

Application of Oligosaccharides to Induce Innate Immunity in Plants

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Abstract. Plants have innate immune systems and protective mechanisms to resist the attack of pathogenic microorganisms. Unlike mammals, they lack mobile defensive cells and therefore rely on autonomous cell events for protection. These cells have a wide range of recognition capabilities for detecting pathogens, thus filling the gap of the adaptive immune system. These protective mechanisms will remain inactive or latent until they are activated after exposure to inducers or application of stimuli. Only after they are affected by pathogens or the same elicitors do they begin to show an active state. The role of oligosaccharides in plant immunity is gradually attracting widespread attention. Therefore, this paper summarizes the functions of oligosaccharides related to plant immunity and provides examples of induced defense events. The recognition of sugar molecules as signal molecules in plants has also been proposed. In this review, we focus on the development and application of oligosaccharides in plant immunity and their potential value in agricultural field.

1 Introduction

Plants have an immune system that can resist the invasion of bacteria, oomycetes and fungi. The initiation of defense responses involves the detection of microbial presence through highly conserved molecular patterns called PAMPs, which are secreted by microorganisms or released by cell wall hydrolases during plant-microbe interactions. These PAMPs trigger the so-called PAMP-induced immune (PTI) defense response during pathogen infection. Therefore, they are considered to be universal elicitors, that is, compounds that can trigger plant defense responses. These universal elicitors may also be derived from plant microorganisms that degrade pathogen cell walls through enzymes, and are therefore called DAMPs^[1]. These general agonists contain a variety of biochemical categories, such as carbohydrates, lipids and proteins. In plant-microbe interactions, sugar is essential for the energy required for fuel defense, and also acts as a signal for defense gene regulation. Induced resistance to pathogens is a crop protection strategy currently under study^[2]. Although pesticides are still the main means of crop protection, some of their effects on environmental quality, human health, and selection of resistant strains have prompted the development of new strategies in the context of sustainable agriculture. In view of these circumstances, induced resistance has become a focus of attention, because it helps to activate plant defense mechanisms, reduce the use of chemical

synthetic pesticides, and may even produce efficient, stable and environmentally friendly next-generation commercial products, thereby increasing yield, reducing losses, and promoting plant growth. Exploring the characteristics of different glucans in fungi, oomycetes and bacteria can not only deeply analyze and understand the interaction between plants and pathogens, but also provide a new perspective for revealing their potential as plant protection defense inducers. For example, trehalose is a non-reducing disaccharide widely found in many species. Studies have shown that the application of trehalose treatment in wheat can significantly enhance the resistance of wheat to powdery mildew. Further studies revealed that trehalose treatment significantly enhanced the defense response of wheat and promoted the activities of resistance-related enzymes phenylalanine ammonia lyase and peroxidase, thereby enhancing plant resistance. In addition, kelp polysaccharides belong to a class of water-soluble β -1,3-glucans, which can guide a variety of plants such as tobacco, Arabidopsis and rice to produce defensive responses. This induction makes the plant's immune pathway dependent on the salicylic acid signaling pathway, resulting in the accumulation of phytoalexins and increased expression of a series of pathogenesis-related proteins. This article focuses on the functions of oligosaccharides related to plant immunity and provides examples of induced defense events. It is also proposed

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to identify sugar molecules as signal molecules in plants. In summary, this review summarizes the role and recognition mechanism of oligosaccharides as defense inducers in plant immunity, and focuses on the potential application of oligosaccharides in agriculture, in order to provide reference for further research and application of oligosaccharides in plant immunity.

