ISSN 0101-2061 (Print)
ISSN 1678-457X (Online)
Food Science and Technology

DOI: https://doi.org/10.1590/fst.19919

Antibacterial activity of polyvinyl alcohol (PVA)/ε-polylysine packaging films and the effect on longan fruit

Yana LI[1*], Yuwen WANG[1], Juying LI[1]

Abstract

Biodegradable and antimicrobial films via combing ε-polylysine (ε-PL) and polyvinyl alcohol (PVA) is a promising material for food packaging. We prepared the ε-PL/ PVA films with the ε-PL content of 0%, 1%, 3%, 5% and 7% (wt., relative to PVA) by casting method and their physical and chemical properties were characterized. SEM indicated the surface of ε-PL/PVA films was smooth with well compatibility between ε-PL and PVA at the ε-PL addition content of 1%. The tensile strength and barrier properties for water vapor and oxygen of the ε-PL/PVA films initially elevated and then decreased when ε-PL content exceeded 1%. The ε-PL doped endows antibacterial activity of PVA and the antibacterial property of ε-PL/PVA films was increasing with increase of ε-PL content. Compared with the control, the ε-PL/PVA packaging films could inhibit effectively the pericarp browning and pulp breakdown for longan, consequently the rate of commercially acceptable fruit packaged by ε-PL/PVA films was improved dramatically. However, the effect of ε-PL/PVA on weight loss rate for longan was worse than the control.

Keywords: polyvinyl alcohol; ε-polylysine; antimicrobial films; food packaging; longan.

Practical Application: Prolonging the shelf life of longan fruit using a kind of packaging films characterized by environmental friendly and excellent antibacterial activity.

1 Introduction

Plastic films based on polymer are widely used for food packaging due to the favorable physiochemical properties and low cost, whereas most of polymeric materials would lead to a number of environmental problems on account of their nondegradability. Recently environmentally friendly natural and sustainable biopolymers give rise to more and more attentions and is promising to replace the traditional polymeric plastic (Guo et al., 2014), in which, poly(vinyl alcohol) (PVA) as one of biodegradable polymers, are brought into focus due to the outstanding biocompatibility, film formation, and gas barrier performance(Wang et al., 2018). However, being short of bactericidal properties of PVA limits the wider application in food packaging.

ε-polylysine (ε-PL) (Li et al., 2018) is a biodegradable and water-soluble bacteriostatic agent with a wide range of antimicrobial activity and food safety. It has been approved as food preservative by FDA. It is found that the addition of ε-PL to polymer matrix can improve the antibacterial activity of chitosan (Li et al., 2018; Lin et al. 2018a) or alginate (Liu et al., 2018). To the best of our knowledge, it has no reports to date about ε-PL in combination with PVA.

Longan (*Dimocarpus longan* Lour.) is one of the consumed subtropical fruit with rich nutritional and medicinal value in southern China, also in many other countries in the world. However, it is known that the shelf life of longan fruit is short due to rapid microbial decay and pericarp browning caused by high sugar and moisture content (Suwanamornlert et al., 2018). Previous studies using fumigation, dipping and coating with antifungal agents, such as ClO_2–SO_2 (Wu et al., 2011), hydrogen peroxide (Lin et al., 2017), propyl gallate (Lin et al., 2018b), essential oil (Suwanamornlert et al., 2018) or chitosan (Shi et al., 2013) could delay longan fruit rot and pericarp browning. Nevertheless, the research of packaging films applied in longan fruit is rare.

The aim of this study was to develop an active flexible films based on PVA that combines ε-PL with food safety and environmental friendliness, simultaneously to find alternatives instead of conventional bactericide to extending the shelf life of longan fruit via antibacterial packaging films such as ε-PL/PVA. The antibacterial activity and physiochemical properties, as well as the effect on longan fruit of ε-PL/PVA films were also investigated.

