi and oomycetes

Chitosan has been shown to have fungicidal effect against a variety of fungus and oomycetes [38]. The lowest amounts that inhibited development ranged from 10 to 5,000 parts per million [39]. Chitosan's highest antifungal activity is frequently seen in the vicinity of its pKa (pH 6.0). Using a radial hyphal growth bioassay of *B. cinerea* and *P. grisea*, Rabea et al. [40] reported on the fungicidal activity of 24 novel derivatives of chitosan (i.e., N-alkyl, N-benzyl chitosans) and shown that all derivatives had a stronger fungicidal effect than the original chitosan. With EC50 values of 0.57, 0.57, and 0.52 gL⁻¹, respectively, N-dodecylchitosan, N-(p-isopropylbenzyl) chitosan, and N-(2,6-dichlorobenzyl) chitosan were the most active against *B. cinerea*. N-(m-nitrobenzyl) chitosan was the most effective against *P. grisea*, with 77% inhibition at 5 g.L⁻¹. The most active compound against *B. cinerea* (EC50 = 1.02 g.L⁻¹) was O-(decanoyl) chitosan at a mol ratio of 1:2 (chitosan to decanoic acid), and the most active compound against *P. grisea* (EC50 =

1.11 g.L⁻¹) was O-(hexanoyl) chitosan. At relatively high concentrations (1.0, 2.0, and 5.0 g.L⁻¹), several of the derivatives also inhibited the production of spores [41]. Chitosan may permeabilize the plasma membrane of *Neurospora crassa* and kill the cells in an energy-dependent way, as recently shown by Palma-Guerrero et al. [42]. Chitosan may permeabilize the plasma membrane of *Neurospora crassa* and kill the cells in an energy-dependent way. With the exception of Zygomycetes, which have chitosan integrated into their cell walls, most fungus and oomycetes may be inhibited in vitro by applying chitosan at a rate of 1 mg/mL [43]. Nemato-/entomopathogenic fungi with extracellular chitosanolytic activity represent another group of fungi that appear to be resistant to chitosan's antifungal effects [42].

4. Chitosan (CHT) in Plant Disease Control

Elicitors that increase host resistance to infections include CHT [44]. Low molecular weight CHT has the ability to stimulate biotic responses in plants by triggering defence mechanisms and activating several pathways that enhance plant resistance. Via a series of biochemical and molecular processes, chitosan is an effective "resistance elicitor" that may activate the plant's innate immune and defence systems. Plant defence genes are activated by chitosan through the octadecanoid pathway. The biopolymer causes a number of reactions, including the manufacture and accumulation of phytoalexins in different host cells [45], callose formation, lignification responses, and the creation of proteinase inhibitors [46]. Depending on the pathosystem, the derivatives used, the concentration, the degree of deacylation, the viscosity, and the applied formulation (i.e., soil amendment, foliar application; chitosan alone or in association with other treatments), chitosan has been extensively explored as a plant pathogen control method with varying degrees of success (Fig. 2).

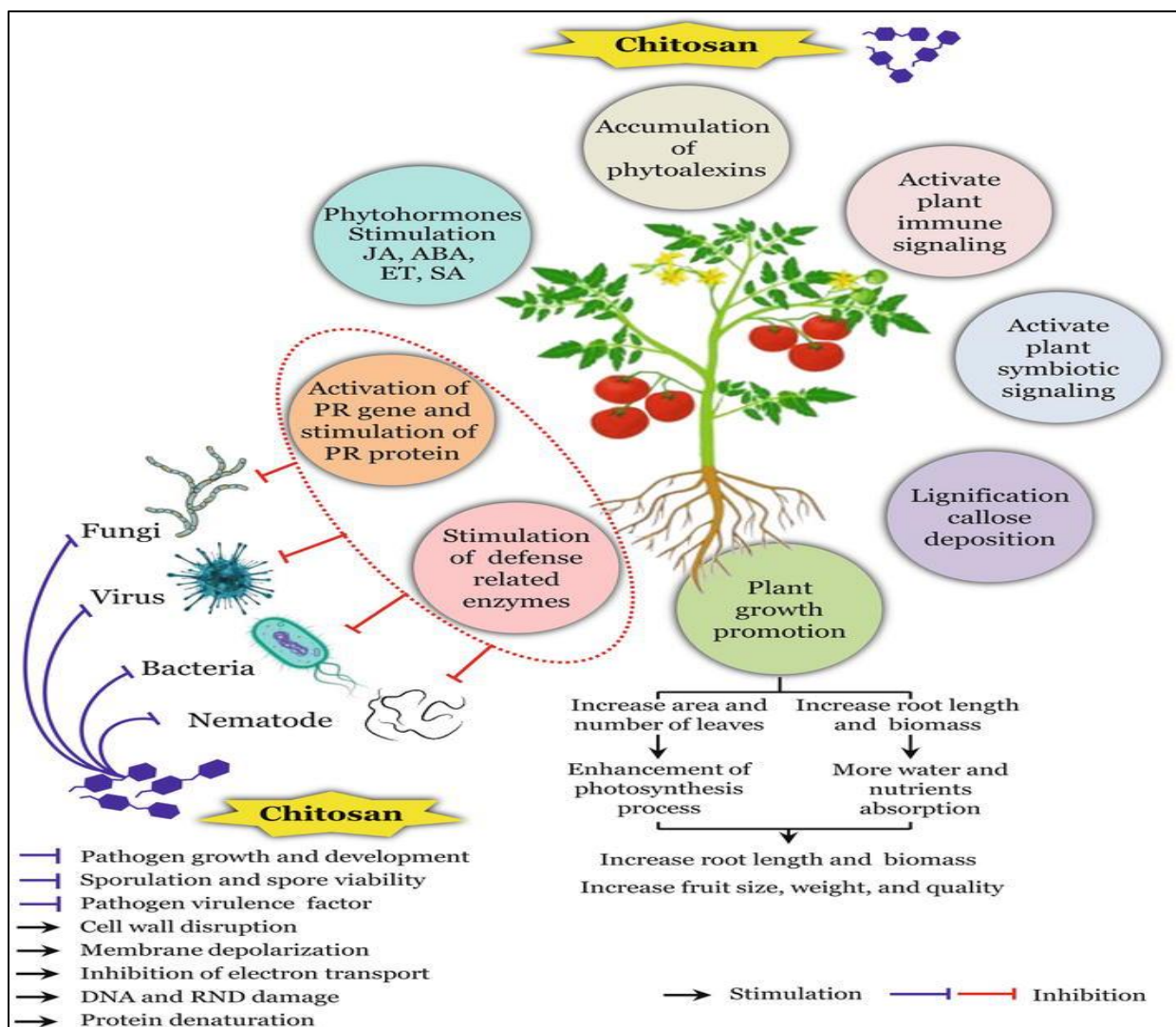


Fig. 2: Chitosan in the enhancement of plant defense system and inhibition of phytopathogens

4.1 Chitosan as seed coating agents in plant disease control

Since chitosan has so many different properties—including a molecular weight that may span a wide range of aspects—it might be useful for seed coating. Because low molecular weight CHT induces resistance, it has a significant deal of potential to protect plants against illnesses [45]. For example, CHT induced resistance in plant defence against *Verticillium dahliae*-induced potato verticillium wilt, reducing vascular invasion and wilt symptoms. Tomato seedlings and seeds have been successfully resistant to *Fusarium oxysporum f.sp. radisis-lycopersici* by the use of CHT [47]. Treating tomato seeds with CHT under in vivo conditions reduced tomato wilt incidence.

