CHEMICAL ENGINEERING TRANSACTIONS
VOL. 91, 2022
A publication of AIDIC
The Italian Association of Chemical Engineering
Online at www.cetjournal.it
Guest Editors: Valerio Cozzani, Bruno Fabiano, Genserik Reniers

ISBN 978-88-95608-89-1; ISSN 2283-9216
DOI: 10.3303/CET2291098

Anti-Microorganism Activity of Chitosan and Nano-Silicon Dioxide Mixture on Damage Causing Strains Isolated from Postharvest Guava

Dien T. V. To[a,*], Pham T. H. Van[b], Le S. Ngoc[b], Them T. Le[a], Huy C. T. Phan[a], Tran D. Manh[c,*]

[a] Van Lang University, 45 Nguyen Khac Nhu Street, Co Giang District, Ho Chi Minh City, Vietnam
[b] Research & Development Center for Hi-Tech Agriculture, 1 Hamlet, Pham Van Coi Village, Cu Chi District, Ho Chi Minh City, Vietnam
[c] Institute of Applied Technology, Thu Dau Mot University, 6 Tran Van On street, Thu Dau Mot City, Binh Duong 820000, VietNam
dien.tvt@vlu.edu.vn; manhtd@tdmu.edu.vn

Postharvest decay of guava is mainly caused by microorganism species during storage time. Therefore fungal and bacterial species that may cause decay on postharvest guava were isolated and evaluated the antimicrobial and antifungal ability by Low-molecular-weight (LMW) chitosan in combination with nano-silicon dioxide (nano SiO_2) compound was carried out. The study successfully isolated four fungal species, namely *Chrysosporium tropicum, Cladosporium sphaerospermum, Aspergillus wentii, Colletotrichum acutatum* and three bacterial species, namely *Azotobacter* sp*., Escherichia coli, Bacillus subtilis*, which is most likely to cause decay on postharvest guava. It is found that a mixture of 0.04% nano SiO_2 and 1% Low-molecular-weight chitosan 44.5 kDa are capable confront microorganism tested with the highest antibacterial zone diameter and the lowest diameter of growing fungi. This work contributes the potential compound for prolonging shelf-life of postharvest guava.

1. Introduction

The guava (*Psidium guajava* L.) is one of the most popular tropical climacteric fruits and commonly eaten fresh, contains extremely nutritious as well as substances with natural biological activities. However, easy spoilage and short shelf-life are negative features of ripening guava, resulting in long-distance transportation (Hong et al., 2012). It was reported that attacking by microorganisms caused spoilage including *Bacillus* sp*., Listeria* sp*., Staphylococcus* sp*., Vibrio* sp*., E.coli, Pseudomonas* sp*., Aeromonas* sp*., Colletotrichum gloeosporioides*.
Chitosan is a natural edible biopolymer, biodegradable, antimicrobial and antifungal activities (Aider, 2010). Chitosan has been used as a coating material in many fields such as biotechnology, agricultural medicine, food industries, and environmental protection (Nadia et al., 2019). Moreover, chitosan is also efficiently resistive to bacteria, fungi, filamentous fungi, and yeasts which are responsible for plant diseases and spoilage in various fruits and vegetables (Abdullah, 2019; Chao et al., 2019). Low-molecular-weight (LMW) chitosan has been used to inhibit the growth of some positive and negative Gram bacteria (Abdullah, 2019), antifungal agent (Kulikov et al., 2014) reducing of postharvest damage of fruits and vegetables (Chao et al., 2019), fresh food storage (Aider, 2010) as restrict lipid peroxidation on guava storage (Hong et al., 2012).
Nano-silicon dioxide (Nano SiO_2) can improve material properties (Dhanasingh et al., 2011) and for the best properties is at 0.04% (Sun et al., 2016), especially, it in combination with chitosan to enhance chitosan activities applied in food storage (Yu et al., 2012). The combination of chitosan and nano SiO_2 is applying in extensive fields (Witoon et al., 2009). Thanks to the creation of hydro-associated with chitosan molecule, this hybrid compound enhanced permeability and mechanical properties (Yu et al., 2012) improved film formation as well as a semipermeable coating for chitosan (Silva et al., 2011). This combination can also increase the anti-