2 Plant immune response and resistance induction

Plants are different from mammals. Mammals have defensive cells and somatic cells, which move in animals to cope with diseases in various parts of the body. In contrast, plants rely on the innate immunity of each cell and activate their own immune responses through signals from the site of infection. In essence, the immune system of plants is mainly divided into two mechanisms. One is microbial-associated molecular pattern-induced immunity (MTI), which uses transmembrane pattern recognition receptors (PRRs) to respond to microbial conserved molecular patterns (MAMPs) [3] on plant cell membranes. Another is effector-induced immunity (ETI), which includes binding nucleotide-leucine-rich repeats (NB-LRR), a receptor kinase that recognizes effectors released by different pathogens and activates the corresponding defense response. MTI immunity first recognizes MAMPs by pattern recognition receptors in plants, and then pathogens release pathogenic effectors to inhibit the MTI immune response. These pathogenic effectors are recognized by NB-LRR receptors in plant cells, triggering the ETI immune response of plants [4]. Compared with MTI, ETI reaction is stronger. In addition to the inherent immune system, plants also have the ability to sense specific biotic and abiotic stimuli. This ability is triggered by an inducible defense mechanism, resulting in plants showing a variety of rapid or strong cellular defense responses, or showing both types of responses at the same time [5]. About induced systemic resistance (ISR) [6], when there is a beneficial interaction between plants and microorganisms, microorganisms contribute to plant nutrition, while overcoming biotic or abiotic stresses, improving plant defense capabilities, and effectively preventing a wide range of pathogens [7]. This type of induced resistance is considered to be independent of SA and does not involve changes in the expression of major defense genes. This may be because such changes may lead to a significant increase in resource input, thereby reducing host fitness, as revealed by previous studies [8]. In a broad sense, elicitors refer to chemical substances or biological factors from a variety of sources that can cause physiological and morphological changes in the target organism. They include non-biological properties, such as metal ions and inorganic compounds, and biological properties, such as from fungi, bacteria, viruses (or their fragments), plant cell walls and chemicals released when attacked, or pathogens present in plants. [9] Another defense response produced by the plant itself, such as the bean leaves extracted from the same plant, can trigger a strong defense response and

release reactive oxygen species. It is worth emphasizing that these elicitors have many advantages in the field of crop protection, mainly because they are not limited by specific mechanisms of action.

3 Oligosaccharides as immune inducers

Different types of PAMP / MAMP can be used as inducers, such as oligosaccharides (OGA). Because these molecules play an important role in many pathogenesis, recent studies have focused on these molecules. [10] In nature, when plants interact with pathogens, plants secrete β -1,3-glucanase, chitinase and chitosanase to degrade the cell wall of pathogens to resist the invasion of pathogens. Pathogens degrade plant cell walls by releasing polygalacturonases and pectinases to invade plant cells. In this interaction process, oligosaccharides are produced, and these oligosaccharides can be recognized by plants, thereby activating their own immune response and enhancing disease resistance. In recent years, significant progress has been made in the research of oligosaccharide inducers, and good results have been achieved in many plants such as *Arabidopsis thaliana*, rice and tobacco. For example, in 2010, Yin et al. reported the role of oligosaccharides in plant vaccines, which made oligosaccharides plant immune inducers a hot topic for researchers. At present, the application of chito-oligosaccharides has been widely discussed in the field of oligosaccharide plant immune inducer research, and its effect is remarkable. As one of the earliest studied oligosaccharide inducers, chito-oligosaccharides are derived from the deacetylation of chitin, the main component of fungal cell wall. β -1,3-glucan is one of the main components in the cell walls of some bacteria, fungi and plants [11]. This oligosaccharides may be produced when plants interact with pathogens. Linear β -1,3-glucan oligosaccharides extracted and purified from brown algae, such as kelp starch, can induce the accumulation of ROS, salicylic acid, phenylalanine aminotransferase (PAL) and lipoxygenase activity in tobacco cells [12]. In addition, oligogalacturonic acid is a linear polysaccharide composed of 2-20 α -1,4-galacturonic acids, which is the product of pectin degradation of plant cell wall components. The study also found that 30 % acetylated oligogalacturonic acid can increase the number of papillae on the surface of wheat cells, thereby reducing the number of haustoria when pathogenic fungi infect plants. In addition to the above-mentioned oligosaccharides, many other oligosaccharides have been further studied because of their potential to induce plant disease resistance, as shown in Table 1. In recent years, alternatives to commercial fungicides have emerged. Starting in 2014, a team of scientists began studying a complex containing oligosaccharides as a resistance inducer to explore its potential advantages. The results showed that spraying the compound within an interval of 7-14 days could reduce the severity of the disease. In addition, this product shows a persistent and cumulative effect. In a subsequent study, the researchers found that the complex increased resistance to the disease, and also seemed to

involve the salicylic acid pathway (SA) in plant defense. [13] Previous studies have reported that the induction of plant immunity induced by COS-OGA involves the up-regulation of PR protein and protease expression and the increase of peroxidase activity during the accumulation of SA, suggesting the mechanism of inducer in systemic acquired resistance. These findings make us realize that oligosaccharides are a promising research topic in the development of ecological agriculture. However, because the structure of the sugar chain is much more complex than that of nucleic acids and proteins, [14] the determination and chemical synthesis of sugar bonds are also more difficult than nucleic acids and proteins, which limits the research and development of glycan functions. Therefore, in the field of plant protection, the development and research of glycans still face great challenges.