2 Materials and methods

2.1 ε-PL/PVA fabrication

The ε-PL/PVA solutions with 0%, 1%, 3%, 5% and 7% (wt., relative to PVA) content of ε-PL prepared by adding a certain amount of ε-PL (Baina Biological Engineering Co., China) into PVA aqueous solutions (2% w/v, ShangPu Chemical Co., China) with magnetic stirring for 4 h and an ultrasonator (KQ-300DV, Kunshan, China) for 30 min were poured into a silicone mold (280 mm × 280 mm) and dried in oven at 40 °C for 24 h. Finally the ε-PL/CS films with thickness of about 60 ± 2 um peeled off from the mold were fabricated.

Received 30 Aug., 2019
Accepted 29 Oct., 2019
[1] *Mechanical Engineering, Wuhan Polytechnic University, Wuhan, China*
*Corresponding author: myllyn@126.com

2.2 Characterization of ε-PL/PVA films

Morphology

The sample films of ε-PL/PVA were fractured in liquid nitrogen and the morphology was probed through scanning electron microscope (SEM, S-4800, Hitachi Inc., Japan).

Tensile strength

The tensile strength of ε-PL/PVA films (130 mm × 20 mm) was measured via a testing machine (SANS JTM4104, Shenzhen, China) in a speed of 50 mm/min at 23 °C and 60%RH. Six replicates were tested for each sample.

Barrier properties

Barrier properties were characterized by the water vapor permeability rate (*WVT*) and oxygen permeability rate (*OT*) determined respectively with a moisture permeable meter (W-E-11A, Labthink Inc., China) and a gas permeability testing instrument (OX2/230, Labthink Inc., China) at 23 °C and 60%RH.

Antibacterial activity

The antibacterial activity against *Escherichia coli* (*E. coli*) and *Staphylococcus aureus* (*S. aureus*) was characterized by optical density (OD) according to (Li et al., 2018). The OD value was measured by Bioscreen C system via an automatic growth curve analyzer (FP-1100-C, Oy Growth Curves Ab Co., Finland) at 600 nm and 37 °C for 48 h.

2.3 Effect of ε-PL/PVA films on longan fruit

Sample treatment and packaging

The longan fruit used in this test was provided by an orchard in Guangdong Province, China. Fruit with uniform size and color and without any physical damages was picked out for this measurement.

Longan were divided into five treatment groups, which were packed respectively with ε-PL/PVA bags (140 mm×140 mm) prepared by ε-PL/PVA films with 0%, 1%, 3%, 5% and 7% concentration of ε-PL. The fruit packed by PE bags with the thickness of 12μm branded by Miaojie from supermarket was as control. The *WVT* and *OT* of the PE films are shown in Table 1. There are ten fruit in each bag. All samples were stored in a warehouse at 10 °C with 60%RH.

Fruit quality determination

Weight loss (%) of longan fruit was measured according to (Li et al., 2018).

Pericarp browning index was calculated by the equation: Σ(browning scale × percentage of corresponding fruit within each class), in which, browning scale is defined as 1, 2, 3, 4 and 5, respectively for no browning, 25%, 25-50%, 50-75%, and > 75% of brown area/total surface on each fruit.

Pulp breakdown index was subjectively assessed using an expert panel and calculated by the equation: Σ(pulp breakdown scale × percentage of fruit within each class), in which, pulp breakdown scale is defined as 0, 1, 2, and 3 for none, slight, moderate and severe in the light of firmness.

Rate of commercially acceptable fruit was measured by counting fruit with a percentage of pericarp browning index<3 as well as no evident mildew on the surface.

2.4 Statistical analysis

For determination of weight loss, pericarp browning and pulp breakdown, there were three replicates per treatment and ten fruit (one bag) per replicate for each time point. For rate of commercially acceptable fruit analysis, three replications were for each time point with 50 fruits per replicate. All the data were subjected to analysis of variance (ANOVA), and comparisons of means were carried out by Duncan's multiple range test, considering differences significant when $P < 0.05$.