Paper Received: 18 January 2022; Revised: 8 April 2022; Accepted: 28 April 2022
Please cite this article as: To D.T., Van P.T., Ngoc L.S., Le T.T., Phan H.C.T., Manh T.D., 2022, Anti-Microorganism Activity of Chitosan and Nano-Silicon Dioxide Mixture on Damage Causing Strains Isolated from Postharvest Guava, Chemical Engineering Transactions, 91, 583-588 DOI:10.3303/CET2291098

microorganism capacity of the film (Dhanasingh et al., 2011), and inhibit disease on agricultural products (Silva et al., 2011; Sun et al., 2016). Nearly, 1% (w/v) chitosan in combination with 0.04% (w/v) nano SiO_2 compounds was applied in the fruit storage (Yu et al., 2012) for very well efficiency.

Even though excellent reported characteristics regarding the combination of chitosan and nano SiO_2, there is not any study conducted to find out detail impact of this mixture on the improvement of preservation for postharvest guava, especially anti-microorganisms capacity. Therefore, in this study, the impact of chitosan and nano SiO_2 mixture to microorganisms causing spoilage postharvest guava was carried out with the purpose to find out the suitable concentration of chitosan and nano SiO_2 for postharvest guava preservation that would broaden the fresh fruits consuming markets as well as facilitate abroad export.

2. Materials and methods

2.1. Isolation of bacterial and fungal causing damage postharvest guava

Taiwan guava was grown in Cai Be district, Tien Giang province, harvested after fully blossomed 75 ± 2 days, grouped as uniform size and form regardless of the sign of impact injury and disease. Then, they were transported to the laboratory, washed with distilled water. Each fruit was caused damaged by sterilized 11 blades on its skin and stored at room temperature until there was a sign of damage. These damaged positions were then removed from the fruits, left in 250 mL Erlenmeyer flasks containing 40 mL sterilized distilled water. After shaking for 20 mins to obtain cell supernatant during precipitation, this solution was then diluted to concentration 10^{-1} to 10^{-3}. 0.1 mL diluted solution was spread on tryptic soy agar (TSA), potato–dextrose–agar (PDA) plates medium (Merck), and incubated in 8 days to obtain pure cultures. Then species of bacteria and fungi are sent to Bio-Techem Co., Ltd. to determine identify species.

2.2. Assay of *in vitro* anti-microorganism activities of SiO_2 nanoparticle and LMW chitosan

Chitosan with LMW of 16, 44.5, 80, and 109 kDa, degree of deacetylation > 80% and nano SiO_2 with the size of 20-30 nm were purchased from the Center for Radiation Technology Research and Development Ho Chi Minh. 1% (m/v) each LMW chitosan sample was dissolved in 1% acetic acid (v/v) and then adjusted to 5.5 pH with continuous stirring to obtain a homogeneous chitosan solution. The solution was then added with 0.04% (m/v) nano SiO_2, adjusted to 5.5 pH to obtain a uniform mixture. The mixture should be used within 1 hour to avoid nano SiO_2 precipitation.The experiment was designed to randomized complete containing 10 treatments with triplicate (5 Petri dishes per replication) follow by chitosan LMW 16; 44.5; 80 and 109 kDa (1%); 0.04% nano SiO_2 alone and in the combination of each chitosan LMW 16; 44.5; 80 and 109 kDa (1%), and the control. Antibacterial activity was assessed by disk diffusion method (Chand, 2013). Tested bacteria were prepared to spread on TSA media. Sterilized impregnated papers (6 mm in diameter) were used to spread on the plates. Then 10 µL of tested solution dispense into the middle of impregnated papers, sterilized distilled water used as the negative control. Plates were incubated at 32 ºC for 24 hours. Aftermath, the diameter antibacterial zones were measured and determined by following formula: $\Delta D = D - d$ (mm). Where D is the diameter of the antibacterial zone (mm), d is the diameter of sterile impregnated papers (mm).

Antifungal activity was assessed by agar-well diffusion method (Chand, 2013). PDA media containing tested solution was prepared and then perforated with 6 mm diameter into in the middle of the plates containing pure growth of fungi tested, sterilized distilled water used as the negative control. Plates were incubated at 32 ºC for 7 days to observe and measure the growth rate of fungi. Diameter of formulated fungi was determined by following formula: $\Delta A = A - a$ (mm). Where A is the diameter of formulated fungi (mm); a is the diameter perforated agar (mm).

