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Eco-Friendly Food Preservative

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Abstract: Chitosan (CS), an antibacterial biopolymer with the ability to form films, has been used topically to preserve foods like fruits, vegetables, and even seafood. We must look for more materials with these qualities if we are to have a variety of options for preservation. Sapindus mukorossi, often known as soap nut (SN), is a plant with similar characteristics. The main substance in soap nuts, known as saponin, is what gives them their antibacterial and film-forming properties. There are numerous reports that suggest saponin can be utilised for culinary purposes up to a point. This study assessed the synergistic preservation effect of soap nut and chitosan aqueous solutions on fruits and vegetables. The antibacterial effects of CS, SN, and [CS+SN] are initially seen. Staphylococcus aureus and Escherichia coli were the first microorganisms against which the antibacterial effects of CS, SN, and [CS+SN] in film form were investigated. As anticipated, the film CS+SN demonstrated greater antibacterial action, with zones of inhibition against Staphylococcus aureus and Escherichia coli of 25 mm and 23 mm, respectively. Spraying different regional fruits like lemons, bananas, tomatoes, and oranges with a known concentration of CS+SN allowed researchers to assess the substance's potential as a preservative. The control was water. Different concentrations of [CS+SN] -250ppm and 125ppm, together with CS-500ppm, SN-500ppm, and water, were compared. In comparison to control fruits, fruits sprayed with [CS+SN]-250ppm solution displayed a considerable delay in the change in weight loss, decay percentage, and pH. Additionally, it kept its visual quality better than CS, SN, and control samples.

Keywords: Eco-friendly, edible preservation, chitosan, soapnut, synergistic preservative effect, topical coating

I. INTRODUCTION

Natural way of preserving food is the need of present situation as number of untreatable diseases is growing day by day. The ill effects of utilizing chemicals, synthetic polymers and radiations to preserve food are threatening our livelihood. We need alternative natural ways of preserving food for a sustainable living. Traditionally salt, turmeric, tamarind, oil, and lemon were added to food in order to preserve them in the form of pickles. For preservation of fruits and vegetables which are easily perishable commodities topical application of preservative will be preferred. A preservative should be selected in such a way that it should preserve the colour, texture, flavour and nutrients present in the fruit or vegetable. At the same time it should be harmless even if it is present in it. Biopolymers are being explored to find out their potential as natural preservative. As they can form film, that will act as a shield and protect the food from microbes. Chitosan (CS), the 3-8edible polysaccharide, derived from marine waste is being studied for this purpose. It has antioxidant, antibacterial and antifungal properties. It can be dissolved in vinegar, which is nothing but naturally derived acetic acid. Dilute solutions of chitosan with concentrations ranging from 500-1000 ppm to 1-2% are utilized. These solutions can be sprayed on the fruit or it can be dipped into the solution, to give a coating of chitosan. The demerits of chitosan is that if high concentrations were used the film restricts the respiration of the fruit. In order to enhance its property and also to utilise a new material for preservation, soapnut (SN), with botanical name Sapindus mukorossi is used. Many research works have been published on the 10-14antimicrobial and anticancer activity of Sapindus mukorossi. But not even a single report has been published about the preservative effect of soapnut. Traditionally this natural surfactant has been used for bathing and washing purposes as it exhibits amphiphilic nature. The hydrogel nature of chitosan and soapnut will be an added feature to enhance their activity as topical preservatives. In the present study for the first time soapnut (SN) solution was studied for its preservative effect along with chitosan (CS) solution. They were used as topical preservative agents. The hydrogel nature of CS, SN, [CS+SN] was evaluated based on the values obtained for the % equilibrium water content of the materials in the film form. The antimicrobial effects of CS, SN, [CS+SN] films were also evaluated against Staphylococcus aureus and Escherichia Coli. The potential of [CS+SN] as topical preservative in different concentrations viz. 250 ppm and 500 ppm was compared with CS (500 ppm), SN (500 ppm) and water. For this purpose different regional fruits like lemon, tomato, orange and banana were sprayed with the above mentioned solutions. Properties like change of weight loss, decay percentage fruit gloss and pH were evaluated at regular time intervals.