3 Results and discussion

3.1 Morphology

The surface morphology of ε-PL/PVA films are shown in Figure 1. It is found that the surface of ε-PL/PVA films with the ε-PL concentration of 1% (Figure 1b) is smooth, the same with PVA surface shown in Figure 1a, and no significant phase separation appears, which indicates well compatibility between ε-PL and PVA matrix at a low doped content of ε-PL. When the addition content of ε-PL was improved to 5%, a large amount of white point or mass is observed on the surface of film as shown in Figure 1c. That suggests the heavy aggregation of ε-PL occurred.

3.2 Tensile strength

Table 1 shows the tensile strength of the ε-PL/PVA films as a function of ε-PL content. As can be seen from Table 1, the tensile strength initially increased due to well dispersibility of ε-PL in PVA and then decreased dramatically to be lower than

Table 1. Tensile strength and barrier properties of ε-PL/PVA films.

ε-PL (wt.%)	Tensile strength/MPa	*WVT* (g/m^2.d)	*OT* (mL/m^2.d)
0	35.7 ± 1^a	845.6 ± 20.2^a	2.4 ± 0.2^a
1	43.3 ± 2^b	787.0 ± 15.1^b	1.5 ± 0.1^b
3	28.1 ± 1.5^c	842.0 ± 10.2^a	3.5 ± 0.2^c
5	23.1 ± 1.8^d	854.5 ± 18.3^a	6.6 ± 0.3^d
7	18.1 ± 1.3^e	872.2 ± 20.3^a	6.8 ± 0.2^d
PE	-	48.3 ± 5.2^c	145 ± 10.8^e

