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Microbial Chitinases: Role in Fungal Plant Disease Resistance

(*Ranjna Sharma[1], Sakshi[2], Shilpa Katoch[2], Shalini Verma[3] and HS Negi[4])
[1]Assistant Professor, Microbiology, Department of Basic Sciences, Dr YSPUHF, Nauni, Solan, Himachal Pradesh, India
[2]Research Scholar, Department of Basic Sciences, Dr YSPUHF, Nauni, Solan, India
[3]Senior Scientist, Plant Pathology, Dr YSPUHF, Nauni, Solan, Himachal Pradesh, India
[4]Assistant Professor, Department of Plant Pathology, College of Agriculture, Banda University of Agriculture and Technology, Banda, UP, India
*Corresponding Author's email: ranjnassharma@gmail.com

Plant diseases caused by infectious pathogens have seriously affected human society and nature through their damage to food production, economic development, ecological resilience, and natural landscapes over human history. Plant diseases can occur in the entire crop production chain and remain one of the greatest threats to the sustainable development of society, resulting in annual losses. In the long history of agriculture, humans have developed a variety of approaches to manage plant diseases. Microbial chitinases are one of the modern approaches toward plant disease resistance. These are enzymes produced by various microorganisms, including bacteria and fungi, which break down chitin, a biopolymer found in the exoskeletons of insects and the cell walls of fungal pathogens. These enzymes play a significant role in plant resistance against pathogens having chitin-containing structures. These enzymes directly attack the chitin, main cell wall constituent thus kill the pathogen.

Microbial Chitinases

Enzymes are biocatalysts that catalyze the rate of chemical reaction. Chitinase enzymes belong to the glycosyl hydrolase superfamily, which catalyzes the hydrolysis of glycosidic bonds in chitin (Nayak *et al.* 2021) Chitinases mainly belong to families 18 and 19 of glycosyl hydrolases based on characteristics. Viz. N-terminal sequence, enzyme localization, isoelectric pH, signal peptides, and inducers. Family 18 contains chitinases of classes III and V, whereas family 19 includes chitinases of classes I, II, and IV. Chitinases are produced naturally by a wide range of organisms i.e., fungi, bacteria, yeasts, plants, and actinomycetes. Most of the chitinases from microbial sources have been grouped into family 18 of glycosyl hydrolases, except some of the Gram-positive bacteria that are included in family 19. Chitinase has a specific affinity toward chitin, an insoluble linear β-1,4-linked polymer of N-acetylglucosamine (GlcNAc) (Kumar et al. 2018) Chitin is a polymer found in many organisms such as cell walls of fungi and algae to cuticle of insects, shells of mollusks and crustaceans. It isa linear un-branched polysaccharide and found as crystalline microfibrils. Chitin chains comprise β-(1 → 4) linked N-acetyl-D-glucosamine (GlcNAc) residues, while glucosamine residue is very low. It is less soluble in water and solvents. Chitin is known to have many valuable properties and has been applied in various fields. Chitin can be used as a source of nitrogen and carbon to isolate chitinase-producing microorganisms (Gomaa 2021)

Mode of Action

Chitinase activity can be divided into two types: endochitinases and exochitinases. Endochitinase degradation results in soluble, low-molecular mass multimers of GIcNAc such as chitotetraose and chitotriose with the dimer, di-acetylchitobiose, being predominant.On the other hand, exochitinases degradation results in the release of diacetylchitobiose. Cleavage occurs only at the nonreducing end of chitin microfibril.The chitinases hydrolyze β-1,4 linkages in chitin, yielding predominately N-N-diacetyl chitobiose, which is further degraded by N-acetyl-glucosaminidases to the GlcNAc monomer (Sahai *et al.* 1993) Furthermore, the waste products of chitin and other value-added products formed by chitinases are also studied (Gomaa 2021)