According to Photchanachai et al. [48], CHT treatment of seeds stimulated the lignification process in chilli plants, boosting seedling survival and decreasing the anthracnose disease-causing pathogen, *Colletotricum sp.* According to Sharathchandra et al. [49], pearl millet was 48% less susceptible to downy mildew disease in greenhouse settings when treated with Elexa, a formulation containing 4% CHT. Studies on the processes governing illness control have shown that systemic resistance is the cause. Another study looked into how CHT seed priming affected pearl millet's ability to develop disease resistance against *Sclerospora graminicola*-caused downy mildew. By raising the levels of the defence-related enzymes CHTase and peroxidase in comparison to the untreated pearl millet seedlings, the results demonstrated that CHT was efficient in suppressing downy mildew under in vivo and in situ circumstances [50]. By causing phenolics and lignin to accumulate, chitosan treatment of wheat seeds reduced *F. graminearum* infection and boosted resistance in seedlings [51]. In sunflower seedlings treated with CHT, Nandeeshkumar et al. [52] found an increase in the enzymes catalase (CAT), phenylalanine ammonia lyase, peroxidase, polyphenol oxidase, and chitinase (CHI). This resistance was produced against downy mildew caused by *Plasmopara halstedii*. The antifungal activity of CHT against *R. solani* was discovered by analysing the effects of three different forms of CHT against the rice sheathe blight.

4.2 Chitosan applied as foliar treatment in plant disease control

Foliar CHT use has been documented in several systems and for a variety of uses. Applying CHT topically to barley plants, for instance, caused an oxidative burst and the deposition of phenolic compounds in the treated leaves, creating an unsuitable environment that inhibited the growth of fungus [53]. Through the induction of defence mechanisms in tobacco plants, CHT demonstrated antiviral action against enhanced tobacco necrosis necrovirus (TNV) and reduced virus symptoms. Micro-oxidative bursts, microlesions, and callose apposition were associated with this resistance [54]. Iriti et al. [55] looked at the effectiveness of a CHT formulation [Kendal (Kc)] in protecting grapevines against powdery mildew disease in a different research. The results obtained indicated that the induction of plant defence mechanisms and polyphenol production led to a considerable decrease in the severity of the illness. Tomato plants developed resistance against *B. cinerea* after receiving foliar spraying of CHT solution. The buildup of JA and its conjugated JA, as well as the priming of callose at the infection site, were all linked with the CHT-induced resistance. Furthermore, illness resistance was linked to Avr9/cf-9 expression [56]. By using CHT as a foliar

therapy, rice plants were shielded from bacterial late blight (BLB) and their production of the defence enzymes peroxidase and oxidase was enhanced [57]. When rice seedlings were sprayed with CHT, many modes of bacterial suppression by the compound have been documented. In addition to the extraction of phenylalanine ammonia-lyase, peroxidase, and polyphenol oxidase against *Xanthomonas oryzae* pv. *oryzae* and *Xanthomonas oryzae* pv. *oryzicola*, which cause leaf blight and leaf streak, respectively, the biofilm destruction and membrane lysis as a direct activity. Applying CHT to watermelon seedlings against *Acidovorax citrulli*, which causes bacterial fruit blotch, had similar effects [58].

4.3 Chitosan applied as soil amendment in plant disease control

According to research, adding CHT to soil enhances its structure and modifies the rhizosphere's habitat, shifting the microbial balance in favour of helpful bacteria and away from pathogens. *Actinomycetes* and *pseudomonas* populations have been shown to rise in soil supplemented with CHT [59]. When CHT was added to strawberries, the *Bacillus subtilis* population grew [60]. Mishra et al. [61] discovered that CHT had an effective anti-tomato leaf curl virus (ToLCV) effect on tomato plants and increased the quantity of *Pseudomonas* sp. bacterial cells. Vesicular-arbuscular mycorrhizal fungi were more prevalent in soil amended with CHT [62]. Additionally, due to its chelating properties, CHT may shield plants from the damaging effects of pesticides, fertilisers, and hazardous materials. CHT has a beneficial effect on both the plant and the microbial organisms in the soil. According to several investigations, CHT stimulates and evokes the host defence mechanism in response to pathogens that are carried by soil. For example, CHT depolarized the plasma membrane of the root cell and produced hormones, lipid signalling, and substances involved in plant defence, such as phenolics, which in turn induced the release of tomato root exudates. Under in vitro settings, these reactions significantly inhibited the growth of *M. javanica* and *F. oxysporum* f. sp. *radices-lycopersici* [63]. The causative agent of bell pepper root and crown rot, *Phytophthora capsica*, was successfully managed by soil amendment using CHT [64]. Subsequent research has demonstrated that tomato plants developed resistance to *F. oxysporum* f.sp. *radices-lycopersic* when exposed to a soil amendment with a mixture of *B. pumilus* and CHT [65]. According to Algam et al. [66], in a greenhouse setting, CHT used as a soil drench inhibited *R. solanacearum* growth and induced some plant defence responses, such as the synthesis of chitinase and B-2,3-glucanase.

5. Chitosan (CHT) against Insects and Pest control

While several chitosan derivatives have shown pesticide action against a variety of agricultural pests, chitosan itself can improve the availability and stabilisation of certain insecticides or botanicals [13] (Fig. 3). As a safe and ecologically acceptable substitute for synthetic pesticides, chitosan is presently approved by the EU as an active and non-toxic basic chemical for plant protection (Regulation EC No 1107/2009) [67]. A range of chitosan formulations that were enhanced with synthetic or natural insecticides were investigated for their potential to safeguard food as well as be used in agriculture and public health to combat insect pests [68].