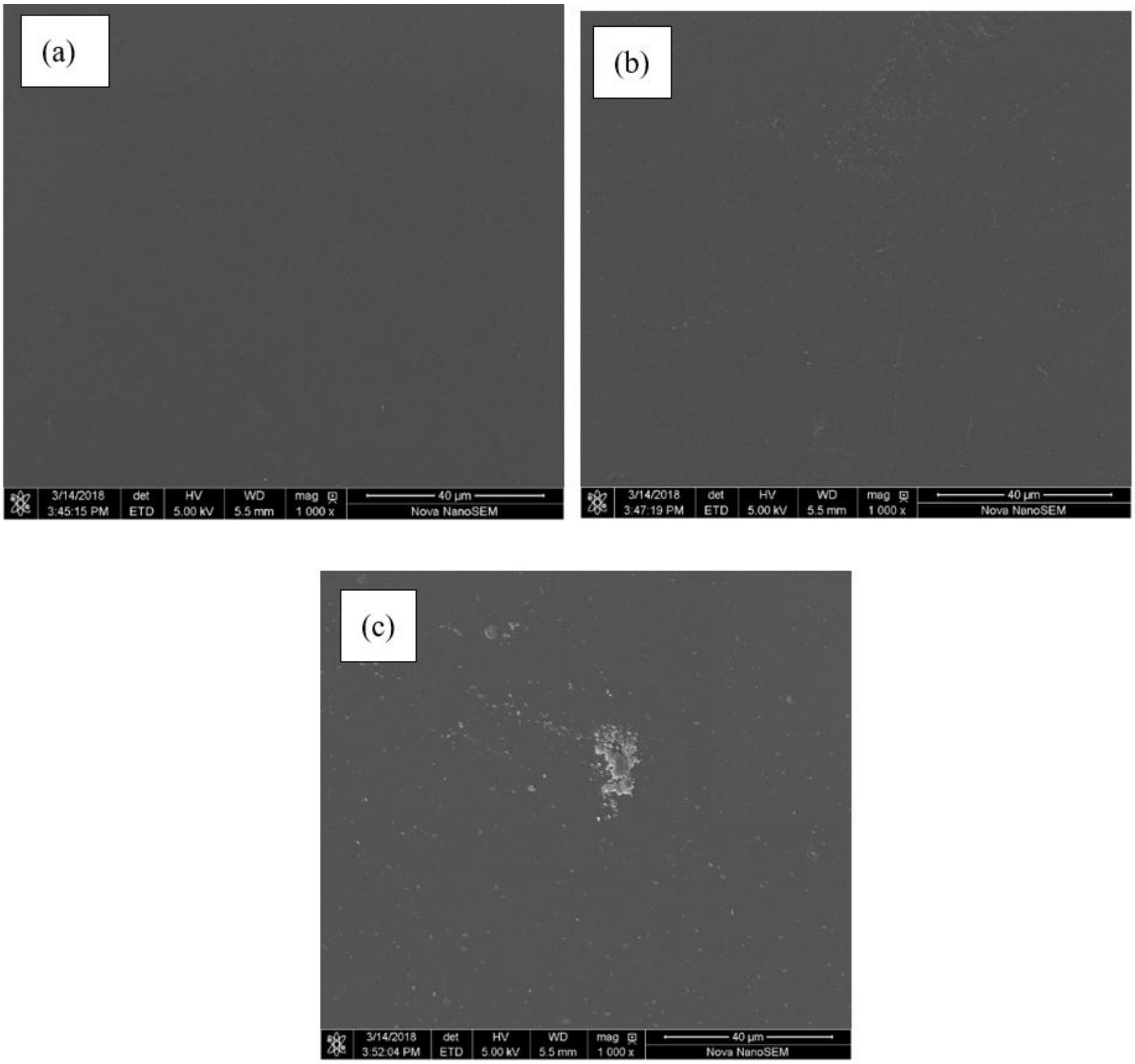

Figure 1. SEM of the surface for PVA film (a) and ε-PL/PVA films with the ε-PL content of 1% (b) and 5% (c).

that of the neat PVA film (0%), which may be due to more defects created by heavier aggregation of ε-PL in PVA matrix when the ε-PL content was greater than 1% as seen from SEM.

The values are presented as mean ± standard deviation. Any two means in the same column followed by the same superscript (a, b, c) are not significantly ($P > 0.05$) different by Duncan's multiple range tests.

3.3 Barrier properties

The barrier properties of ε-PL/PVA composite films to water vapor and oxygen is shown in Table 1. It is found that the decline of *WVT* and *OT* of the PVA films occurs after the addition of ε-PL to PVA matrix, then they are increasing when the ε-PL content is exceed 1%, which indicates the improvement of the barrier properties of ε-PL/PVA films with low doped content of ε-PL. It is worth mentioning that it is characteristic of the low barrier against moisture and high barrier property against oxygen for ε-PL/PVA films contrast with the PE films.

3.4 Antibacterial activity

Figure 2 shows the OD value of ε-PL/PVA films against *E. coli* and *S. aureus* as a function of incubation time. It is found that with the increase of ε-PL content, the OD value of ε-PL/PVA films is deceased. That indicates the antibacterial activity of ε-PL/PVA films is enhanced as increase of the ε-PL amount since higher antibacterial property of films brings out lower OD value (Li et al., 2018). It is also observed that when the ε-PL content is higher than 3%, the OD value of ε-PL/PVA films tends to equilibrate within an incubation time of 15 h for *E. coli* and 10h for *S. aureus*. The ε-PL/PVA films with the ε-PL concentration of 7% has the strongest antibacterial activity against *E. coli* and *S. aureus*.

3.5 Effect of ε-PL/PVA films on longan fruit

Weight loss

The different package on weight loss of longan fruit is shown in Figure 3. As can be seen from Figure 3, the weight loss percentage in all experimental fruit increased with extended