Table 1. Application of oligosaccharides to their respective pathogens in different cultures

Inductor	Plant	Disease	Pathogen	Application	Quote
Curdlan oligosaccharide	Potato	Late blight	Phytophthora infestans	Infiltration	[15]
Oligogalacturonides	Arabidopsis		Pectobacterium carotovorum sp.	Foliar.	[16]
Laminarin (β-1,3-glucan)	Tobacco	Bacteriosis	Erwinia carotovora subsp.	Addition of elicitor	[12]
Chito-oligosaccharides (COS)	Blackberry	Biotic and abiotic stress		Foliar spray	[17]

4 Perception of sugars in plants

Sugar also acts as a signal molecule in plant immunity [18]. The primary induced defense line shared by plants and animals is MAMP-triggered immunity and PAMP-triggered immunity (MTI/PTI). This immune mechanism is based on the recognition of highly conserved structures of microorganisms that do not exist in host organisms. In plants, these MAMPs / PAMPs are detected by pattern recognition receptors, and a variety of carbohydrate structures are identified as MAMPs / PAMPs, such as peptidoglycan (PGN) [19]. Such as trehalose, raffinose or galactose, etc., can activate the plant's defense response, enhance plant resistance to pathogens. In Arabidopsis cell suspension, sucrose, glucose and fructose induce PR protein transcripts PR-Q and PAR-1 in tobacco by SA-independent pathway [20]. The mechanism of plant sensing sugar is extremely complex, and its perception is achieved through hexokinase [21]. For example, in Arabidopsis thaliana cells, the glucose sensor HXK1 plays a role in sensing hexose [22]. However, there are also studies reporting an independent pathway for hexokinase [23]. In addition, sucrose and other disaccharides seem to be perceived at the plasma membrane level [24]. At present, research on

sugar transporters is still in progress, which will help to unravel the mechanisms related to sugar perception.

5 Conclusion

At present, it can not be ignored that oligosaccharides play a certain role in plant immunity. However, due to the high complexity of the related mechanisms, their practical significance in plant-microorganism interaction is still partially unrevealed. Unlike pesticides that directly affect pathogens, induced resistance involves plant perception of inducers and subsequent plant responses, and is undoubtedly affected by many factors. In the field of crop protection, induced resistance is seen as an alternative approach aimed at limiting or reducing the use of chemical synthetic insecticides. Nowadays, consumers are increasingly inclined to produce food in a more environmentally friendly and sustainable way, reducing pesticide use, which is beneficial not only to farmers and consumers in terms of economy, environment and quality of life [25]. At present, the oligosaccharide products that have been temporarily registered for pesticides include the following : 0.4 % oligosaccharide produced by Guangdong Yuanfeng Bioengineering Co., Ltd., Haopu (containing 2.8 % and 2.0 % amino oligosaccharide) launched by Dalian Kaifei Chemical Co., Ltd., and OS-Sterling (containing 0.5 % amino oligosaccharide) developed by Hebei Nangong Biochemical Plant. Different types of oligosaccharides have diverse plant immune function activation and mechanism of action. Since the structure of glycans is much more complex than nucleic acids and proteins, for example, linear molecules composed of three nucleotide bases or three amino acids have six possible sequences, while trisaccharides composed of three hexoses have up to 1,056 to 27,648 possible sequences. Therefore, it is much more difficult to determine and chemically synthesize glycan bonds than nucleic acids and proteins, which greatly limits the research and development of glycan functions. Oligosaccharides can degrade pesticide residues in plants and stimulate plant immunity. At present, oligosaccharides are mainly used as plant sprays in the agricultural field and have considerable research potential. However, it is much more difficult to determine and chemically synthesize glycan bonds than nucleic acids and proteins, which greatly limits the research and development of glycan functions. Therefore, the development and research of glycans in the field of plant protection still faces many challenges and arduous tasks.

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