Properties and Bioactivity of Chitosan

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Chitosan (CS) is a natural biopolymer derived by deacetylation (N-acetyl-D-glucosamine to D-glucosamine unit) of chitin.

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1. Properties of Chitosan

Chitosan (CS) is a natural biopolymer derived by deacetylation (N-acetyl-D-glucosamine to D-glucosamine unit) of chitin, which has been studied for its effectiveness against bacterial, fungal, and viral pathogens [1]; in addition, CS has demonstrated minimal toxicity in mammals and humans [2]. CS owns a molecular weight (MW) between 10 and 1000 kDa [3], but the MW and degree of acetylation (DA) depend on the type of source [4]. In principle, the DA is defined as the proportion of N-acetyl-D-glucosamine units with respect to the total number of units [3][5]. CS can be obtained from several sources including sea animals (e.g., annelida, mollusca, coelenterate, and crustaceans), insects (e.g., scorpions, spiders, ants, cockroaches, and beetles), microorganisms (e.g., algae, yeast, fungi, ascomycetes, and spores), among others [6]. The different MW and DA of chitosan also influence its physicochemical properties, such as solubility, appearance, rheological properties, among others [3], directly impacting the bioactivity and toxicity of the polysaccharide [2]. CS is a viscous polysaccharide that forms more structured gels when its MW increases; furthermore, it is considered as a pseudoplastic material since its viscosity is a function of concentration, temperature, and rate of shear [3]. The solubility of CS varies depending on DA, and it is insoluble in neutral-alkaline pH but highly soluble in acid pH due to the proportion of protonated amino groups ($-NH_2$) that are positively charged [7].

Depending on its DA, the CS-polyelectrolyte takes different behaviors in solution; for example, if DA is greater than 50%, the molecule maintains the hydrophobic characteristics of chitin, while for a DA between 20% and 50%, the molecule becomes less hydrophobic, and CS, having a DA less than 20%, is considered a highly hydrophilic cationic polyelectrolyte [8]. CS can form amorphous and complex three-dimensional structures due to its polycationic nature that allows it to interact electrostatically (by hydroxyl groups), or to form covalent bonds by $-NH_2$ groups, with other molecules, such as metals, surfactants, proteins, and polyanions [6].

2. Bioactivity of Chitosan

Bioactivity is defined as the ability of a material to chemically interact with compounds, microorganisms, or pathogens [9]. The bioactivity of CS has been related to the antimicrobial, antifungal, and antiviral properties, providing important contributions in different applications, such as additives, agro-food, pharmaceutical, and medical [10]. Bioactivity is determined by intrinsic, environmental, and microbial factors. The intrinsic properties can be modified by varying the temperature and time of exposure when CS is in dispersion [8]. The antimicrobial activity is totally dependent on the pH of the medium and the solubility of CS [11]. For instance, a stronger inhibitory effect has been demonstrated at lower pH, such inhibitory effect decreases as the pH increases, and this is attributed to the neutralization of the $-NH_2$ groups in the alkaline medium [8][12]. CS can also act as a chelator for metal ions (e.g., nickel, zinc, cobalt, iron, magnesium, and copper) [13], and some studies promote its addition to modify the ionic strength [8], however divalent ions have been shown to attenuate the inhibitory activity of CS [14]. According to the insights documented by several authors, the antibacterial effect of CS on prokaryotic cells (e.g., Gram-negative and Gram-positive bacteria) depends on the electrostatic interactions between the biopolymer and bacterial cell wall components (e.g., teichoic and lipoteichoic acids, and lipopolysaccharides), altering stiffness and eventually entering into the cell [15]. The antifungal effect of CS on eukaryotic cells is thanks to the biopolymer cationic power acting on the residues of the mannoproteins (i.e., sialic acid) located on the surface; once inside the cells, CS can chelate metals (e.g., potassium, calcium, and sodium), denature proteins due to the effect of the net negative charge, or interact with DNA via nucleic acid phosphate groups [15]. The antiviral effect may

be due to the interaction between CS and the RNA of viruses via phosphate groups [16]. Thanks to its demonstrated bioactivity, CS has become an alternative in post-harvest food treatment to control the decomposition and extend shelf-life by the inhibitory effect against bacteria, fungi, and viruses [18].

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