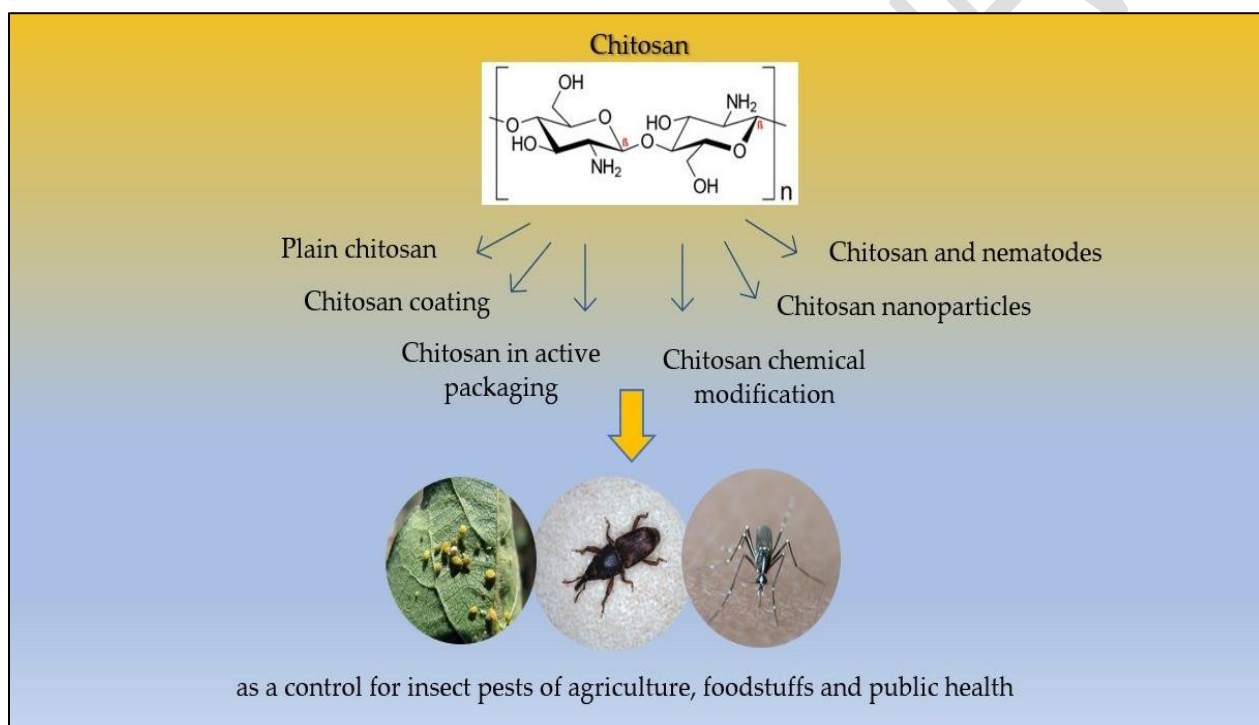


Fig. 3: Chitosan for insect pest control

The polymer has specifically been used in a wide range of formulations, such as a basic formulation of chitosan that is sprayed on leaves, wood, and paper; derivatives of chitosan; chitosan–metal complexes; chitosan nanoparticles that are loaded with insecticides and essential oils (EOs); chitosan coating; and chitosan formulations that contain nematodes. Numerous investigations have been conducted on the use of chitosan in various formulations.

5.1 Plain Chitosan

Chitosan has just come to light in a few publications as a useful technique for managing insect pests in food, agriculture, and public health. However, it appears that the chitosan's antimicrobial action, which disrupts the gut microbiota of the insects, is connected to the insecticidal activity. According to Muryeti and co-authors [69], termites treated with milled paper combined with chitosan had higher death rates (80 and 45%, respectively) than the control group. This appears to be because termites use chitosan paper to obstruct their eating, which disrupts the symbiotic protists' capacity to function in the termites' digestive tract. This sparked an investigation into how chitosan affected the kind and quantity of protists in termites' bellies. Additionally, it was noted that chitosan could be useful in protecting wood from termites [70]. Specifically, after being exposed to 2% of chitosan for 28 days, wood treated with the substance demonstrated a statistically significant mortality of over 94% against subterranean termites for *Reticulitermes flavipes* (*Blattodea, Rhinotermitidae*) [71]. Furthermore, the impact of treating wood with chitosan on the protists of *Reticulitermes virginicus* (*Blattodea, Rhinotermitidae*) was examined. According to the findings, termites in the control group had 10 different species of protists in their hindguts, but only two species were present in the specimens that had chitosan treatment [72].

5.2 Chitosan Coating

In agriculture, chitosan coatings have also been explored for treating fruit and seeds to promote plant development and reduce disease [73]. Specifically, it has been observed in certain investigations that the chitosan covering deters insects and prevents some developmental instars from settling. The growth of eggs and larvae of Mexican fruit flies, *Anastrepha ludens* and *Anastrepha obliqua* (*Diptera Tephritidae*), was in fact suppressed by a chitosan coating on mango fruit, as reported by Salvador-Figueroa et al. [74] and Limon et al. [75]. In each instance, the fruit's weight was decreased and the ripening process was markedly slowed down by the chitosan covering. Regarding the mechanism of action, the suppression of egg and larval development was caused by an increase in the concentration of phenolic chemicals and in the fruit's gas exchange. Furthermore, because the treated fruits had distinct hues and were less permeable, the chitosan coating acted as a barrier, making the fruits less alluring to pests and preventing the oviposition of insects [76].

5.3 Chitosan Coating with Essential Oils (EO)

The efficacy, durability, and availability of EOs have been improved by the coating of chitosan loaded with various EOs, which has shown antifeedant and anti-toxic effects. Ascriczzi et al. [77] found that the chitosan coating film produced by adding *Ferulago campestris* EO to bean seeds demonstrated a statistically significant dose-dependent repellent effect (93.3% repellence at the highest concentration 57.7 $\mu\text{L}/\text{L}$ of air) against the seed pest *Acanthoscelides obstectus* (*Coleoptera Bruchidae*), without negatively affecting the germination and growth of bean plants, while simultaneously being effective in suppressing the germination of weed seeds in vitro. A chitosan covering with eight distinct EOs was created by Hossain et al. [78] in conjunction with ionising radiation, notably gamma radiation at 100 and 300 Gy. The rice treated with eucalyptus and tea tree essential oils coated in a chitosan coating had the highest efficacy against *Sitophilus oryzae* (*Coleoptera Curculionidae*), resulting in 100% mortality at the lowest doses of 0.2 $\mu\text{L}/\text{mL}$ after 24 to 48 hours of incubation. Even after 14 days of treatment, 100% of the insects died from the combination of gamma radiation, giving the food packaging total pest control.

