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Review Article

Chitinase production in organisms: a review

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Abstract
Chitin has dive in various forms of life (bacteria to higher plants and animals) as one of the most durable, richest biopolymers distributed widely in nature both in the terrestrial and marine environments. Chitinase are chitin degradable enzyme have control of phytopathogens, physiological functions and degradation of chitinous waste. Interest in chitin wastes utilization is increasing day by day because of its natural resistance against degradation. The review focused on different sources of naturally chitinase production in organisms was discussed.
Key words: Chitin, sources of chitinases, chitinase functions

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INTRODUCTION

Chitin is the second most bounteous, non-toxic, inelastic, biocompatible and biodegradable natural polymer after cellulose. Structurally, chitin is a rigid, toughest and crystalline, consist of monomers of N-acetyl-glucosamine (NAG) which are attached by 1→4 linkages (Shahidi *et al.,* 1999; Tamura *et al*., 2011). Chitin is categories into three different forms on the basis of X-ray diffraction namely; α, β and γ chitin. Parallel and anti-parallel chains, arrangement of small units, in these structures are existed in β chitin and α chitin, respectively. The α-chitin forms crystalline, tight and compact structure. While β chitin possess weak bond between molecules, therefore, forms less stable structure (Zhang *et al*., 2014).

Most abundant form of chitin is αchitin, present notably in the cell wall of fungi and invertebrates. Chitin present in enormous amount in the form of insect exoskeleton, fungi cell wall and in the shell of crustaceans (Souza *et al.,* 2011; Arbia *et al.,* 2013; Zhang *et al*., 2014). Chitinases are chitin degrading enzymes that break down 1→4 β-glycoside bond in chitin to produce monomers and oligomers of NAG (Howard *et al.,* 2003). There are two main categories of chitinases (endo-chitinases and exo-chitinases) differentiated on the basis of their mode of action (Hamid *et al.,* 2013). Endo-chitinases breakdown chitin from the inner sites at random position and give rise to small multimer units such as chitotetrase and chitotriose. Exo-chitinasesas name indicating attack chitin structure at external ends, categories into two (chitobiosidases and β-1, 4 N-acetylglucosaminidases) subgroups on the basis of its step by step gradually degradation. Release of diacetylchitobioseat occurs as a result of enzymatic action of chitobiosidases. The β-1,4 N- acetylglucosaminidases, which further breakdown chitobiosidases product into oligomer units.

On the basis of amino acid sequences, chitinases grouped into three main families of glycosyl hydrolases (18, 19 and 20). Different mechanisms (including acid-base and substrate based) are used for the breakdown of chitin by chitinases (Henrissat and Bairoch, 1993; Brameld and Goddard, 1998). In general, type of organism comes under the category of family No. 18are viruses, bacteria, fungi, insect and mammals, while, family 19 include plant chitinase. Human chitinase belong to family 20of chitinases (Henrissat and Bairoch, 1993; Watanabe *et al.,* 1999; Funkhouser and Aronson, 2007). Different organisms like bacteria, fungi, insect, mammals and plants have potential to produce chitinases (Kasprzewska, 2003; Merzendorfer and Zimoch, 2003; Karthik *et al.,* 2014). This review focused together information to know when chitinases

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produce in the organisms. What are the roles of chitinases in different organisms in normal circumstances?

Bacterial Chitinases

Bacterial chitinase comes in the category offamily 18 of glycosyl hydrolases. Based on the amino acid sequence, bacterial chitinases are divided into three main subfamilies A, B, and C (Watanabe *et al.,* 1999). Research conducted on characterization and purification dictates variation in the bacterial chitinases, molecular weight ranging from 20 to 80kDa (Liu *et al.,* 2010; Beier and Bertilsson, 2014; Karthik *et al.,* 2015). Optimum pH varies from 5 to 8, while, optimum temperature ranges from 30º to 40º C (Beier and Bertilsson, 2014; Karthik *et al.,* 2015). Bacteria produce chitinase to fulfil its nutritional requirement and for parasitism. (Bhattacharya *et al.,* 2007; Funkhouser and Aronson, 2007). Among microorganisms, bacterial chitinases play substantial part in the chitin degradation process, to accomplish its energy demand (Keyhani and Roseman, 1999; Patil *et al*., 2000; Huang and Chen, 2005; Bhattacharya *et al*., 2007). Chitinase activity has been extensively found mainly in *Streptomyces, Serratia, Clostridium, Aeromonas, Arthrobacter, Vibrio, Klebsiella, Pseudomonas, Chromobacterium* and *Bacillus* species (Wang *et al.,* 2008; Han *et al.,* 2009; Narayana and Vijayalakshmi, 2009; Liu *et al.,* 2010; Kuddus and Ahmad, 2013; Islam and Datta, 2015).

Fungal chitinases

Chitinases have been found in fungus. Fungal chitinasecarries multiple functions such as nutritional, morphogenesis, as a major structural component andpathogenesis (Sahai and Manocha, 1993; Cohen-Kupiec *et al.,* 1998). Lee *et al.* (2009) reported fungal chitinase of *Penicillium sp.*, The fungi metabolism machinery like bacterial chitinaseshas ability to produce chitinases so that it degrade chitin effectively and derive energy from it for survival. The fungal cell consists of chitin. During fungal development, chitinases assist in breakdown of old cell wall chitin as a result of which new chitin formation take place (Dahiya *et al.,* 2006).

