Antibacterial Performance Evaluations for Silver Colloid Incorporated Thermoplastics

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Abstract: Anti-bacterial performance efficacies of various thermo-plastics, such as medium-density polyethylene (MDPE), polystyrene (PS), polyethylene terephthalate (PET) and polyvinyl chloride (PVC) containing nano-silver colloids were quantitatively assessed under a wide range of testing conditions. The effects of nano-silver colloid content and the silver-polymer contact time were of our main interests through a plate-count-agar (PCA) testing method using *Escherichia coli* as testing bacteria. Two different methods were used for incorporating the nano-silver colloids into the thermoplastics, these being spray-coating onto the thermoplastic surface, dry-blending in the thermoplastic matrices. The experimental results suggested that coating silver colloid onto all types of thermoplastic substrates could inhibit the growth of the *E. coli* bacteria up to 99%, different thermoplastics showing different optimum contents of nano-silver content to achieve the maximum bacterial inhibition. For a given nano-silver content, the spray-coating technique was much more effective than that dry-blending technique.

Introduction

In packaging applications, thermoplastics have been widely used and their microbial contaminations are recently of main concern. The application of antimicrobial agents into the polymer products is one of the methods to prevent the thermoplastic products from microbial contaminations **[1]**. Many antibacterial agents including nisin, nano-silver, triclosan and sorbic acid anhydride have been used in packaging thermoplastics for inhibition of the bacterial growth in various processing techniques **[2]**. Methods to incorporate the anti-bacterial agents into the polymer matrices could be done by either blending into the polymer matrices or coating onto the surface of the polymer products. The anti-bacterial efficacies are dependent on type, concentration and diffusability of the incorporated bacteria through the polymer matrices, as well as the testing methods used for such evaluations **[3-4]**. In this present paper, anti-bacterial performance efficacies of various thermo-plastics, such as medium-density polyethylene (MDPE), polystyrene (PS), polyethylene terephthalate (PET) and polyvinyl chloride (PVC) containing nano-silver colloids were quantitatively assessed under a wide range of testing conditions and the effects of nano-silver colloid content and the silver-polymer contact time.

Materials and Methods

Materials: Medium-density polyethylene (MDPE, M380RU/RUP, Thai Polyethylene Co., Ltd., BKK, Thailand), Polystyrene (PS, Styron 656D267; Siam Polystyrene Co., Ltd, BKK, Thailand), Polyethylene terephthalate (PET; Indorama Co., Ltd., BKK, Thailand), and polyvinyl chloride (PVC, VP Wood Co., Ltd., BKK, Thailand), were used as matrices. Nano-silver colloid (designated as SNSE, supplied by Koventure Co., Ltd, Bangkok, Thailand) was used as the anti-bacterial agent. *Escherichia coli* (ATCC 25922) was used as testing bacteria.

Incorporating method: The thermoplastics were incorporated with nano-silver colloid using two different techniques. For spray-coating, the polymeric substrates were pre-heated at 120^{o}C for 5 min before being spray-coated with nano-silver colloids using a constant spraying condition (spray distance of 10 cm, spraying temperature of 25^{o}C, spraying time of 10 sec and spray rate of 5 ml/min). The surface area covered by the nano-silver was 10x10 cm^2 for a required dosage. For dry-blending method, the thermoplastic powder were dry-blended with nano-silver colloid using a fluidized bed before being compression moulded to obtain the silver-incorporated thermoplastic film with 0.2mm thick. The resultant films from the two incorporating techniques were then cut into square samples of 2.5x5cm^2 for plate count agar (PCA) test.

Preparation of film specimens: The experimental procedure was commenced by compression moulding for making a film specimen of 0.2 mm thick. The mould pressure, temperature and time used in the compression moulding process were 150 kg.cm^{-3}, 160^{o}C and 5 min, respectively, for MDPE, 150 kg.cm^{-3}, 170^{o}C and 5 min, respectively, for PS, 50 kg.cm^{-3}, 255^{o}C and 1 min, respectively, for PET, and 180 kg.cm^{-3}, 180^{o}C and 2 min, respectively, for PVC.