5.4 Chitosan Nanoparticles

Lately, polymeric nanoparticles have become more and more common in many different applications, primarily in the field of agriculture [79]. Because of their regulated delivery methods, chitosan nanoparticles in particular are being researched for their potential as active material carriers as well as for stabilising biological components like proteins, peptides, or genetic material [80]. Chitosan nanoparticles are created via ionotropic gelation, polyelectrolyte complex, microemulsion, emulsion solvent diffusion, and inverse micellar methods. Nonetheless, the first two methods listed above are the most often used ones. Nanospheres, which are solid structures with a homogeneous matrix in which the active ingredient coats them uniformly on the outside, and nanocapsules, which are hollow structures with a polymer membrane and an inner core containing the active ingredient, are the two main groups of nanoparticles based on their morphology [81]. The size, shape, surface area, and area/volume ratio of nanoparticles, which have diameters ranging from 1 to 100 nm, are essential qualities for their effective application in pest management [82]. The smaller size of the nanoparticles guarantees improved permeability, bioavailability, and coverage of the active ingredient. It also improves the formulation stability, slow-release capability, mobility, resistance to degradation, and increased insecticidal activity.

Additionally, it increases water solubility for otherwise insoluble active ingredients. As a result, the most often used particle size in chitosan formulations is nanoscale [83].

Adding EOs to chitosan nanoparticles to boost the oils' stability and bioactivity is one of the formulation's most popular applications. In comparison to the plain product, chitosan nanoparticles help to slow down the release of essential oils and extend their effects over time. Chitosan nanoparticles loaded with *Piper nigrum* EO were assessed against *S. oryzae* and *Tribolium castaneum* (*Coleoptera Tenebrionidae*) Rajkumar et al. [84]. Utilising an impregnated paper assay for fumigation toxicity tests, chitosan nanoparticles loaded with *P. nigrum* EO demonstrated statistically significant prominent mortality after 24 hours, with LC50 values of 25.03 and 29.02 $\mu\text{L/L}$ air for *S. oryzae* and *T. castaneum*, respectively, in comparison to the pure EO (LC50 values of 48.97 and 55.77 $\mu\text{L/L}$ air, respectively). Furthermore, in antifeedant bioassays, the nanoparticles demonstrated higher wheat grain protection against *T. castaneum* and *S. oryzae* assault (0% of damages detected for both species) in comparison to the control (74% damages observed for *S. oryzae* and 86% for *T. castaneum*). Fumigation studies using chitosan nanocapsules loaded with *Rosmarinus officinalis* EO revealed an increase in *T. castaneum* mortality over time (20% mortality after 60 days compared to 2.1% mortality for the pure EO at the same time) (Soltani et al., 2021). The rosemary oil nanoencapsulated by chitosan demonstrated 82% and 50.7% mortality after 60 days against *Oryzaephilus surinamensis* (*Coleoptera, Silvanidae*) and *Carpophilus hemipterus* (*Coleoptera, Nitidulidae*), respectively, according to Soltani and co-authors [85-86].

Extracts from plants have inherent insect-repelling qualities [87]. Furthermore, it seems that the insecticidal action of plant extracts is amplified when paired with chitosan nanoparticles. Indeed, 48 hours after exposure, chitosan nanoparticles loaded with *Nerium oleander* leaf extract had a statistically significant larvicidal activity against *M. domestica* when added to an artificial diet. The pupation rate and adult emergence were reduced by 27% and 60%, respectively, and the LC50 was 0.64 ppm [88]. Additionally, chitosan nanoparticles have been used as agrochemical nanocarriers [89]. In this context, *H. amigera* larvae were used as test subjects for the chitosan-based nano-encapsulation of Ponem, a botanical insecticide that contains neem oil, Karanja oil, azadirachtin, and Karanjin [90]. Tripolyphosphate (TPP) or glutaraldehyde (GLA), which are frequently utilised as crosslinking agents in the manufacture of chitosan nanoparticles, were used to obtain the chitosan nanoparticles, hence augmenting their durability. Furthermore, 48 hours

after treatment, the chitosan nanoparticles containing insecticidal metabolites from the fungal biocontrol agent *Nomuraea rileyi* that were sprayed on leaves demonstrated up to 99% larvicidal activity for the fourth larval stage of *Spodoptera litura* (*Lepidoptera Noctuidae*) [91].

Chitosan nanoparticles have been more widely used recently as an ingredient in formulations for double-strand RNA (dsRNA) and small interfering RNA (siRNA) [92]. Due to its cationic properties, chitosan nanoparticles may be applied for RNA interference (RNAi), a non-invasive method that allows particular RNA fragments (dsRNA and siRNA) to stifle gene expression in mosquito larvae by feeding [93]. By interfering with the locomotor and metabolic processes involved for growth and development, RNAi technology, specifically, employing chitosan nanoparticles, has enhanced its efficiency as a pest control tool, especially for *Anopheles gambiae* (*Diptera Culicidae*) and *Ae. aegypti*.

Future prospect

Chitosan has been the subject of much research, but its exact method of action in controlling plant immunity and inhibiting the pathogen has not been fully understood. It is believed that there may be more overlapping features in the method of action of chitosan, which will require more research in the future. Among the modern fields that has proven effective in examining the worldwide fluctuations in protein expression in living things under many environmental circumstances is proteomics. It is important to think about how these derivatives could affect the ecosystem. Contradictory reports on the toxicity of these metal-complexed derivatives on humans and ecosystems make the case for more research to rule out any potential negative effects on living things. Chitosan's low solubility restricts its applicability; however, it can be enhanced chemically by adding hydrophilic groups to the macromolecular chain of chitosan, for example. In fact, compared to ordinary chitosan, the chitosan derivatives NAC, NBC, and OAC containing alkyl, benzyl, and acyl groups are more soluble and, more importantly, have more insecticidal efficacy. Certain modified chitosan types, such oligosaccharides and chitosan-metal complexes, might be deemed appropriate based on their practicality, efficiency, and environmental friendliness.

Conclusion

Pathogens have seriously harmed agricultural goods in recent decades, leading to decreased growth, lower yields, poor quality, and enormous financial losses. Therefore, it is important to take

pest and plant pathogen control into consideration in order to preserve the quality and health of agricultural goods. Overuse of pesticides over the past century has led to the emergence of resistant strains and worsened effects on the environment and human health. Offering substitute techniques would address the actual issues with agriculture output. Therefore, it is crucial to provide ecologically safe insecticides and treatment techniques for plant diseases. Plant diseases and pests can be controlled using natural compounds like CHT, which also lessen the harmful effects of pesticides on people and the environment. Because of its biological qualities—such as its biocompatibility, biodegradability, nontoxicity, antibacterial activity, and ability to boost immunity—chitosan has been given consideration by several businesses. Thus, as this article reviews, several research has demonstrated that CHT may either directly or indirectly suppress plant pathogens. Even though CHT has been studied extensively for its ability to make plants resistant to pests and diseases, much research still needs to be done. However, additional research is needed since it's likely that the CHT mode of action and the activity caused by plant innate immunity are more intricate than what was previously stated.