2.3. FT-IR and SEM analysis

Guavas were dipped in prepared solution (1% 44.5 kDa chitosan and 0.04% nano SiO2) for 1 min with the guava ratio about 1:3, then taken out and drained well with a blower. The coated guava peel was collected with the thickness around 1–1.5 mm, which used for FT-IR and SEM analysis.

FT-IR spectra were recorded using a Perkin-Elmer MIR/NIR Frontier instrument (PerkinElmer, USA) in the region from 4000–400 cm^{-1}. Scanning Electron Microscopy (SEM) was performed on a FE-SEM S4800 (Hitachi, Japan).

2.4. Statistical analysis

All data were analyzed by JMP 10.0 software (SAS Institute Inc., Cary, NC, USA). Significant differences between treatments were showed through Duncan test ($p < 0.05$).

3. Results and discussion

3.1. Characterizations of Samples

Figure 1a shows FT-IR spectra of guava before and after coating the mixture of chitosan and nano SiO_2. As seen from the FT-IR spectrum of guava (black curve), a specific peak at 3368 cm^{-1} is related to the hydroxyl groups (O–H) stretching vibrations, whereas the peak at 1632 cm^{-1} is corresponded to C=O stretching vibration of amide groups (Chen et al., 2018). The vibrations of O–H in plane and out of plane blending are recorded at 1404 cm^{-1} and 818 cm^{-1}, respectively. Two vibrations at the wavelength of 1060 cm^{-1} and 896 cm^{-1} are consistent with –C–O–C pyranose ring and β-glycosidic linkages of cellulose (Chen et al., 2018). The presence of the peak at 629 cm^{-1} represents alkyl halides (Mandal et al., 2020). After coating the mixture of chitosan and nano SiO_2, there are shifts of peaks from 3368 cm^{-1}, 1404 cm^{-1}, 1060 cm^{-1}, and 620 cm^{-1} to 3436 cm^{-1}, 1444 cm^{-1}, 1034 cm^{-1}, and 638 cm^{-1}, respectively. These can be explained that chitosan and SiO_2 interact with guava at functional groups as – OH, –C–O–C pyranose ring and alkyl halides. In addition, the intensities of all peaks are decreased. Furthermore, observing a SEM image of guava peel surface after coating the mixture of chitosan and SiO_2 as presented in Figure 1b shows the uniform dispersion of SiO_2 particles. From these, it can conclude that a film of the mixture containing chitosan and nano SiO_2 is formed and covered guava peel surface.

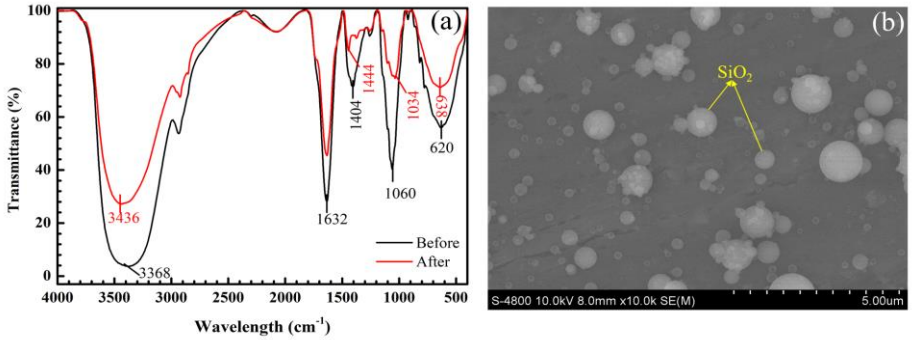

Figure 1. FT-IR spectra of guava peel before and after coating the mixture of chitosan (1%) and SiO_2 nanosilica (0.04%) (a) and SEM image of guava peel surface after coating the mixture of chitosan and SiO_2 (b)

3.2. Isolation of bacterial and fungal causing damage postharvest guava

We isolated Taiwan guava after damage and determined four fungal species, namely *Chrysosporium tropicum* (*Ch. tropicum*), *Cladosporium sphaerospermum* (*C. sphaerospermum*), *Aspergillus wentii* (*A. wentii*), *Colletotrichum acutatum* (*C. acutatum*) and three bacterial species, namely *Azotobacter* sp., *Escherichia coli (E. coli)*, *Bacillus subtilis* (*B. subtilis*) which caused damage postharvest guava.