Measurement of antibacterial performance: Plate Count Agar (PCA) was suitable for quantitative assessment of bacteria reduction, which follows the test standard of ASTM E-2149 (2001). 2 pieces of thermoplastic film sample of 2.5x5 cm^2 were used. Nutrient broth (NB) was used as a growing medium for *E. coli* bacteria and peptone solution (prepared by 1 g / L peptone, pH 6.8 – 7.2) was chosen as a testing medium. Bacteria were cultivated in 5 ml of NB at 37^oC for 24 h. The film samples were placed into a 250 ml flask with peptone solution and the bacteria cell suspensions were diluted with 0.85% sodium chloride solution to the required initial bacteria density. In this work, the dilution factor (n) used was dependent on the preferred number of initial bacteria colonies (ranging from 30 to 300 colonies). The flask was shaken on a reciprocal shaker at a speed of 100 rpm at $37^oC \pm 0.5^oC$ for a contact time of 30, 90, 150, and 210 minutes. 100 µL of bacterial suspension after shaking were placed over the agar into sterilized Petri dishes. The inoculated plates were cultivated at $37^oC \pm 0.5^oC$ for 24 h before calculating the viable count of the testing bacteria and evaluating the anti-bacterial efficacies using Equation 1 **[5]**.

$$R = \frac{A - B}{A} \times 100 \qquad \text{(Equation 1)}$$

where: *R* is the percent reduction of bacteria (%).
A is average number of bacterial colonies from thermoplastics without nano-silver colloid (CFU/ml)
B is average number of bacterial colonies from thermoplastics incorporated with nano-silver colloid (CFU/ml)

Results and Discussion

Neat thermoplastics

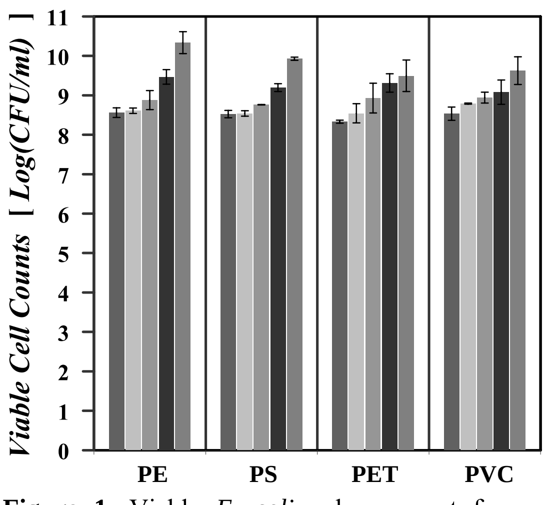

Figure 1. Viable *E. coli* colony count for various thermoplastics with different contact times 0 min (■), 30 min (■), 90 min (■), 150 min (■), and 210 min (■) without incorporation of nano-silver colloid

Figure 1 shows viable colony count for *Escherichia coli* for various thermoplastics under a wide range of contact times without incorporation of nano-silver colloid. It can be seen that all thermoplastics had similar viable count of *E. coli*, and the *E. coli* growth appeared to increase with increasing contact times. However, the increasing magnitude of *E. coli* growth with increasing contact times (differences in viable counts at 0 and 210min) for PET and PVC seemed to be smaller than that for other thermoplastics. The reason was related to polarity and halogen (chlorine atom) that had abilities to reduce *E. coli* population **[6].**

Nano-silver filled thermoplastics by spray-coating

Figure 2 shows the effects of nano-silver content and contact time on viable cell count and antibacterial efficacy for different thermoplastics. For all thermoplastics, it was observed that the viable cell count decreased with increasing the content of nano-silver colloid on the thermoplastic surfaces. This was obviously seen by the percentage reduction of *E. coli*. However, it was interesting to note that the optimum silver content to reach 99% reduction of *E. coli* for all thermoplastics were different. Overall, it was observed that the contact time of 150 min was sufficient for 99% reduction of *E. coli*. At 150 min contact time, 75 ppm nano-silver colloid was required for PE whereas 50 ppm nano-silver were required for PS, PET and PVC for achieving the maximum percentage reduction of *E. coli*. The differences in the silver dosages required for each thermoplastics were associated with a number of factors such as polarity of thethermoplastics used, embedding or diffusability of nano-silver colloid in the thermoplastic surfaces which were related with the softening temperatures of all thermoplastics, and possible formation of AgCl compound (in the case of PVC) during spray-coating.