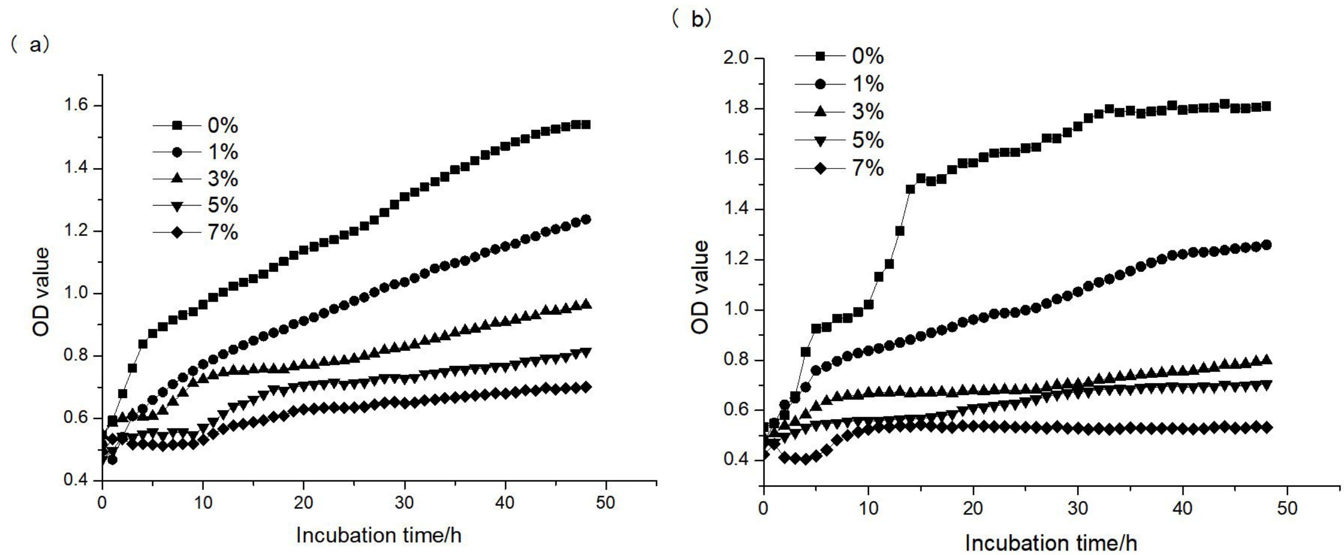

Figure 2. OD value of ε-PL/PVA films against *E. coli* (a) and *S. aureus* (b).

storage time due to transpiration and desiccation. Compared with the control, the longan fruit packed by PVA or ε-PL/PVA bags showed a higher increasing weight loss (P<0.05). It could be explained that the barrier property of ε-PL/PVA films against water vapor is worse than PE films resulted from the high water absorption caused by the hydroxyl groups on PVA (Zhang et al., 2018). That is consistent with the results of barrier properties (Table 1) with the WVT of 845.5 ± 20.2 g/m^2.d for PVA and 48.3 ± 5.2 g/m^2.d for PE films, respectively. However, the concentration of ε-PL on the weight loss of longan fruit has no significant (P > 0.05).

Pericarp browning index

It is well-known the pericarp browning restricts transportation and makes longan loss market value. Recently, more and more data indicate the browning might be accounted for by the disorder of energy metabolism caused by oxidative stress and mitochondrial recession (Lin et al., 2018b; Sun et al., 2011).

The changes of pericarp browning index for longan fruit during storage are reported in Figure 4. From Figure 4, the pericarp browning index exhibited an increasing trend from 5d for the control, while for the other groups, the pericarp browning occurred at 15d then the pericarp browning index was elevated with the increase of storage time. Furthermore, the pericarp browning index of the fruit packaged by 0%-7% ε-PL/PVA films was significant lower (P < 0.05) than that of the control. It indicates the ε-PL/PVA films could effectively inhibit the browning of longan fruit compared with the PE films. The ε-PL content doped has no significant effect on pericarp browning index in contrast with the control (P > 0.05).

Pulp breakdown index

After harvest, the outer peel of longan fruit looks complete, but the texture of its pulp has become soft and the juice has flowed out, which is known as the pulp breakdown. The autolysis of pulp seriously affects the quality of longan fruit, accelerates

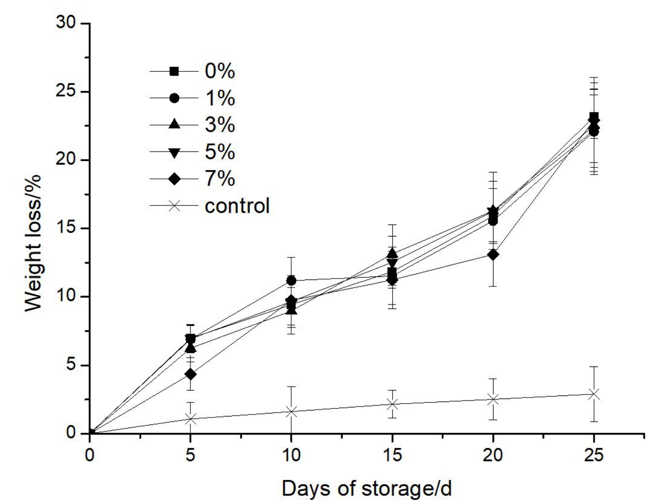

Figure 3. Changes of weight loss during storage.