Due to its distinctive mycoparasitic activity in *Trichoderma* species, makes it effective biocontrol agent against many plant pathogenic fungi (Spiegel and Chet, 1998).Different species of fungi which possess chitin are *Trichoderma, Penicillium, Metharhizium, Aspergillus, Mucor, Neurospora, Lycoperdon, Beauveria, Myrothecium, Conidiobolus, Stachybotrys* and *Agaricus* (Lee *et al*., 2009; Sharma and Shanmugam, 2011; López-Mondéjar *et al.*, 2012; Islam and Datta, 2015).López-Mondéjar *et al.* (2012) depicted the role of *Trichoderma harzianum*in nurseries of greenhouse for the biological control of *Fusarium oxysporum*.

Insect chitinases

In insects, clear picture of chitinases functioning is noticed during molting. Insect chitinase plays major role during this process (removal of external old skeleton). However, chitinase expression in insect is highly specific comes under hormonal control, avoiding its premature expression or over expression. In insect, ecdysis is a two-step process, in which both endo and exo-chitinases work together for transformation from larvae to the adult. Firstly, endochitinases breakdown cuticle into small subunits called chitooligosaccharides. These units of endochitinases, further hydrolyzed into N-acetylglucosamine (NAG) with the help of exo-chitinases.

The NAG act as building block for the synthesis of new exoskeleton (Royer *et al.,* 2002; Takahashi *et al*., 2002; Merzendorfer and Zimoch, 2003).Different chitinases are involve in molting of *Tribolium castaneum* (Zhu *et al.,* 2008). In complete molting cycle, both old cuticle degradation and new cuticle synthesis take place simultaneously, the purpose behind this simultaneous process, is to protect of the nascent cuticle from activity of chitinases and other enzymes of molting (Chaudhari *et al.,* 2011).There are different orders of insects including hymenopterans, coleopterans, lepidoterans, and hemipterans. Similarly, most prominent species (*Bombyx mori, Tribolium castaneum, Culex pipiens, Manduca sexta,* and *Apis mellifera etc*) of insect in which chitinase has been estimated (Takahashi and Kamimura, 2002; Zhu *et al.,* 2008; Nakabachi *et al.,* 2010; Karthik *et al.,* 2014). There is small range of variation from 40 to 80 kDa in molecular weight of chitinases reported in insects. Koga *et al.* (1997) described insect chitinases in the *Bombyx mori*. Takahashi, *et al* (2002) narrated BmChiR1, new chitinase-related gene in the *Bombyx mori* and also explain its function in metamorphosis. Chitinolytic enzymes also play additional role in protection against insect own parasite (Brzezinska *et al*., 2013; Islam and Datta, 2015).

Mammalian chitinases

Mammalian chitinases divided into two main categories; one is true chitinases and other is protein chitinases. The role of both chitinasesare different from each other. True chitinases participated in the breakdown and digesting activity of chitin. Although, protein chitinases not involve in any breakdown process, it just binds with chitin (Hamid *et al.,* 2013). Type of chitinases found in human is called chilectins which are non catalytic activity (Vega and Kalkum, 2011; Adrangi and Faramarzi, 2013).There is not any process of chitinase biosynthesis in human. Dušková *et al.* (2011) narrated chitinolytic bacteria present in the gastrointestine of human tract. In the gaucher patient, the first reported chitinase was chitotrisidase produced by macrophages. This enzyme showed antifungal potential (Stoykov *et al.,* 2015). Another enzyme called acid mammalian chitinase showed potential to degrade chitin and role in innate immunity (Boot *et al.,* 1995; Bussink *et al.,* 2006). Different researchers reported that chitinases facilitate the mammals in protection strategy against the chitin containing pathogens (Renkema *et al.,* 1998; Bussink *et al.,* 2006; Stoykov *et al.,* 2015).

Plant chitinases

Plant do not produce chitinase all the time, there are specific stimuli (phytopathogenic attack) which activates chitinase production in the plant. Production of chitinases in response to various condition of fungal challenge was observed in plant (Schlumbaum *et al.,* 1986; Kubicek *et al.,* 2001; Van *et al.,* 2006). The predictable role of chitinases in the plant is noticed in the form of defense mechanism. Phytopathogenic attack act as a chitinase induction signal and facilitates plant in self defense against pathogens. In some plant chitinase production is also found during embryogenesis, growth of seedling and seed germination (Schlumbaum *et al.,* 1986; Collinge*et al.,* 1993; Kasprzewska, 2003; Van *et al.,* 2006; Kirubakaran and Sakthivel, 2007). Experimentally, the effect of purified form of barley chitinasesis noticed for preventing the growth of parasitic fungal hyphae (Kirubakaran and Sakthivel, 2007). In various plants, the enhanced protection against pathogenic fungal is produce by using expression of heterologous chitinase gene (Schickler and Chet, 1997; Punja, 2001). During fruit ripening, as in the developmental phase of fungi, ethylene acts as an inducer for the chitinase formation (Clendennen and May 1997; Kasprzewska, 2003). Major site of chitinase production in the plant are tissue specific (present in flowers, seeds, stems and tubers) (Van *et al.,* 2006).Environmental stresses such as osmotic pressure are also responsible for chitinases production in the plant (Yun *et al.,* 1996). Researchers upsurge phytopathogens resistance in the plants by introducing chitinase genes of *Trichoderma species* into the plant (Kubicek *et al.,* 2001). Chitinases that produce in the plants are endochitinase that inhibits fungal growth (Islam and Datta, 2015). In plant, molecular weight of chitinases is ranged from 20 to 40 kDa (Hamid *et al.,* 2013; Roopavathi *et al*., 2015).Patel *et al.* (2010) studied new chitinases enzyme, purified and characterized from *Ipomoea carnea.* In genetic engineering, bacterial chitinases gene are employed to provide protection in the plant by generating the transgenic plant, introducing chitinases gene for the control of fungal diseases (Sharma *et al.,* 2011).

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