II. MATERIALS AND METHOD

Lemon, orange, banana, tomato, and sapindus mukorossi (soap nut) were bought at grocery stores in Chennai, India. Chitosan (MMW) was obtained from Aldrich (CAS 44-8869) and utilised as received with a deacetylation percentage of 75–85% and a Brookfield viscosity of 20 cps. Purchased from Merck (India), acetic acid (glacial, 99–100%) and sodium bicarbonate were used without further purification. Throughout the investigation, all solutions were prepared using double-distilled water. 2% w/v of chitosan was dissolved in 0.5M of aqueous acetic acid, and the mixture was stirred at 65°C for 16 hours to create a homogenous stock solution. Using double distilled water, diluted solutions with concentrations of 125, 250, and 500 ppm were created from the stock. The pericarp of Sapindus Mukorossi was dried out in the sun to make the soapnut solution (SN). It was processed via a laboratory mill to create a fine powder with a 40 mesh size. Overnight stirring was used to create an aqueous solution of the Sapindus Mukorossi at 20% w/v. To remove all unextractable material, the extract was filtered through a plastic tea strainer. The gravimetric method was used to assess the extract's water soluble matter content percentage. The pH of the SME solution remained constant (7.2) both before and after extraction, and it was discovered to contain 8% solid. Following the preparation of a 1% w/v stock solution, additional dilutions of 125, 250, and 500 ppm concentrations were made.

III. PREPARATION OF [CS+SN] SOLUTION

The [CS+SN] solution were prepared by simply mixing chitosan and soapnut solutions. It the data on the preparation of different concentrations of [CS+SN] solutions using CS and SN solutions. Chitosan + Soapnut Chitosan (CS) Soapnut (SN) [CS+SN] 125 ppm 125 ppm [CS+SN] 250 ppm 250 ppm. Preparation of [CS+SN] Preparation of CS, SN and [CS+SN] films: 15ml of 1% w/v of CS, SN and [CS+SN] solutions were stirred separately in three different beakers along with 0.1 ml of 25% glutaraldehyde, adjusted to pH 7 with sodium bicarbonate solutions were cast into films and dried using vacuum desiccator. The photographs of CS, SN and [CS+SN] films. The equilibrium water content (EWC) of CS, SN and [CS+SN] films were measured by the weight difference between the swollen hydrogel film and the dehydrated film as described previously.

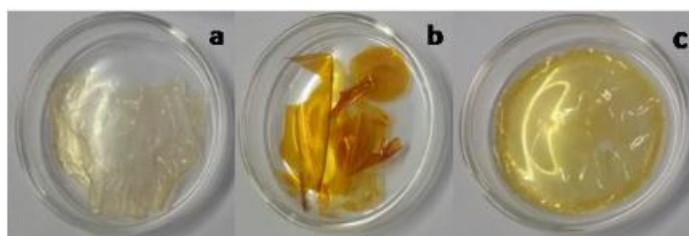


Fig. 1 The photographs of a) chitosan (CS) b) Soapnut (SN) c) [CS+SN] films

A. Antibacterial activity

Nutrient agar was prepared and poured in the sterile Petri dishes and allowed to solidify. 24 h growing bacterial cultures (Staphylococcus aureus and Escherichia coli) were swabbed on it. Then, the test samples was been placed on the nutrient agar plate using sterile forceps. Chloramphenicol was used as standard. The plates were then incubated at 37°C for 24h. After incubation the inhibition diameter was measured. Topical application of CS, SN and [CS+SN] solutions on the fruits. Regional fruits like lemon, orange, banana and tomato were washed with tap water and the excess water on the surface was absorbed by tissue paper. The fruits were placed into a 500 ml plastic container. The CS-500ppm, SN-500 ppm, [CS+SN]-125 & 250 ppm and water were sprayed onto the fruits until wet. Such spraying was done in every alternate day. The observation of morphological features, fruits decay over time was recorded in a notebook and also photographed every day by a digital camera.

B. Fruit Quality Studies

- 1) *Decay Percentage*: The decay percentage of coated and uncoated fruit was calculated as the number of decayed fruit divided by initial number of all fruit multiplied by 100.1
- 2) *Weight Loss*: The fruit samples (3fruit) were weighed at day 0 and at the end of each storage interval. The difference between initial and final weight of fruit was considered as total weight loss at each of the storage interval and calculated as percentage on a fresh weight basis.

- 3) *Determination of Fruit Gloss:* Fruit visual appearance was evaluated subjectively by 5 persons. Fruits gloss was evaluated on a 0 to 10 scale in which 0 = no gloss and 10 = very glossy.
- 4) *pH:* The change in the pH of the fruits used for evaluation was measured from the juices obtained from the fruits at regular intervals. Results and Discussion Chitosan exhibits hydrogel properties due to the presence of groups like -OH and -NH₂ along the polymer backbone. This makes it hydrophilic and retains the water it. Soap nut which is an amphiphilic in nature that is it can absorb both oil as well as water. So in order to evaluate its water absorbing capacity % equilibrium water content was calculated as given in Fig. 2.