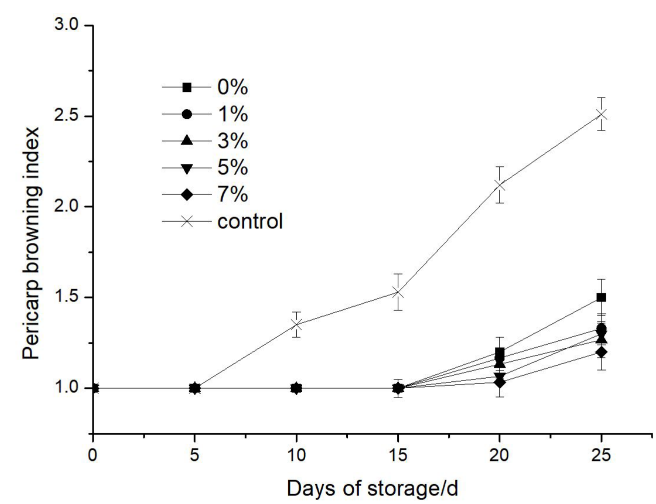

Figure 4. Changes of pericarp browning index during storage.

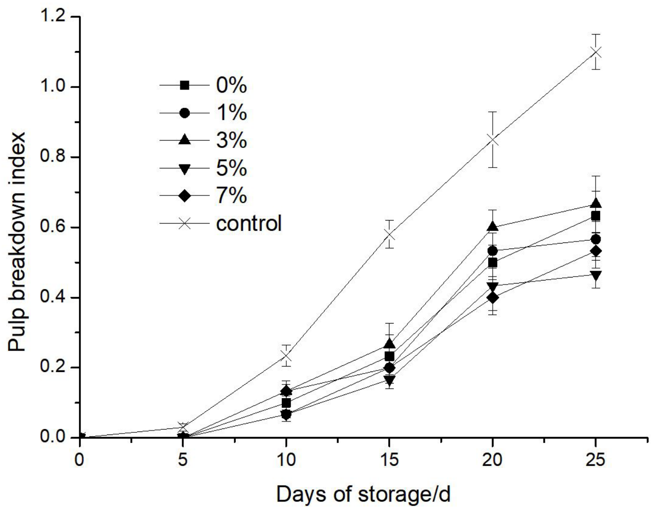

Figure 5. Changes of pulp breakdown index during storage.

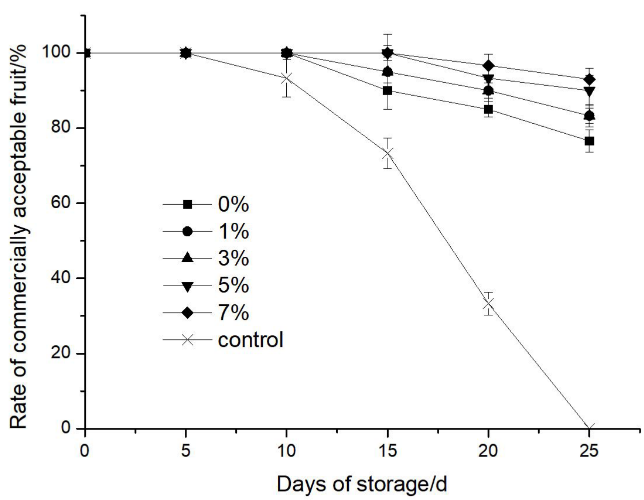

Figure 6. Changes of rate of commercially acceptable fruit during storage.

the aging process of fruit, resulting to shorten the shelf life of longan fruit (Li et al., 2015).