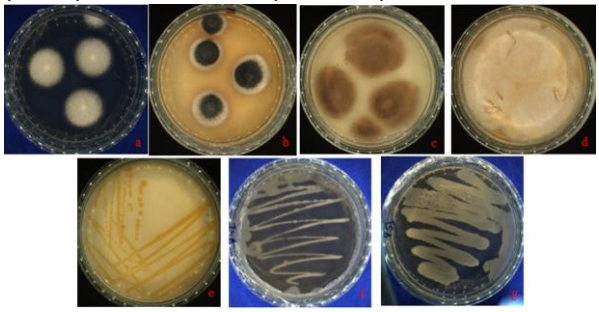

Figure 2. Properties of fungal and bacterial species isolated on damage guava fruits: a - Ch. tropicum; b - C. sphaerospermum; c - A. wentii; d - C. acutatum; e - Azotobacter sp.; f - E. coli; g - B. subtilis.

Characteristic of fungal species was then described on PDA media, with regard to *Ch. tropicum* fungi has in form of fibril, flaked, slow growth, while color hyphae (Figure 2a). *C. sphaerospermum* has in form of the fibril, grew dense, slow growth, on the top face of the fungi having deeper green color, under face having russet color, having wrinkle not smooth (Figure 2b). *A. wentii* has in form of fibril, on the top face and bottom face of the fungi light brown-yellow color (Figure 2c), *C. acutatum* has in form of fibril, on the top face and bottom face of the fungi was while color (Figure 2d) Characteristic of bacterial species was then described on TSA media: *Azotobacter* sp. colony has in form of circle, pale yellow, polished Viscid (Figure 2e). *E. coli* colony has in form

of circle, white opaque color, a polished and wetting surface on TSA media (Figure 2f). *B. subtilis* colony has in form of circle, the edge of the crenated irregular pale yellow-brown color surface and lined (Figure 2g).

3.3. Testing of antibacterial activity of nano SiO2 and LMW chitosan mixture in vitro

The outcome of the antibacterial ability of the LMW chitosan alone or in the combination of and nano SiO_2 on *E. coli*, *Azotobacter* sp., *B. subtilis* are shown in Table 1 and Figure 3. All the treatments applied with nano SiO_2 and LMW chitosan mixture result in the potential antibacteria of *E. coli* and *Azotobacter* sp., *B. subtilis*, and shown a significant difference to the control treatment ($p < 0.05$). nano SiO_2 and 44.5 kDa chitosan mixture exhibited the best effect with diameters of 12.86 ± 0.55 mm; 11.19 ± 0.56 mm; 7.86 ± 0.05 mm, respectively (Table 1, entry 6).

On the plates of *E. coli*, Entry 6 shown a significant difference with Entry 4 which treated with nano SiO_2 and 16 kDa chitosan mixture (11.61 ± 1.09 mm). On the plates of *Azotobacter* sp., Entry 4 containing nano SiO_2 and 16 kDa chitosan mixture (9.81 ± 0.24 mm) after standing Entry 6. On the plates *B. subtilis*, Entry *6* was not significant difference than Entry 5 containing 44.5 kDa chitosan mixture (7.14 ± 0.18 mm).

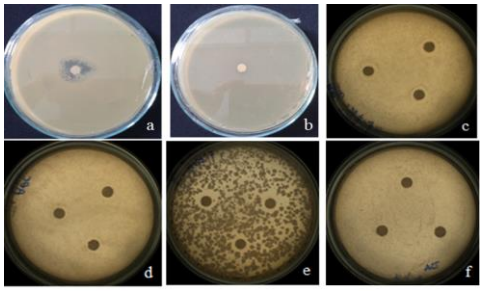

Figure 3. Inhibition ability of 44.5 kDa chitosan and 0.04% nano SiO_2 mixture against E. coli (a: tested sample; b: control sample), Azotobacter sp. (c: tested sample; d: control sample), B. subtilis (e: tested sample; f: control sample) in TSA medium after 24hrs at 32 ºC.