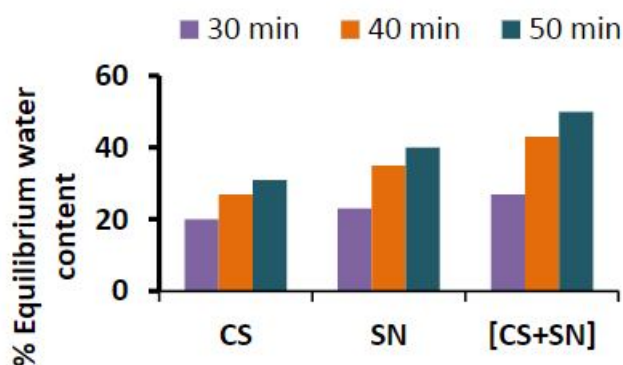


Fig. 2 The % Equilibrium water content of CS, SN and [CS+SN] films

From the Fig. 2 it is clear that compared to chitosan and soapnut, it is the combination of CS and SN [CS+SN] which exhibits higher water intake. This quality is necessary for a preservative to absorb and retain moisture, in order to prevent the surface of the fruit from drying. When the fruit surface is fresh without drying its colour, nutrient, appearance will be enhanced.

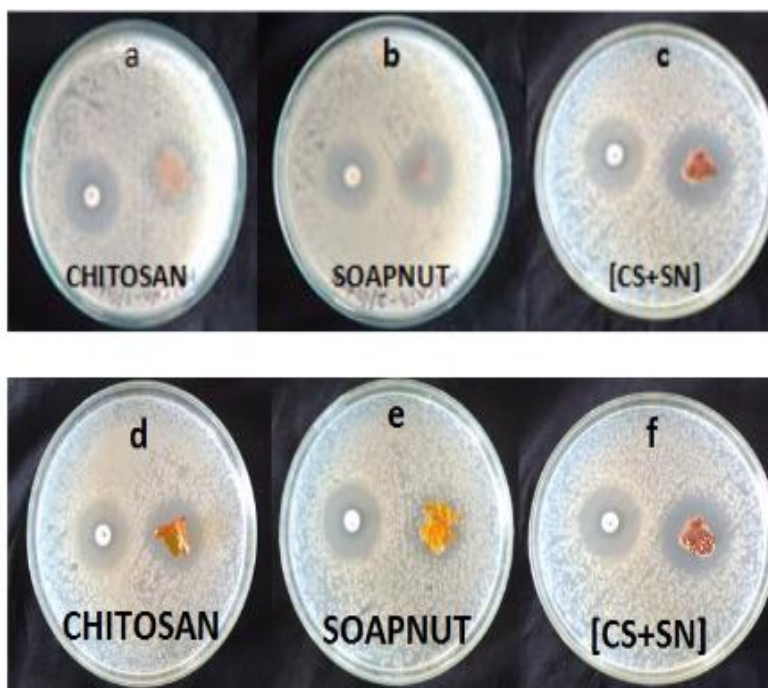


Fig. 3 The photographs showing zone of inhibition of CS, SN and [CN+SN] against (a-c) *Staphylococcus aureus* (d-f) *Escherichia coli*.

Test organism	Sample Name	Zone of inhibition (mm)
<i>S.aureus</i>	Chloramphenicol CS	26 22
	Chloramphenicol SN	23 23
	Chloramphenicol [CS+SN]	25 28
<i>E.coli</i>	Chloramphenicol CS	24 22
	Chloramphenicol SN	24 25
	Chloramphenicol [CS+SN]	24 27

Table 1. The zone of inhibition exhibited by CS, SN and [CN+SN] against Staphylococcus aureus and Escherichia coli.

The antimicrobial efficacy of CS, SN and [CN+SN] against Staphylococcus aureus and Escherichia coli is represented in Fig.3 and Table 1 lists the zones of inhibition (ZOI) values that the samples displayed. By comparing the combined antibacterial impact of [CN+SN] to the separate values, it can be deduced from the ZOI values that it is significantly higher. Thus, it has been demonstrated that the combination of chitosan with soapnut will produce a bio composite with strong antibacterial action. Fig.4 displays the percentage of fruits that have undergone preservation that have decayed. For bananas, it was determined after 15 days of preservation, however for other fruits, it was calculated after 25 days of preservation because they began to rot after that. It is clear from the data that the decay percentage was at its lowest for [CS+SN]125 and [CS+SN]250, while it was at its highest for water, the control.

