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## Antimicrobial Water-soluble Chitosan Based Films for Coating and Packaging Applications

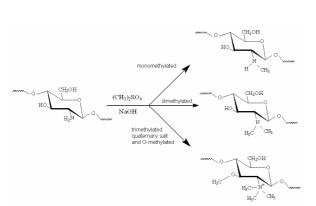
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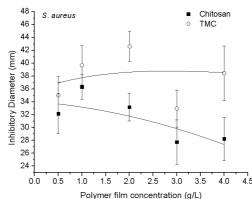
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**Abstract** – Water soluble chitosan derivatives have been explored as antimicrobial agent against a large number of fungi and bacteria both gram-negative and gram-positive. Such derivatives can be obtained by the introduction of permanent positive charges in the polymer chains resulting in a cationic polyelectrolyte characteristic independently of the pH of the aqueous medium. Such solubility makes this polymer interesting for application in foodstuffs such as edible coating, with organized structure, and for the processing of films and nanoparticle arrays for packaging purpose.

Water soluble chitosan derivative can be accomplished by the quaternization of the nitrogen atoms of the amino groups. To attain this, an extensive methylation of chitosan is carried out in suspension of dimethylsulfate, NaOH and NaCl resulting in N,N,N-trimethylchitosan. The synthesis of chitosan derivatives takes place by grafting methyl functionality onto chitosan amino groups at the C-2 position (Figure 1). Evaluation with small concentration of quaternary salts of chitosan in solution reveled that the antimicrobial activity against *E.coli* bacteria is about 5 to 20 times more effective than those reported for commercial chitosans [1]. Such characteristics associated to the non-toxicity and good film forming abilities and high mechanical strength make this derivative attractive for applications as edible coating and active packaging.

In the present study the effectiveness of the parent chitosan polymer and its quaternary salt N,N,N-trimethylchitosan (TMC), both in film form are evaluated against gram-positive (*Staphylococcus aureus*) and gram-negative bacteria (*Escherichia coli*) in function of the polymer concentration. The derivatives were obtained from medium molar weight chitosan purchased from Sigma-Aldrich and the methylation reactions carried out according to adopted procedure [2]. The antibacterial activity, assessed by the inhibition zone formation method [3], showed a higher activity against the gram-positive bacteria for both chitosan and TMC polymers with superior biocide properties to TMC (Figure 2). The activity against *E.coli*, for both polymers, was almost imperceptible for any concentration pointing that in film format the derivative is not effective as reported as in solution against gram-negative bacteria. On the other hand, both films showed to be effective against *S.aureus*, where the film concentration appears to be important, mainly for chitosan. The reduction of antimicrobial activity as the polymer concentration increases (Fig. 2) can be attributed to spatial factor where the active sites can be hindered in more dense films.





**Figure 1:** Schematic representation of the reaction leading to the quaternization of the amino groups of chitosan and resulting in N,N,N-trimethylchitosan.

**Figure 2:** Numeric data of the inhibitory diameter against *S. aureus*, in function of film concentration.

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