References

1. Das, A., Ringu, T., Ghosh, S. Pramanik, N. A comprehensive review on recent advances in preparation, physicochemical characterization, and bioengineering applications of biopolymers. *Polym. Bull.* 80, 7247–7312 (2023). <https://doi.org/10.1007/s00289-022-04443-4>.
2. Hassanisaadi, M., R. Saberi Riseh, A. Rabiei, R.S. Varma, J.F. Kennedy, Corrigendum to “Nano/micro-cellulose-based materials as adsorbents for the remediation of chemical pollutants from agricultural resources” [*Int. J. Biol. Macromol.* 246 (2023) published online/125763], *Int. J. Biol. Macromol.* 250 (2023) 126128.
3. Saberi-Riseh, R., M. Vatankhah, M. Hassanisaadi, J.F. Kennedy, Increasing the efficiency of agricultural fertilizers using cellulose nanofibrils: a review, *Carbohydr. Polym.* 321 (2023) 121313.
4. Raafat D., Sahl H.G. Chitosan and its antimicrobial potential—A critical literature survey. *Microb. Biotechnol.* 2009;2:186–201. doi: 10.1111/j.1751-7915.2008.00080.x.
5. Khor E. Chitin: Fulfilling a Biomaterials Promise. Elsevier Ltd.; Amsterdam, The Netherlands: 2001.
6. No, H.K, Park NY, Lee SH, Meyers SP. Antibacterial activity of chitosans and chitosan oligomers with different molecular weights. *International Journal of Food Microbiology.* 2002;74(1-2):65-72. DOI: 10.1016/S0168-1605(01)00717-6.

7. Ghada E. A. A., Hala R. W., Abeer A. A., Mohamed E. H. A novel alginate–CMC gel beads for efficient covalent inulinase immobilization. *Colloid Polym Sci*, 295, 495–506 (2017). <https://doi.org/10.21608/EJCHEM.2019.6967.1580>.
8. Ghormade V, Pathan E, Deshpande M. Can fungi compete with marine sources for chitosan production? *International Journal of Biological Macromolecules*. 2017;104:1415-1421. DOI: 10.1016/j.ijbiomac.2017.01.112.
9. Laishram B, Devi OR and Ngairangbam H. Insight into Microbes for Climate Smart Agriculture. *Vigyan Varta*. 2023; 4(4):53-56.
10. Devi OR, Sarma A, Borah K, Prathibha RS, Tamuly G, Maniratnam K and Laishram B. Importance of zinc and molybdenum for sustainable pulse production in India. *Environment and Ecology*. 2023; 41(3C): 1853–1859. <https://doi.org/10.60151/envec/lcch4556>.
11. Laishram B, Singh TB, Kalpana A, Wangkheirakpam M, Chongtham SK and Singh W. Effect of Salicylic Acid and Potassium Nitrate on Growth and Yield of Lentil (*Lens culinaris* L.) under Rainfed Condition. *International Journal of Current Microbiology and Applied Sciences*. 2020; 9(11):2779–2791. <https://doi.org/10.20546/ijcmas.2020.911.337>.
12. Devi, O.R, Ojha, N, Laishram, B., Dutta, S. and Kalita, P. (2023). Roles of Nano-Fertilizers in Sustainable Agriculture and Biosafety. *Environment and Ecology*, 41(1B): 457—463.
13. Sharif, R.; Mujtaba, M.; Ur Rahman, M.; Shalmani, A.; Ahmad, H.; Anwar, T.; Tianchan, D.; Wang, X. The Multifunctional Role of Chitosan in Horticultural Crops; A Review. *Molecules* 2018, 23, 872.
14. Hudson SM, Jenkins DW. Chitin and Chitosan, *Encyclopedia of Polymer Science and Technology*. NJ: Wiley Interscience; 2001. DOI: 10.1002/0471440264.pst052.
15. Hadwiger L.A. Anatomy of a nonhost disease resistance response of pea to *Fusarium solani*: PR gene elicitation via DNase, chitosan and chromatin alterations. *Front. Plant Sci*. 2015;12 doi: 10.3389/fpls.2015.00373.
16. Malerba, M.; Cerana, R. Chitosan Effects on Plant Systems. *Int. J. Mol. Sci*. 2016, 17, 996. <https://doi.org/10.3390/ijms17070996>.
17. Laishram B, Singh TB, Devi OR, Khumukcham PS and Ngairangbam H. Yield, Economics, Nutrient uptake and Quality of Lentil (*Lens culinaris* L.) as Influence by Salicylic Acid and Potassium Nitrate under Rainfed Condition. *Environment and Ecology*. 2023; 41(3A):1591–1596. <https://doi.org/10.60151/envec/hdsa3286>.
18. Sudarshan N.R., Hoover D.G., Knorr D. Antibacterial action of chitosan. *Food Biotechnol*. 1992;6:257–272. doi: 10.1080/08905439209549838.
19. Liang C., Yuan F., Liu F., Wang Y., Gao Y. Structure and antimicrobial mechanism of ϵ -polylysine-chitosan conjugates through Maillard reaction. *Int. J. Biol. Macromol.*, 2014;70:427–434. doi: 10.1016/j.ijbiomac.2014.07.012.
20. Reddy M.V.B., Arul J., Ait-Barka E., Angers P., Richard C., Castaigne F. Effect of chitosan on growth and toxin production by *Alternaria alternata* f. sp. *lycopersici*. *Biocontrol. Sci. Technol*. 1998;8:33–43. doi: 10.1080/09583159830414.