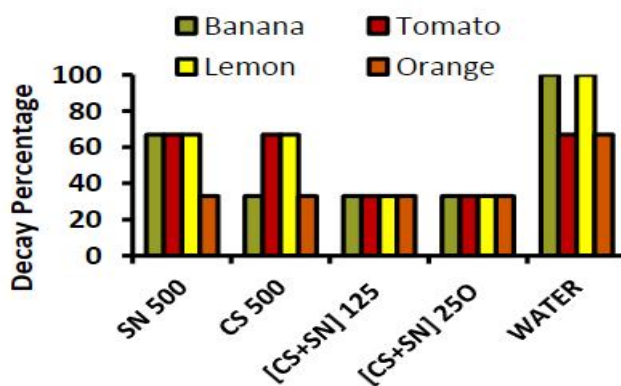


Fig.4 The decay percentage of preserved fruits

In Fig. 5 (a-d), the weight loss percentage of preserved fruits is shown. The bar diagram makes it obvious that bananas might only last for 15 days while other fruits could last up to 25 days with little to no weight loss. Overall, the weight loss for [CS+SN] treated fruits is small for both the concentration. This could be owing to the combined action of hydrogel property of chitosan and soapnut. For soapnut alone the weight loss is larger and can be compared to that of water. This despite being a hydrogel, this might be because to its weak film-forming ability. Table 2 lists the pH of fruits measured at various time intervals. Fruit ripening was indicated by an increase in pH in the samples. Therefore, it is evident that the fruit's preservative coating, which was sprayed on top, has no effect on how ripe the fruit becomes.

Days	Banana	Tomato	Lemon	Orange
0	5.0	4.0	2.1	4.2
5	5.8	4.1	2.4	4.2
10	7.0	4.3	2.5	4.4
15	7.8	4.5	2.9	4.5
20	--	4.7	3.0	5.0
25	--	5.0	3.2	5.2

Table 2 The pH of fruits measured at various time intervals

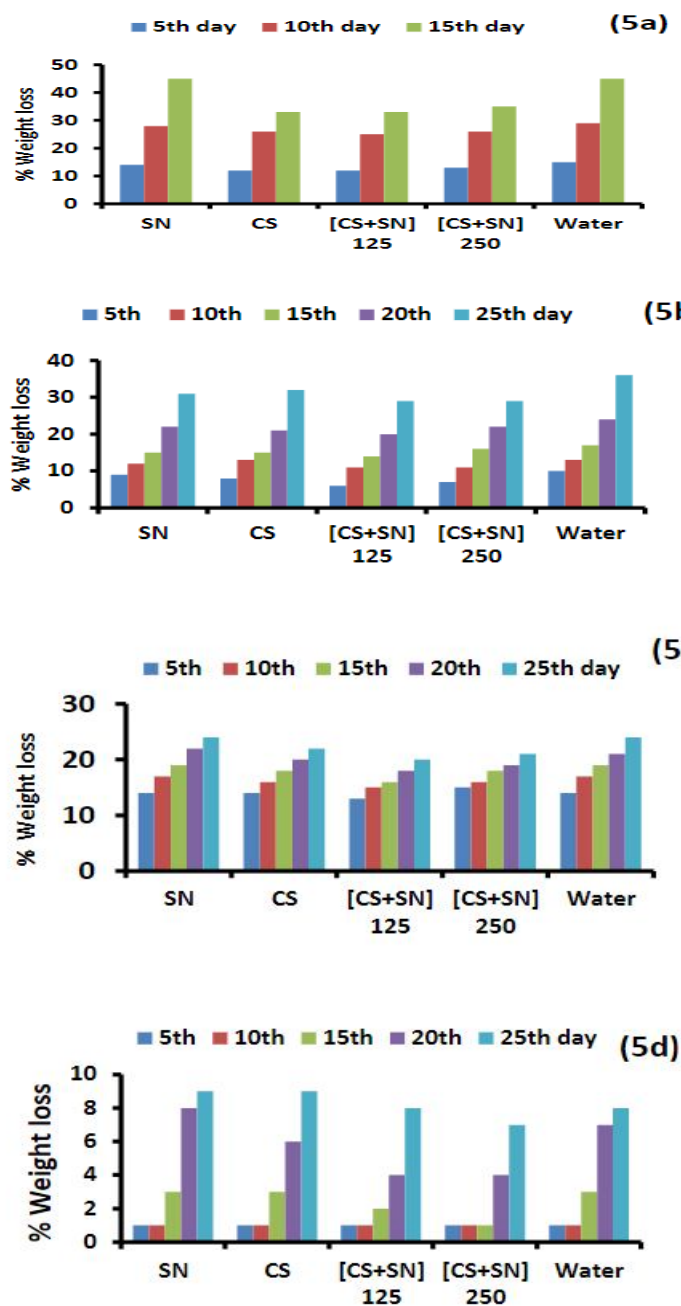


Fig. 5 % weight loss of a) Banana b) Tomato c) Lemon d) Orange

Fruit gloss evaluation done manually also proved that [CS+SN] enhances the preservative effect. Thus among the two concentrations, it is [CS+SN] 250 more effective in preserving the fruit quality.