The results showed antibacterial ability of *B. subtilis* (the positive Gram bacteria) lower than *E. coli and Azotobacter* sp. (the negative Gram bacteria), and these are in agreement with the study of Dutta *et al*. (2009), mentioned that antibacterial ability of negative Gram bacteria is higher than positive Gram bacteria due to its higher hydrophilicity and negative charge ability on the cell surface of Gram-negative bacteria higher, results in more chitosan adsorbed (Abdullah, 2019). Meanwhile, according to Zhong *et al*. (2008) reported that Gram-positive bacteria were more susceptible due to barrier ability of outer membrane of the Gram-negative bacteria. All the treatments containing nano SiO_2 and LMW chitosan mixture showed better antibacterial activity than the one with LMW chitosan alone with identical molecular weight which can be explained by an interaction of nano-silicon dioxide and chitosan to enhance the anti-microorganism ability through improve the properties of films and inhibit disease on agriculture product (Dhanasingh et al., 2011). Due to highly porous structure of chitosan and nano silica compounds via formation of the Si–O–C bonds and hydrogen (N:H) bonds that silica was uniform dispersed and diffused in matrix of chitosan (Dhanasingh et al., 2011) and lead to tightly compacted making improve the mechanical and biological properties of chitosan material.

Table 1. Antibacterial activity of Nano SiO_2 (0.04%)–Si and LMW Chitosan (1%)–CH in vitro

Entry	Compounds	Antibacterial zone diameter (mm)		
		E. coli	*B. subtilis*	*Azotobacter* sp.
1	109 kDa CH	3.45 ± 0.49^{f}	-	4.27 ± 0.23^{f}
2	109 kDa CH, Si	4.80 ± 0.28^{ef}	1.24 ± 0.12^{e}	5.56 ± 0.47^{e}
3	16 kDa CH	9.85 ± 0.36^{c}	5.07 ± 0.17^{c}	7.82 ± 0.36^{c}
4	16 kDa CH, Si	11.61 ± 1.09^{ab}	5.86 ± 0.16^{b}	9.81 ± 0.24^{b}
5	44.5 kDa CH	10.63 ± 0.53^{bc}	7.14 ± 0.18^{a}	9.39 ± 0.49^{b}
6	44.5 kDa CH, Si	12.86 ± 0.55^{a}	7.16 ± 0.23^{a}	11.19 ± 0.56^{a}
7	80 kDa CH	5.23 ± 0.43^{d}	3.65 ± 0.43^{d}	6.31 ± 0.17^{de}
8	80 kDa CH, Si	6.34 ± 0.50^{de}	3.97 ± 0.54^{d}	6.82 ± 0.22^{cd}
9	Control	-	-	-
10	Si	-	-	-

In the same columns followed by the same letter are not significantly different at confidence interval 95%; -: not inhibited.

3.4. Testing of antifungal activity causing damage postharvest guava of nano SiO2 and Low molecular weight chitosan mixture in vitro

The results of antifungal ability of samples with only LMW chitosan or in combination with nano SiO_2 on *Ch. tropicum*, *C. sphaerospermum*, *A. wentii, C. acutatum* are shown in Table 2 and Figure 4. All treatments which contain nano SiO_2 and LMW chitosan mixture showed a potential antifungal activity of *Ch. tropicum, C. sphaerospermum, A. wentii, C. acutatum* and the results are significantly distinguished to the control treatment ($p < 0,05$). Therein, the treatment with 0.04% nano SiO_2 and 44.5 kDa chitosan mixture also showed the best antifungal ability on *Ch. tropicum, C. sphaerospermum, C. acutatum* with lowest growth fungi diameter of 6.32 ± 0.27 mm, 0.00 mm, 0.00 mm, respectively. However, with regard to *A. wentii* the plates treated with nano SiO_2 and 44.5 kDa chitosan mixture have diameter growth of fungi (9.34 ± 0.69 mm) higher negligible than that with 44.5 kDa chitosan (6.65 ± 0.35 mm). These results are corresponding to the previous studies which mentioned that LMW chitosan can be inhibited anthracnose disease caused by *Colletotrichum* sp (Xiangchun et al., 2012).

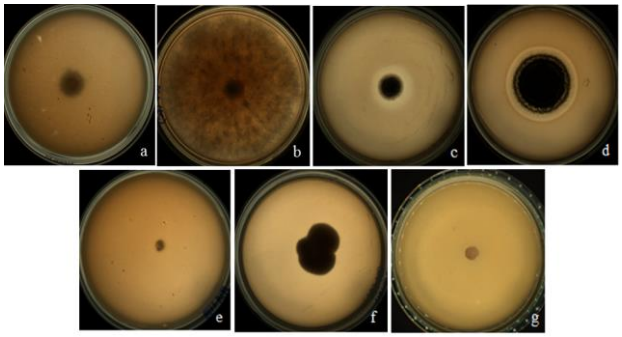

Figure 4. Inhibition ability of 44.5 kDa chitosan and 0.04% nano SiO_2 mixture in PDA medium after 5 days at 32 ºC against Ch. tropicum (a: tested sample; b: control sample), A. wentii (c: tested sample; d: control sample), after 7 days at 32 ºC against C. sphaerospermum (e: tested sample; f: control sample), C. acutatum (g: tested sample; h: control sample).