Factors affecting Chitinase Production

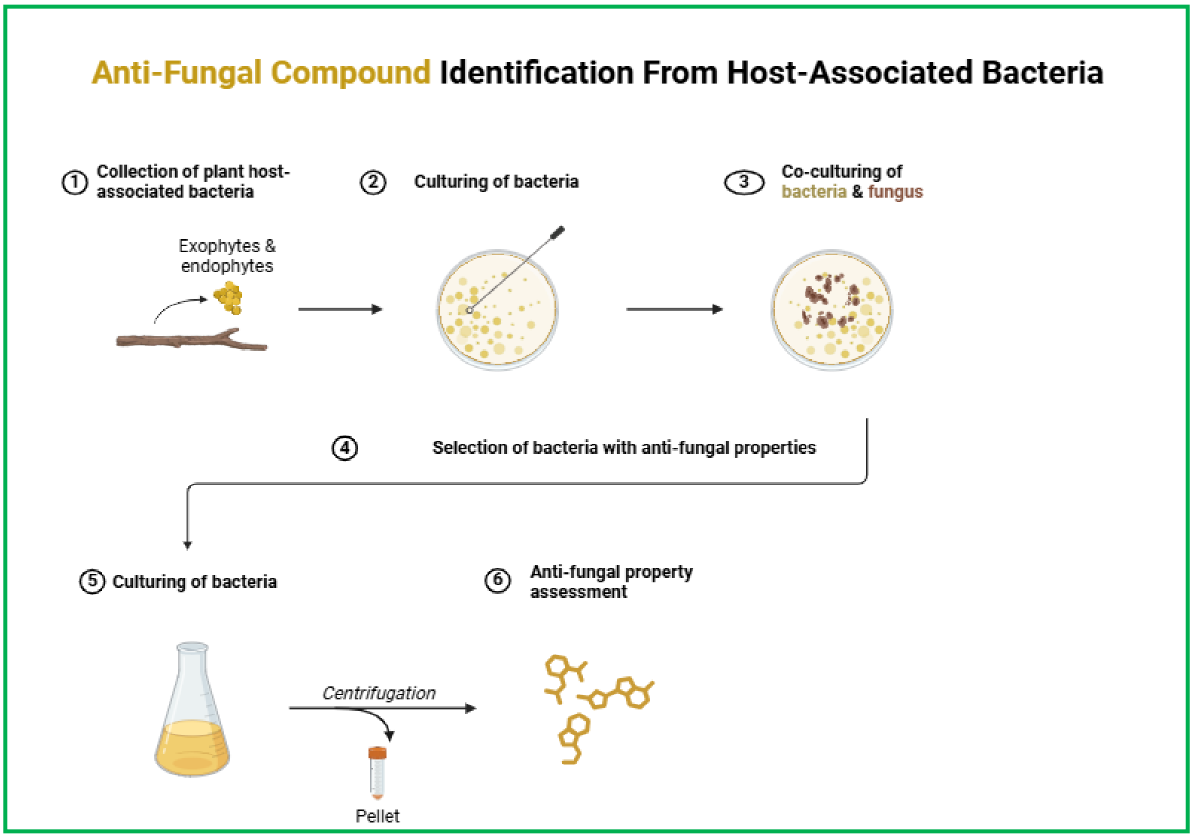
Chitinase production may be influenced by somenutritional and environmental factors. Nutritional factors involve the source of nitrogen and carbon and the presence of organic or inorganic salts. Environmental factor involves the temperature, pH, agitation, and incubation period (Gomaa 2021)