Figure 5 shows the changes of pulp breakdown index during storage. It is found that for the control, the pulp breakdown index slowly increased initially and then increased sharply after day 5, while the pulp breakdown index of longan fruit packaged by ε-PL/PVA was elevated slowly during the storage. Compared with the control, the significant decrease of pulp breakdown index of ε-PL/PVA group ($P < 0.05$) indicates the ε-PL/PVA bags markedly delayed the aging probably caused by preventing the respiration of fruit due to the barrier properties for O_2 of films.

Similar to the result of pericarp browning index, there is no significant difference of ε-PL concentration on pulp breakdown index ($P > 0.05$).

Rate of commercially acceptable fruit

Figure 6 shows the rate of commercially acceptable fruit during storage. It is found that the quality of longan fruit became worse and worse with prolonging the storage time. For the control, the rate of commercially acceptable fruit began to decrease at storage time of 5 d, then dropped rapidly to <50% at day 20. However, the sample fruit packaged by PVA or ε-PL/PVA showed a significantly better commercially quality ($P < 0.05$) that the mild decease of the rate of commercially acceptable fruit occurred at storage time of 10 d with >85% of the rate of commercially acceptable fruit at day 20. At the last storage time of 25 d, the rate of commercially acceptable fruit was above 80% of ε-PL/PVA groups in contrast with 0% of the control. It is also observed that from Figure 6, the rate of commercially acceptable fruit was improved when increasing the ε-PL doped content though there is no significant difference between the samples of 0%-7% ε-PL/PVA films ($P > 0.05$). That suggests ε-PL/PVA films are effective to inhibit the growth of bacteria on pericarp of longan fruit to make the decay rate decreased and the rate of commercially acceptable fruit enhanced in storage time, and bringing in better protecting against microbe with higher ε-PL addition content in accordance with the antibacterial results shown in Figure 2. As a result, the postharvest longan fruit packaged by 7% ε-PL/PVA films maintained a best fruit quality with 93% of the rate of commercially acceptable fruit.

4 Conclusions

The antibacterial ε-PL/PVA films with were prepared by the addition ε-PL to PVA matrix with the amount of 0%, 1%, 3%, 5% and 7%. SEM images shows that the surface of PVA and ε-PL/PVA films for 1% ε-PL addition were smooth. The tensile strength and barrier properties for water vapor and oxygen of the ε-PL/PVA films were enhanced with the ε-PL concentration being lower than 1% and then decreased when ε-PL was greater than 1%. The adding of ε-PL results in a significant antibacterial properties to PVA, and that the antibacterial activity of ε-PL/PVA films increases with the ε-PL increase. The results of the effect of films on longan fruit show that the ε-PL/PVA films is highlighted by restraining the pericarp browning and pulp breakdown effectively, which in turn results in better fruit quality and high rate of commercially acceptable fruit compared with the control of PE. The developed antibacterial ε-PL/PVA films is promising applied in food packaging.

Acknowledgements

This research was financially supported by Hubei Provincial Natural Science Foundation of China (2017CFB494), Science and Technology Major Project of Hubei Province (2017ABA136) and Research and Innovation Initiatives of WHPU (2019y18).

References

Guo, X. K., Yu, L., Chen, L. H., Zhang, H. Y., Peng, L. M., Guo, X. F., & Ding, W. P. (2014). Organoamine-assisted biomimetic synthesis of faceted hexagonal hydroxyapatite nanotubes with prominent stimulation activity for osteoblast proliferation. *Journal of Materials Chemistry*, 2(13), 1760-1763. http://dx.doi.org/10.1039/C3TB21652G.

Li, L., Li, J. M., Sun, J., Li, C. B., Sheng, J. F., Zheng, F. J., Liao, F., He, X. M., Liu, G. M., Ling, D. N., & You, X. G. (2015). Effects of 2-butanol on quality and physiological characteristics of longan fruit stored at ambient temperature. *Postharvest Biology and Technology*, 101(3), 96-102. http://dx.doi.org/10.1016/j.postharvbio.2014.12.002.