Most of the treatment with nano SiO_2 and LMW chitosan mixture showed better antifungal ability than those with chitosan alone at identical molecule weight due to improve properties of films that explained similar to as antibacterial activity. Chitosan has been demonstrated ability against several fungi. Chitosan's antifungal mechanism has been explained from previous studies. On cell surfaces of fungi, polycationic polysaccharides interaction with anionic sites leads to altering membrane permeability and internal osmotic imbalance or inhibit mRNA and protein synthesis (Zhang et al., 2011), diffuse inside hyphae inhibit on the enzymes activity leads to inhibit the fungus growth and act more quickly than on bacteria. Different penetration of chitosan into the fungal cell are also reported, the LMW chitosan (50 kDa) penetrated very easy into cell of *Fulvia fulva*, LMW chitosan 499 kDa penetrated the inner hyphae of *Fulvia fulva* but 1320 kDa was not.

Table 2. Antifungal activity of Nano SiO_2 (0.04%)–Si and LMW Chitosan (1%)–CH in vitro

Entry	Compounds	Diameter of growing fungi (mm)			
		Ch. tropicum	*C. Sphaerospermum*	*A. wentii*	*C. acutatum*
1	109 kDa CH	16.76 ± 0.79^{d}	6.88 ± 0.58^{b}	14.64 ± 0.39^{ef}	14.31 ± 1.04^{e}
2	109 kDa CH, Si	14.47 ± 0.87^{c}	+	13.57 ± 0.43^{de}	13.05 ± 0.27^{de}
3	16 kDa CH	12.56 ± 0.70^{b}	+	10.93 ± 0.82^{bc}	8.22 ± 0.68^{c}
4	16 kDa CH, Si	7.24 ± 0.79^{a}	+	12.09 ± 0.57^{cd}	5.42 ± 1.05^{b}
5	44.5 kDa CH	8.04 ± 0.61^{a}	+	6.65 ± 0.35^{a}	+
6	44.5 kDa CH, Si	6.32 ± 0.27^{a}	+	9.34 ± 0.69^{b}	+
7	80 kDa CH	16.05 ± 0.71^{cd}	+	16.57 ± 0.39^{fg}	$11,76 \pm 0,55^{d}$
8	80 kDa CH, Si	14.56 ± 0.46^{c}	+	18.36 ± 1.05^{g}	$11,18 \pm 0,54^{d}$
9	Control	80.00 ± 0.00^{e}	29.17 ± 1.76^{c}	29.87 ± 1.45^{h}	18.44 ± 1.18^{f}
10	Si	80.00 ± 0.00^{e}	29.30 ± 1.27^{c}	30.28 ± 0.95^{h}	18.13 ± 0.83^{f}

In the same columns followed by the same letter are not significantly different at confidence interval 95%; +: completely inhibited.

4. Conclusions

The study results in an effect of LMW chitosan in combination with nano SiO_2 on anti-microorganisms efficiency that mainly cause decay of postharvest guava fruit. Four fungal species, namely *Chrysosporium tropicum*, *Cladosporium sphaerospermum*, *Aspergillus wentii*, *Colletotrichum acutatum* and three bacterial species, namely *Azotobacter* sp., *Escherichia coli*, *Bacillus subtilis* was isolated from damage of guava fruit. In addition, chitosan with LMW of 44.5 kDa (1%) in combination with 0.04% nano SiO_2 shown the best capability to inhibit activity of four fungi species, i.e. *Ch. tropicum, C. sphaerospermum, A. wentii, C. acutatum*, and three bacteria species, i.e. *Azotobacter* sp., *E. coli*, *B. subtilis* on postharvest guava. It is recommended to utilize this mixture for further studies.

References

Abdullah, A.A., 2019, Preparation, Characterization and Antibacterial Effect of Chitosan Nanoparticles against Food Spoilage Bacteria, Journal of Pure and Applied Microbiology 13 (2), 1273–1278.