Comparison of spray-coating and dry-blending techniques

Polyethylene (PE) was selected for studying the effect of incorporating technique on the anti-bacterial performance evaluation using nano-silver colloid. Figure 3 shows a comparison of viable cell count of PE sample having different dosages of nano-silver which were incorporated by spray-coating (a) and dry-blending (b) techniques. It was found that for a given nano-silver content, the spray-coating technique was much more effective than that dry-blending technique. This was simply explained that the silver concentration to contact with the *E. coli* by the spray-coating technique was greater. The nano-silver particles from the dry-blending were embedded and trapped in the PE matrix and they were not able to contact with the bacteria. This was why the concentration to reduce the viable bacteria in the dry-blending technique occurred at the silver content of 100 ppm whereas that in the spray-coating technique was 50 ppm.

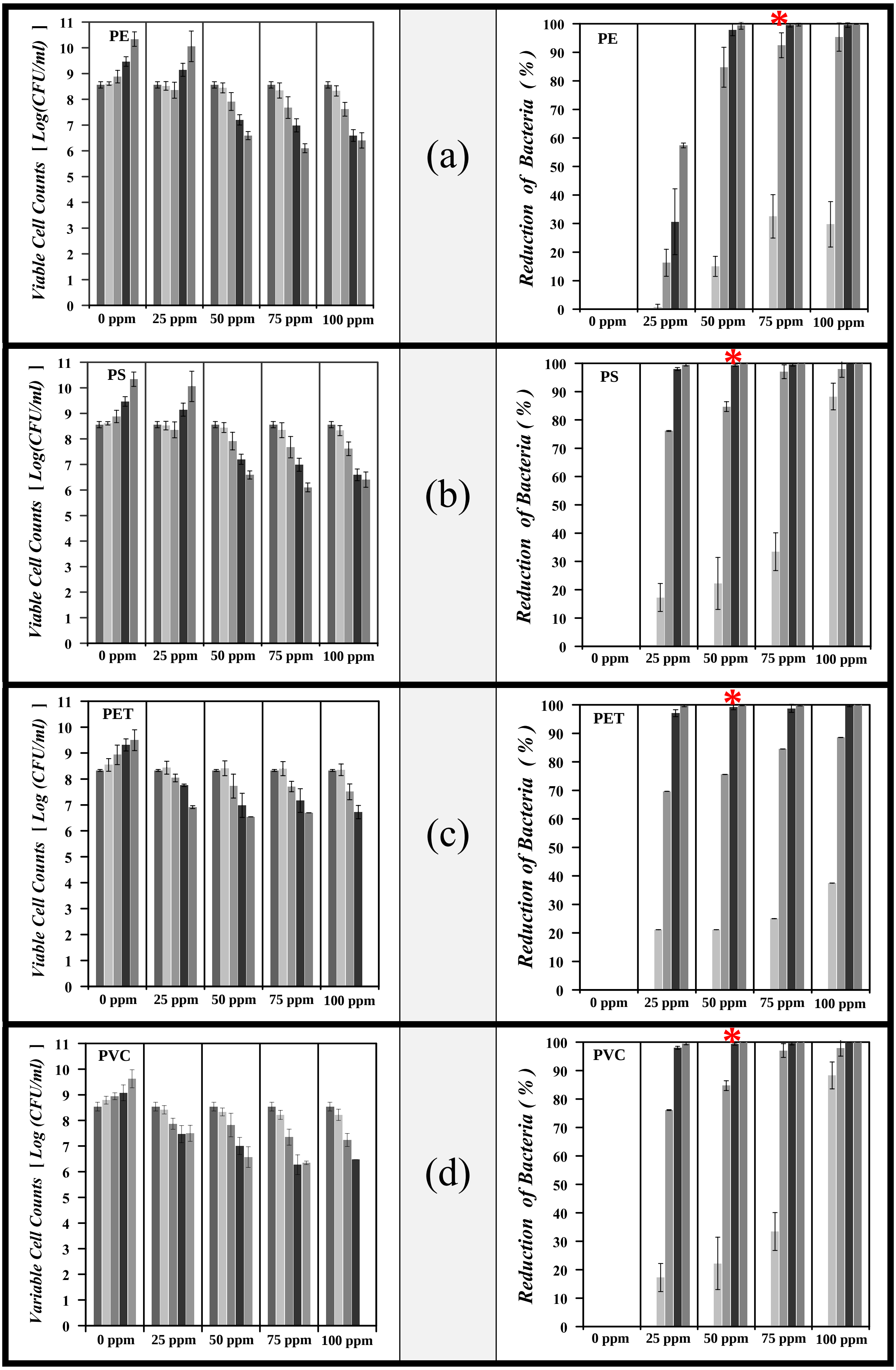