21. Geisberger G., Gyenge E.B., Hinger D., Käch A., Maake C., Patzke G.R. Chitosan-thioglycolic acid as a versatile antimicrobial agent. *Biomacromolecules*. 2013;14:1010–1017. doi: 10.1021/bm3018593.
22. Islam MM, Md. Humayun K, Mamun ANK, Islam M, Md. Islam M, Das P. Studies on yield and yield attributes in tomato and chilli using foliar application of oligochitosan. *GSC Biological and Pharmaceutical Sci.* 2018;3(3):020-028. DOI: 10.30574/gscbps.2018.3.3.0038.
23. Lizarraga-Paulin EG, Miranda-Castro SP, MorenoMartinez E, Lara-Sagahon AV, Torres-Pacheco I. Maize seed coatings and seedling sprayings with chitosan and hydrogen peroxide: their influence on some phenological and biochemical behaviors. *J of Zhejiang Uni. Sci. B.* 2013;14(2):87-96. DOI: 10.1631/jzus.b1200270.
24. Dzung NA, Khanh VTP, Dzung TT. Research on impact of chitosan oligomers on biophysical characteristics, growth, development and drought resistance of coffee. *Carbohydrate Polymers*. 2011;84(2):751-755.
25. Mondal MMA, Malek MA, Puteh AB, Ismail MR, Ashrafuzzaman M, Naher L. Effect of foliar application of chitosan on growth and yield in okra, *Australian J Crop Sci.* 2012;6(5):918-921.
26. Pospieszny, H.; Chirkov, S.; Atabekov, J. Induction of antiviral resistance in plants by chitosan. *Plant Sci.* 1991, 79, 63–68.
27. Rabea, E.I.; El Badawy, M.T.; Stevens, C.V.; Smagghe, G.; Steurbaut, W. Chitosan as antimicrobial agent: Applications and mode of action. *Biomacromolecules* 2003, 4, 1457–1465.
28. Kulikov, S.N.; Chirkov, S.N.; Il'ina, A.V.; Lopatin, S.A.; Varlamov, V.P. Effect of the molecular weight of chitosan on its antiviral activity in plants. *Prik. Biokhim. Mikrobiol.* 2006, 42 (2), 224–228.
29. Savard, T.; Beaulieu, C.; Boucher, I.; Champagne, C.P. Antimicrobial action of hydrolysed chitosan against spoilage yeasts and lactic acid bacteria of fermented vegetables, *J. Food Prot.* 2002, 65, 828–833.
30. Abdellatef, M. A., Elagamey, E., & Kamel, S. M. (2023b). Chitosan Is the Ideal Resource for Plant Disease Management under Sustainable Agriculture. In *Biochemistry*. <https://doi.org/10.5772/intechopen.107958>.
31. Palma-Guerrero J, Lopez-Jimenez JA, Pérez-Berná AJ, Huang IC, Jansson HB, Salinas J, et al. Membrane fluidity determines sensitivity of filamentous fungi to chitosan. *Molecular Microbiology*. 2010;75(4):1021-1032. DOI: 10.1111/j.1365-2958.2009.07039.x.
32. Divya K, Vijayan S, George TK, Jisha MS. Antimicrobial properties of chitosan nanoparticles: Mode of action and factors affecting activity. *Fibres and Polymers*. 2017;18(2):221-230. DOI: 10.1007/s12221-017-6690-1.
33. Wang X, Du Y, Fan L, Liu H, Hu Y. Chitosan-metal complexes as antimicrobial agent: Synthesis, characterization and structure-activity study. *Polymer Bulletin*. 2005;55(1):105-113. DOI: 10.1007/s00289-005-0414-1.

34. Goy RC, Britto DD, Assis OB. A review of the antimicrobial activity of chitosan. *Polímeros*. 2009;19:241-247. DOI: F10.1590/S0104-14282009000300013.
35. Yan D, Li Y, Liu Y, Li N, Zhang X, Yan C. Antimicrobial properties of Chitosan and Chitosan derivatives in the treatment of enteric infections. *Mol.* 2021;26(23):7136. DOI: 10.3390/molecules26237136.
36. Jia, Z.; Shen, D.; Xu, W. Synthesis and antibacterial activities of quaternary ammonium salt of chitosan. *Carbohydr. Res.* 2001, 333, 1–6.
37. Kim, J.H.; Shin, J.H.; Lee, H.J.; Chung, I.S.; Lee, H.J. Effect of chitosan on indirubin production from suspension culture of *Polygonum tinctorium*. *J. Ferm. Bioeng.* 1997, 83, 206–208.
38. Vasyukova, N.I.; Chalenko, G.I.; Gerasimova, N.G.; Perekhod, E.A.; Ozeretskovskaya, O.L.; Irina, A.V.; Varlamov, V.P.; Albulov, A.I. Chitin and chitosan derivatives as elicitors of potato resistance to late blight. *Appl. Biochem. Microbiol.* 2005, 36, 372–376 (translated from *Prik. Biokhim. Mikrobiol.* 2000, 36, 433–438).
39. Rhoades, J.; Roller, S. Antimicrobial actions of degraded and native chitosan against spoilage organisms in laboratory media and foods. *Appl. Environ. Microbiol.* 2000, 66, 80–86.
40. Rabea, E.I.; El Badawy, M.T.; Rogge, T.M.; Stevens, C.V.; Höfte, M.; Steurbaut, W.; Smagghe, G. Insecticidal and fungicidal activity of new synthesized chitosan derivatives. *Pest Manag. Sci.* 2005, 61, 951–960.
41. Badawy, M.E.I.; Rabea, E.I.; Rogge, T.M.; Stevens, C.V.; Steurbaut, W.; Höfte, M.; Smagghe, G. Fungicidal and insecticidal activity of O-acyl chitosan derivatives. *Polymer Bull.* 2005, 54, 279–289.
42. Palma-Guerrero, J.; Huang, I.C.; Jansson, H.B.; Salinas, J.; Lopez-Llorca, L.V.; Read, N.D. Chitosan permeabilizes the plasma membrane and kills cells of *Neurospora crassa* in an energy dependent manner. *Fungal Gen. Biol.* 2009, 46, 585–594.
43. Palma-Guerrero, J.; Jansson, H.B.; Salinas, J.; Lopez-Llorca, L.V. Effect of chitosan on hyphal growth and spore germination of plant pathogenic and biocontrol fungi. *J. Appl. Microbiol.* 2008, 104, 541–553.
44. Malerba, M., R. Cerana, Chitosan effects on plant systems, *Int. J. Mol. Sci.* 17 (7) (2016) 996.
45. Orzali, L., B. Corsi, C. Forni, L. Riccioni, Chitosan in agriculture: a new challenge for managing plant disease, in: *Biological Activities And Application of Marine Polysaccharides*, 2017, pp. 87–96.
46. Riseh, R. S., Hassanisaadi, M., Vatankhah, M., Babaki, S. A., & Barka, E. A. (2022). Chitosan as a potential natural compound to manage plant diseases. *International Journal of Biological Macromolecules*, 220, 998–1009. <https://doi.org/10.1016/j.ijbiomac.2022.08.109>.