Aider, M., 2010, Chitosan application for active bio-based films production and potential in the food industry: Review, LWT - Food Science and Technology 43, 837–842.

Chand, B., 2013, Antibacterial effect of garlic (Alliumsativum) and ginger (Zingiber officinale) agains staphylococcus aureus, salmonella typhi, escherichia coli and bacillus cereus, Journal of Microbiology, Biotechnology and Food Sciences 2, 2481–2491.

Chao, D., Xin, M., Jingru, M., Md, . I.H.K., Lei, D., Avik, K., Xingye, A., Junhua, Z., Tanzina, H., Yonghao, N., 2019, Chitosan as A Preservative for Fruits and Vegetables: A Review on Chemistry and Antimicrobial Properties, Journal of Bioresources and Bioproducts 4, 11–21.

Chen, W., He, H., Zhu, H., Cheng, M., Li, Y., Wang, S., 2018, Thermo-Responsive Cellulose-Based Material with Switchable Wettability for Controllable Oil/Water Separation, Polymers 10, 592.

Dhanasingh, S., Mallesha, J., Hiriyannaiah, J., 2011, Preparation, characterization and antimicrobial studies of chitosan/silica hybrid polymer, Biointerface Res Appl Chem 1, 048–056.

Dutta, P.K., Tripathi, S., Mehrotra, G.K., Dutta, J., 2009, Perspectives for chitosan based antimicrobial films in food applications, Food Chemistry 114, 1173–1182.

Hong, K., Xie, J., Zhang, L., Sun, D., Gong, D., 2012, Effects of chitosan coating on postharvest life and quality of guava (Psidium guajava L.) fruit during cold storage, Scientia Horticulturae 144, 172–178.

Kulikov, S.N., Lisovskaya, S.A., Zelenikhin, P.V., Bezrodnykh, E.A., Shakirova, D.R., Blagodatskikh, I.V., Tikhonov, V.E., 2014, Antifungal activity of oligochitosans (short chain chitosans) against some Candida species and clinical isolates of Candida albicans: Molecular weight–activity relationship, European journal of medicinal chemistry 74, 169–178.

Mandal, A., Mukhopadhyay, P., Das, S.K., 2020, Adsorptive removal of phenol from wastewater using guava tree bark, Environmental Science and Pollution Research 27, 23937–23949.

Nadia, M.-C., Eric, L., Giangiacomo, T., Grégorio, C., 2019, Applications of chitosan in food, pharmaceuticals, medicine, cosmetics, agriculture, textiles, pulp and paper, biotechnology, and environmental chemistry, Environmental Chemistry Letters 17, 1667–1692.

Silva, G.S., Oliveira, P.C., Giordani, D.S., Castro, H.F.D., 2011, Chitosan/siloxane hybrid polymer: synthesis, characterization and performance as a support for immobilizing enzyme, Journal of the Brazilian Chemical Society 22, 1407–1417.

Sun, T., Wu, C.-l., Hao, H., Dai, Y., Li, J.-r., 2016, Preparation and preservation properties of the chitosan coatings modified with the in situ synthesized nano SiOx, Food Hydrocolloids 54, 130–138.

Witoon, T., Chareonpanich, M., Limtrakul, J., 2009, Effect of acidity on the formation of silica–chitosan hybrid materials and thermal conductive property, Journal of sol-gel science technology 51, 146–152.

Xiangchun, M., Yanxia, T., Aiyu, Z., Xuemei, H., Zhaoqi, Z., 2012, Effect of oligochitosan on development of Colletotrichum musae in vitro and in situ and its role in protection of banana fruits, Fruits 67, 147–155.

Yu, Y., Zhang, S., Ren, Y., Li, H., Zhang, X., Di, J., 2012, Jujube preservation using chitosan film with nano-silicon dioxide, Journal of Food Engineering 113, 408-414.

Zhang, H., Li, R., Liu, W., 2011, Effects of chitin and its derivative chitosan on postharvest decay of fruits: A review, Int J Mol Sci 12, 917–934.

Zhong, Z., Xing, R., Liu, S., Wang, L., Cai, S., Li, P., 2008, Synthesis of acyl thiourea derivatives of chitosan and their antimicrobial activities in vitro, Carbohydrate Research 343, 566–570.