Figure 2. Effects of nano-silver content and contact times 0 min (■), 30min (■), 90 min (■), 150 min (■), and 210 min (■) on viable cell count and antibacterial efficacy of nano-silver colloid coated specimens: (a) PE (b) PS (c) PET (d) PVC

(a)

(b)

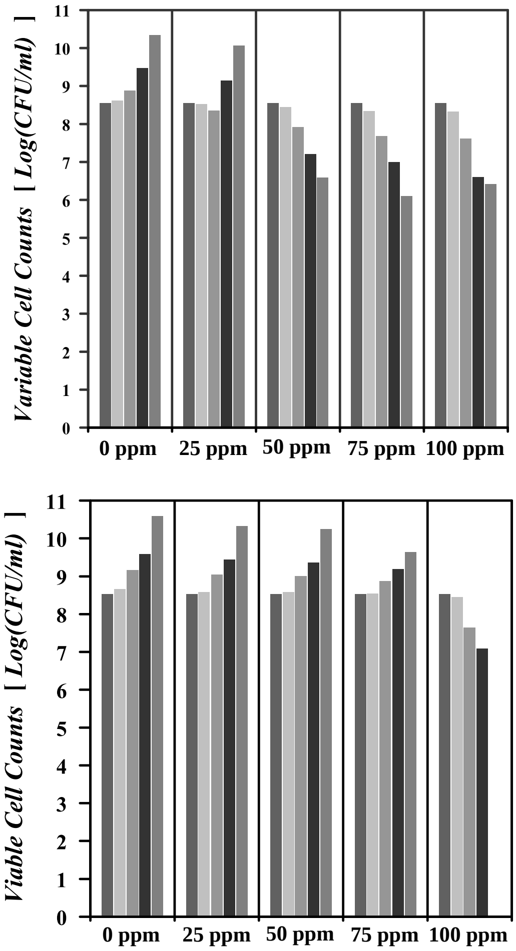

Figure 3. Comparison of viable cell count of PE sample having different dosages of nano-silver which were incorporated by spray-coating (a) and dry-blending (b) techniques with contact times 0 min (■), 30min (■), 90 min (■), 150 min (■), and 210 min (■)

Conclusion

In all thermoplastics used, it was observed that the viable cell count decreased with increasing the content of nano-silver colloid. The contact time at 150 min was sufficient for 99% reduction of *E. coli*. 75 ppm nano-silver colloid was required for PE whereas 50 ppm nano-silver were required for PS, PET and PVC for achieving the maximum percentage reduction of *E. coli* using the contact time of 150 min. For a given nano-silver content, the spray-coating technique was much more effective than that dry-blending technique

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