47. Benhamou, N., P. Lafontaine, M. Nicole, Induction of systemic resistance to Fusarium crown and root rot in tomato plants by seed treatment with chitosan, *Phytopathology* 84 (12) (1994) 1432–1444.
48. Photchanachai, S., J. Singkaew, J. Thamthong, Effects of chitosan seed treatment on *Colletotrichum* sp. and seedling growth of chili cv. jinda, in: IV International Conference on Managing Quality in Chains-The Integrated View on Fruits and Vegetables Quality 712, 2006, pp. 585–590.
49. Sharathchandra, R., S.N. Raj, N. Shetty, K. Amruthesh, H.S. Shetty, A chitosan formulation Elexa™ induces downy mildew disease resistance and growth promotion in pearl millet, *Crop Prot.* 23 (10) (2004) 881–888.
50. Manjunatha, G., K. Roopa, G.N. Prashanth, H. Shekar Shetty, Chitosan enhances disease resistance in pearl millet against downy mildew caused by *Sclerospora graminicola* and defence-related enzyme activation, *Pest Manag. Sci.* 64 (12) (2008) 1250–1257.
51. Bhaskara Reddy, M., J. Arul, P. Angers, L. Couture, Chitosan treatment of wheat seeds induces resistance to *Fusarium graminearum* and improves seed quality, *J. Agric. Food Chem.* 47 (3) (1999) 1208–1216.
52. Nandeeshkumar, P., J. Sudisha, K.K. Ramachandra, H. Prakash, S. Niranjana, S. H. Shekar, Chitosan induced resistance to downy mildew in sunflower caused by *Plasmopara halstedii*, *Physiol. Mol. Plant Pathol.* 72 (4–6) (2008) 188–194.
53. Faoro, F., D. Maffi, D. Cantu, M. Iriti, Chemical-induced resistance against powdery mildew in barley: the effects of chitosan and benzothiadiazole, *Biocontrol* 53 (2) (2008) 387–401.
54. Iriti, M., M. Sironi, S. Gomasasca, A. Casazza, C. Soave, F. Faoro, Cell death mediated antiviral effect of chitosan in tobacco, *Plant Physiol. Biochem.* 44 (11–12) (2006) 893–900.
55. Iriti, M., S. Vitalini, G. Di Tommaso, S. D’Amico, M. Borgo, F. Faoro, New chitosan formulation prevents grapevine powdery mildew infection and improves polyphenol content and free radical scavenging activity of grape and wine, *Aust.J. GrapeWine Res.* 17 (2) (2011) 263–269.
56. De-Vega, D., N. Holden, P.E. Hedley, J. Morris, E. Luna, A. Newton, Chitosan primes plant defence mechanisms against *Botrytis cinerea*, including expression of Avr9/Cf-9 rapidly elicited genes, *Plant Cell Environ.* 44 (1) (2021) 290–303.
57. Stanley-Raja, V., S. Senthil-Nathan, K.M.-P. Chanthini, H. Sivanesh, R. Ramasubramanian, S. Karthi, N. Shyam-Sundar, P. Vasantha-Srinivasan, K. Kalaivani, Biological activity of chitosan inducing resistance efficiency of rice (*Oryza sativa* L.) after treatment with fungal based chitosan, *Sci. Rep.* 11 (1) (2021) 1–15.
58. Li, B., Y. Shi, C. Shan, Q. Zhou, M. Ibrahim, Y. Wang, G. Wu, H. Li, G. Xie, G. Sun, Effect of chitosan solution on the inhibition of *Acidovorax citrulli* causing bacterial fruit blotch of watermelon, *J. Sci. Food Agric.* 93 (5) (2013) 1010–1015.

59. Hallmann, J., D. Bell, B. Kopp-Holtwiesche, R. Sikora, in: Effects of natural products on soil organisms and plant health enhancement, Mededelingen (Rijksuniversiteit te Gent. Fakulteit van de Landbouwkundige en Toegepaste Biologische Wetenschappen) 66(2b), 2001, pp. 609–617.
60. Lowe, A., S.M. Rafferty-McArdle, A.C. Cassells, Effects of AMF-and PGPR-root inoculation and a foliar chitosan spray in single and combined treatments on powdery mildew disease in strawberry, *Agric. Food Sci.* 21 (1) (2012) 28–38.
61. Mishra, S., K.S. Jagadeesh, P.U. Krishnaraj, S. Prem, Biocontrol of tomato leaf curl virus (ToLCV) in tomato with chitosan supplemented formulations of *Pseudomonas* sp. under field conditions, *Aust. J. Crop. Sci.* 8 (3) (2014) 347–355.
62. Park, K-C., T.-H. Chang, Effect of chitosan on microbial community in soils planted with cucumber under protected cultivation, *Hort.Sci.Technol.* 30 (3) (2012) 261–269.
63. Suarez-Fernandez, M., F.C. Marhuenda-Egea, F. Lopez-Moya, M.B. Arnao, F. Cabrera-Escribano, M.J. Nueda, B. Guns´e, L.V. Lopez-Llorca, Chitosan induces plant hormones and defenses in tomato root exudates, *Front. Plant Sci.* 1677 (2020).
64. Kim, K.D., S. Nemeč, G. Musson, Control of phytophthora root and crown rot of bell pepper with composts and soil amendments in the greenhouse, *Appl. SoilEcol.* 5 (2) (1997) 169–179.
65. Benhamou, N., J.W. Kloepper, S. Tuzun, Induction of resistance against fusarium wilt of tomato by combination of chitosan with an endophytic bacterial strain: ultrastructure and cytochemistry of the host response, *Planta* 204 (2) (1998) 153–168.
66. Algam,S., G. Xie, B. Li, S. Yu, T. Su, J. Larsen, Effects of *Paenibacillus* strains and chitosan on plant growth promotion and control of *Ralstonia* wilt in tomato, *J. Plant Pathol.* (2010) 593–600.
67. Žabka, M.; Pavela, R. The dominance of chitosan hydrochloride over modern natural agents or basic substances in efficacy against *Phytophthora infestans*, and its safety for the non-target model species *Eisenia fetida*. *Horticulturae* 2021, 7, 366.
68. Rajkumar, V.; Gunasekaran, C.; Paul, C.A.; Dharmaraj, J. Development of encapsulated peppermint essential oil in chitosan nanoparticles: Characterization and biological efficacy against stored-grain pest control. *Pestic. Biochem. Phys.* **2020**, 170, 104679.
69. Muryeti, M.; Pratiwi, F.E.; Yuniastuti, R.T.; Mulyani, E.B. Termiticidal activity of chitosan on paper. *Prog. Chem. Appl. Chitin Deriv.* 2020, 25, 164–173.
70. Liibert, L.; Treu, A.; Meier, P. A two-step wood protection process using alternative wood protection agents in combination with an oil treatment. In Proceedings of the 7th meeting of the Nordic-Baltic Network in Wood Material Science & Engineering (WSE), Oslo, Norway, 27–28 October 2011.
71. Raji, O.; Tang, J.D.; Telmadarrehei, T.; Jeremic, D. Termiticidal activity of chitosan against the subterranean termites *Reticulitermes flavipes* and *Reticulitermes virginicus*. *Pest Manag. Sci.* 2018, 74, 1704–1710.