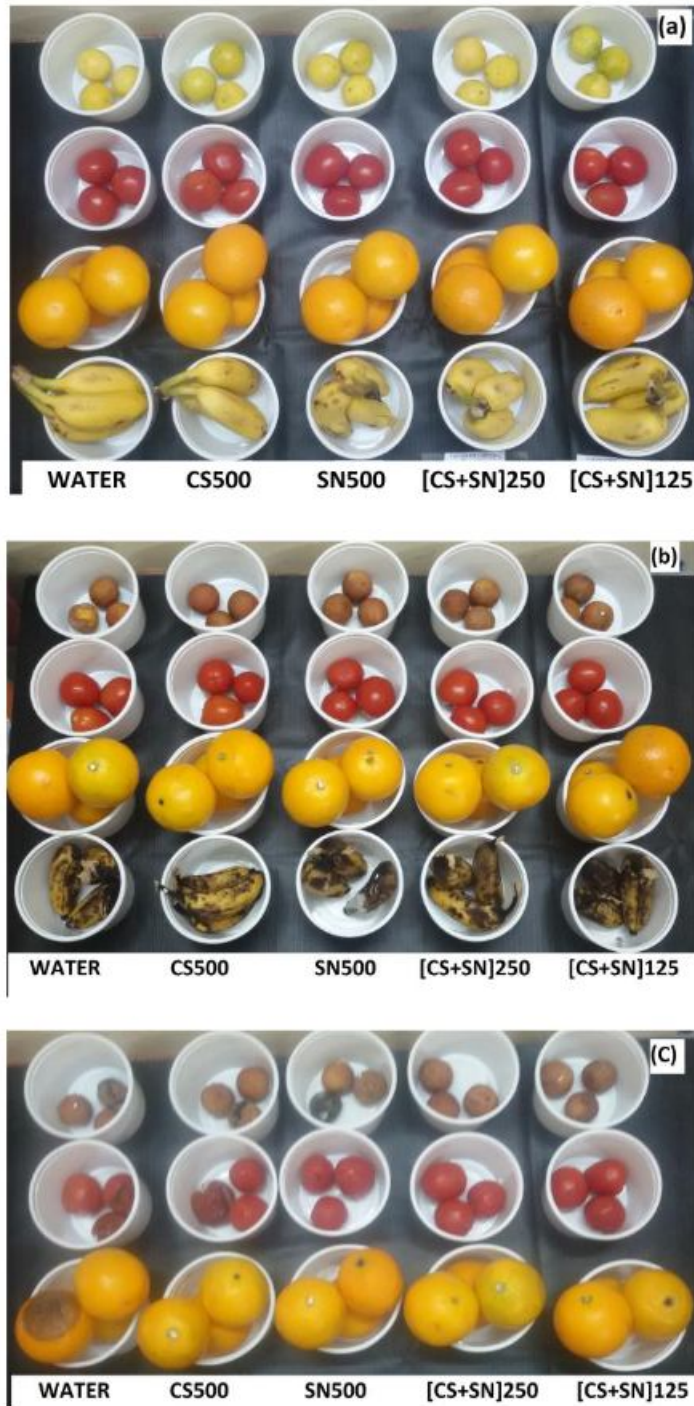


Fig. 6 The photographs of fruits a) 0 day b) 15thday c) 25thday of preservation

When the quality of the fruits were compared, orange, since it is naturally juicy, is extremely efficiently preserved, followed by tomato, then comes lemon, and finally, the least preserved fruit is banana, as can be seen in the picture in Fig. 6. Similar findings have been reported previously, and it has been suggested that lesser quantities of chitosan can be used as a safe substitute to dangerous formalin as a natural fruit preservative.

IV. CONCLUSION

For the first time, the main hypothesis that the mixture of chitosan and soapnut solution will act as an environmentally benign, palatable preservative has been proven. This was demonstrated by using local fruits as models. Topical use of a 250 ppm solution of soapnut and chitosan has produced positive outcomes. This synergistic preservation effect is caused by the hydrogel, antibacterial, and film-forming properties of chitosan as well as soapnut. Out of the fruits used, orange has produced the best outcomes, followed by tomato, lemon, and banana, which has been preserved the least.

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