Li, Y. N., Ye, Q. Q., Hou, W. F., & Zhang, G. Q. (2018). Development of antibacterialε-polylysine/chitosan hybrid films and the effect on citrus. *International Journal of Biological Macromolecules*, 118(10), 2051-2056. http://dx.doi.org/10.1016/j.ijbiomac.2018.07.074. PMid:30026100.

Lin, L., Xue, L., Duraiarasan, S., & Haiying, C. (2018a). Preparation of ε-polylysine/chitosan nanofibers for food packaging against *Salmonella* on chicken. *Food Packaging and Shelf Life*, 17(9), 134-141. http://dx.doi.org/10.1016/j.fpsl.2018.06.013.

Lin, Y. F., Lin, Y. X., Lin, H. T., Chen, Y. H., Wang, H., & Shi, J. (2018b). Application of propyl gallate alleviates pericarp browning in harvested longan fruit by modulating metabolisms of respiration and energy. *Food Chemistry*, 240(2), 863-869. http://dx.doi.org/10.1016/j.foodchem.2017.07.118. PMid:28946353.

Lin, Y. F., Lin, Y. X., Lin, H. T., Ritenour, M. A., Shi, J., Zhang, S., Chen, Y. H., & Wang, H. (2017). Hydrogen peroxide-induced pericarp browning of harvested longan fruit in association with energy metabolism. *Food Chemistry*, 225(15), 31-36. http://dx.doi.org/10.1016/j.foodchem.2016.12.088. PMid:28193430.

Liu, J. H., Xiao, J., Li, F., Shi, Y., Li, D., & Huang, Q. (2018). Chitosan-sodium alginate nanoparticle as a delivery system for ε-polylysine: preparation, characterization and antimicrobial activity. *Food Control*, 91(9), 302-310. http://dx.doi.org/10.1016/j.foodcont.2018.04.020.

Shi, S. Y., Wang, W., Liu, L. Q., Wu, S. J., Wei, Y. Z., & Li, W. C. (2013). Effect of chitosan/nano-silica coating on the physicochemical characteristics of longan fruit under ambient temperature. *Journal of Food Engineering*, 118(1), 125-131. http://dx.doi.org/10.1016/j.jfoodeng.2013.03.029.

Sun, J., You, X., Li, L., Peng, H., Su, W., Li, C., He, Q., & Liao, F. (2011). Effects of a phospholipase D inhibitor on postharvest enzymatic browning and oxidative stress of litchi fruit. *Postharvest Biology and Technology*, 62(3), 288-294. http://dx.doi.org/10.1016/j.postharvbio.2011.07.001.

Suwanamornlert, P., Sangchote, S., Chinsirikul, W., Sane, A., & Chonhenchob, V. (2018). Antifungal activity of plant-derived compounds and their synergism against major postharvest pathogens of longan fruit in vitro. *International Journal of Food Microbiology*, 271, 8-14. http://dx.doi.org/10.1016/j.ijfoodmicro.2018.02.009. PMid:29459244.

Wang, Z. Y., Qiao, X. Y., & Sun, K. (2018). Rice straw cellulose nanofibrils reinforced poly(vinyl alcohol) composite films. *Carbohydrate Polymers*, 197, 442-450. http://dx.doi.org/10.1016/j.carbpol.2018.06.025. PMid:30007633.

Wu, B., Li, X., & Chen, W. (2011). Effects of bi-component ClO_2–SO_2 on the quality of harvested longan fruit. In *Proceedings of 2011 International Conference on Agricultural and Biosystems Engineering*. USA: Information Engineering Research Institute.

Zhang, J., Lei, W. W., Chen, J. Y., Liu, D., Tang, B., Li, J. L., & Wang, X. G. (2018). Enhancing the thermal and mechanical properties of polyvinyl alcohol (PVA) with boron nitride nanosheets and cellulose nanocrystals. *Polymer*, 148, 101-108. http://dx.doi.org/10.1016/j.polymer.2018.06.029.