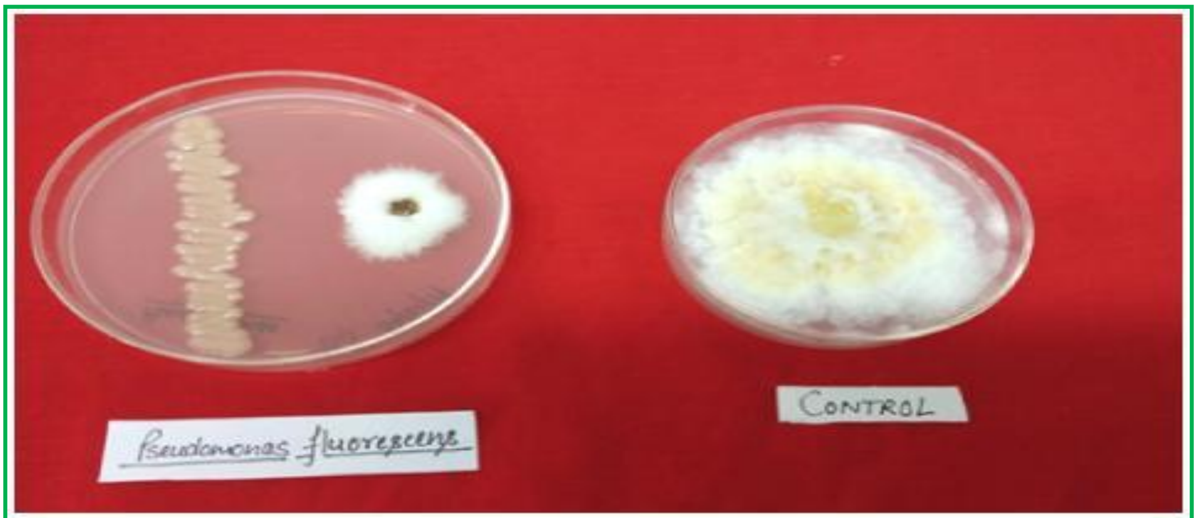
Microbial Chitinases in Plant Defense

Plants are known to produce certain enzymes against phytopathogens and this mechanism can be involved in strengthening the defense. Nowadays, chitinase genes have been cloned and expressed into various plant species, resulting in improved disease resistance in the developed transgenic plants. Chitinases break down the chitin in the cell walls of pathogenic fungi and insect exoskeletons. By degrading these structural components, chitinases weaken the pathogens and help reduce their ability to infect or damage plant tissues. The activity of chitinases can trigger plant defense responses. When plants encounter chitinase activity, they often respond by activating various defense mechanisms, including the production of phytoalexins (antimicrobial compounds), pathogenesis-related proteins, and reinforcement of cell walls. Some microorganisms produce chitinases as part of their strategy to outcompete plant pathogens. By breaking down chitin, these microorganisms can inhibit the growth and colonization of pathogenic fungi in the rhizosphere or on plant surfaces.Certain beneficial microorganisms that produce chitinases can enhance plant growth and health by improving nutrient availability, reducing pathogen loads, and promoting overall plant resilience. This symbiotic relationship helps plants to better withstand stress and disease. Microbial chitinases are often used in biocontrol strategies. Microorganisms that produce these enzymes can be applied to crops to prevent or reduce infections by pathogenic fungi and insects, offering a more environmentally friendly alternative to chemical pesticides.Several bacteria like *Xanthomonas, Serratia, Chromobacterium, Klebsiella, Pseudomonas, Clostridium, Vibrio, Arthrobacter, Beneckea, Aeromonas*, and *Streptomyces* produce chitinase. In fungi including *Trichoderma, Penicillium, Lecanicillium, Neurospora, Mucor, Beauveria, Lycoperdon, Aspergillus, Myrothecium, Conidiobolus, Metharhizium, Stachybotrys* and *Agaricus* were found as main source of chitinase.The transgenic plants show disease resistance and even late appearance of disease symptoms.

Application

The endochitinase gene derived from *Trichoderma harzianum* has been studied for disease resistance against wilt problems in Guava.The technique of expressing the endochitinase gene in the plant system to induce resistance against fungal pathogens has been successfully demonstrated in apples, cotton, broccoli, and rice. In transgenic plants, the efficacy of the chitinase gene is confirmed by a reduction in disease symptoms by pathogenic fungi. Transgenic tobacco and *Brassica napus* both revealed escalated resistance towards *Rhizoctonia solani*, following constitutive expression of the chitinase gene. Transgenic peanut plants with tobacco chitinase showed partial resistance to *Cercospora arachidicola*. To enhance the antifungal response of litchi (*Litchi chinensis* Sonn.), transgenic plants were generated by transferring the rice chitinase gene driven by a maize ubiquitin promoter. Co-expression of basic chitinase gene from rice and acidic â 1-3 glucanase gene from alfalfa in tobacco against *Cercospora nicotianae*, it has been observed that when both genes were expressed in a single plant enhanced more resistance as compared to expression of either of the single genes (Jalil *et al.* 2015)





In Vitro effect of bacterial antagonist on pathogenic fungal growth

Conclusion

Chitinases are prime molecules of interest in today's scenario to utilize them in a variety of ways to improve the soil as well as plant health. These enzymes are classified into various types based on their structural and functional properties. One class of chitinase is not always equally effective against other source of chitin. Thus, there is a need to isolate and identify the chitinase of broad-spectrum activity. In addition, the reaction condition of the enzyme must be known before its use in an open environment and affected site. Besides this, there is a need to enhance the basal level of chitinase production by using recent approaches to genetic engineering. Therefore a coordinated strategy using the above plans may be meaningful to access the full potential of chitinolytic organisms in rendering plant defense.

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