72. Telmadarrehei, T.; Tang, D.J.; Raji, O.; Rezazadeh, A.; Jeremic, D. Effect of chitosan on diversity and number of protists insubterranean termites. In Proceedings of the 114th Annual Meeting of the American Wood Protection, Seattle, WA, USA, 22–24 April 2018; pp. 22–24.
73. Li, K.; Xing, R.; Liu, S.; Li, P. Chitin and chitosan fragments responsible for plant elicitor and growth stimulator. *J. Agric. Food Chem.* 2020, 68, 12203–12211.
74. Salvador-Figueroa, M.; Hernández-Ortiz, E.; Ventura-González, C.; Ovando-Medina, I.; Adriano-Anaya, L. Effect of chitosan coatings on the development of *Anastrepha ludens* (Loew) in mango fruits (*Mangifera indica* L.) cv. Ataulfo. *Rev. Iberoam. De Tecnol. Postcosecha* 2013, 14, 14–20.
75. Limon, T.; Birke, A.; Monribot-Villanueva, J.L.; Guerrero-Analco, J.A.; Altúzar-Molina, A.; Carrión, G.; Goycoolea, F.M.; Moerschbacher, B.M.; Aluja, M. Chitosan coatings reduce fruit fly (*Anastrepha obliqua*) infestation and development of the fungus *Colletotrichum gloeosporioides* in Manila mangoes. *J. Sci. Food Agric.* 2021, 101, 2756–2766.
76. Aluja, M.; Mangan, R.L. Fruit Fly (Diptera: Tephritidae) Host Status Determination: Critical Conceptual, Methodological, and Regulatory Considerations. *Annu. Rev. Entomol.* 2008, 53, 473–502.
77. Ascrizzi, R.; Flamini, G.; Bedini, S.; Tani, C.; Giannotti, P.; Lombardi, T.; Conti, B.; Fraternali, D. *Ferulago campestris* Essential Oil as Active Ingredient in Chitosan Seed-Coating: Chemical Analyses, Allelopathic Effects, and Protective Activity against the Common Bean Pest *Acanthoscelides obtectus*. *Agronomy* 2021, 11, 1578.
78. Hossain, F.; Follett, P.; Salmieri, S.; Vu, K.D.; Harich, M.; Lacroix, M. Synergistic effects of nanocomposite films containing essential oil nanoemulsions in combination with ionizing radiation for control of rice weevil *Sitophilus oryzae* in stored grains. *J. Food Sci.* 2019, 84, 1439–1446.
79. An, C.; Sun, C.; Li, N.; Huang, B.; Jiang, J.; Shen, Y.; Wang, C.; Zhao, X.; Cui, B.; Wang, C.; et al. Nanomaterials and nanotechnology for the delivery of agrochemicals: Strategies towards sustainable agriculture. *J. Nanobiotechnology* 2022, 20, 11.
80. Ghormade, V.; Deshpande, M.V.; Paknikar, K.M. Perspectives for nano-biotechnology enabled protection and nutrition of plants. *Biotechnol. Adv.* 2011, 29, 792–803.
81. Shoueir, K.R.; El-Desouky, N.; Rashad, M.M.; Ahmed, M.K.; Janowska, I.; El-Kemary, M. Chitosan based-nanoparticles and nanocapsules: Overview, physicochemical features, applications of a nanofibrous scaffold, and bioprinting. *Int. J. Biol. Macromol.* 2021, 167, 1176–1197.
82. Devi OR, Laishram B, Singh S, Paul AK, Sarma HP, Bora SS and Devi SS. A Review on Mitigation of Greenhouse Gases by Agronomic Practices towards Sustainable Agriculture. *International Journal of Environment and Climate Change.* 2023;13(8):278–287. <https://doi.org/10.9734/ijecc/2023/v13i81952>.

83. Athanassiou, C.G.; Kavallieratos, N.G.; Benelli, G.; Losic, D.; Usha Rani, P.; Desneux, N. Nanoparticles for pest control: Current status and future perspectives. *J. Pest Sci.* 2018, 91, 1–15.
84. Rajkumar, V.; Gunasekaran, C.; Dharmaraj, J.; Chinnaraj, P.; Paul, C.A.; Kanithachristy, I. Structural characterization of chitosan nanoparticle loaded with *Piper nigrum* essential oil for biological efficacy against the stored grain pest control. *Pestic. Biochem. Phys.* 2020, 166, 104566.
85. Soltani, A.; Labidi, A.; Ben Jemaa, J. Development of formulation based on essential oils of rosemary to manage pests of stored cereal foodstuffs. In *Proceedings of the 1st International Electronic Conference on Entomology*, 1–15 July 2021; p. 10407.
86. Soltani, A.; Haouel-Hamdi, S.; Sadraoui Ajmi, I.; Djebbi, T.; Ben Abada, M.; Yangui, I.; Chouachi, N.; Hassine, K.; Majdoub, H.; Messaoud, C.; et al. Insights for the control of dried-fruit beetle *Carpophilus hemipterus* (Nitidulidae) using rosemary essential oil loaded in chitosan nanoparticles. *Int. J. Environ. Health Res.* 2022, 33, 1243–1253.
87. Lahlali, R.; El Hamss, H.; Mediouni-Ben Jemâa, J.; Barka, E.A. The Use of Plant Extracts and Essential Oils as Biopesticides. *Front. Agron.* 2022, 4, 921965.
88. El-Monairy, O.M.; Abdel-Meguid, A.D.; Emara, M.M. Efficacy of Methanol Leaf Extract, Biosynthesized Silver and Chitosan Nanoparticles Using *Nerium oleander* against *Musca domestica*. *Egypt. Acad. J. Biol. Sci. F. Toxicol. Pest Control* 2020, 12, 35–45.
89. Maluin, F.N.; Hussein, M.Z. Chitosan-based agronanochemicals as a sustainable alternative in crop protection. *Molecules* 2020, 25, 1611.
90. Paulraj, G.M.; Ignacimuthu, S.; Gandhi, M.R.; Shajahan, A.; Ganesan, P.; Packiam, S.M.; Al-Dhabi, N.A. Comparative studies of tripolyphosphate and glutaraldehyde cross-linked chitosan-botanical pesticide nanoparticles and their agricultural applications. *Int. J. Biol. Macromol.* 2017, 104, 1813–1819.
91. Upadhyay, N.; Singh, V.K.; Dwivedy, A.K.; Das, S.; Chaudhari, A.K.; Dubey, N.K. Assessment of *Melissa officinalis* L. essential oil as an eco-friendly approach against biodeterioration of wheat flour caused by *Tribolium castaneum* Herbst. *Environ. Sci. Pollut. Res.* 2019, 26, 14036–14049.
92. Kashyap, P.L.; Xiang, X.; Heiden, P. Chitosan nanoparticle based delivery systems for sustainable agriculture. *Int. J. Biol. Macromol.* 2015, 77, 36–51.
93. Kumar, D.; Saravana Kumar, P.; Gandhi, M.R.; Al-Dhabi, N.A.; Paulraj, M.G.; Ignacimuthu, S. Delivery of chitosan/dsRNA nanoparticles for silencing of wing development vestigial (vg) gene in *Aedes aegypti* mosquitoes. *Int. J. Biol. Macromol.* 2016, 86, 89–95.