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Antimicrobial activity of EVOH based nanocomposite prepared by simple saponification

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ABSTRACT

Various EVOH based nanocomposite particles were prepared by incorporating nano-sized clay, pozzolan or TiO_2 in a poly(ethylene-*co*-vinyl acetate) (EVA)/toluene solution and precipitating in ethanol/KOH solution. After 6 h saponification time, the nanocomposite particles shape recovered was almost the same as the parent particles. The antibacterial activity of prepared nanocomposite powders was determined by shake flask test against *S. aureus* and *E. coli*. Among the nanocomposites used in this study, 6 h saponified EVA/MMT-50% nanocomposite exhibited strong antibacterial activity against the two kinds of bacteria. The biocidal nanocomposites were also tested for resistance to fungal growth in accelerated tests according to ASTM D5590. As the saponification time increased, the antifungal activity against *A. niger* and *P. funiculosum* of all nanocomposites was increased. After 7 days of incubation, saponified EVA/Loess nanocomposite film exhibited highest inhibitory ability against both fungal strains.

KEYWORDS

Nanocomposite;
Copolymer;
Morphology;
Antimicrobial;
Poly(ethylene-*co*-vinyl acetate).

INTRODUCTION

Microbial contamination of paint and coating layer leads to a variety of detrimental outcomes like mould formation, odor development, staining, discoloration and spread of surface borne diseases to humans. Even though they cannot be directly assimilated by microorganisms, microbes can grow and propagate using bioassimilable contaminants on the surface of the coating layer. One possible way to avoid microbial contamination is to develop materials that possess antimicrobial activities[1-9]. Moreover, increased efficiency, selectivity, and handling safety are additional benefits which may be realized[5].

Antimicrobial additives are a crucial component of paints and coating agents. Because of increased environmental legislation, coatings technology is moving toward low volatile organic compounds systems such as water-borne latexes. Antimicrobial agents are especially critical in these cases because the coatings are water-based and contain carbon-based polymers and surfactants that are an excellent food source for bacteria, algae, and fungi. They are needed to prevent the growth of bacteria and fungi in paint cans before the cans are opened and used, and to protect paint from attack by algae and fungi after it is applied and dried.

Antimicrobial agents used in antimicrobial-processed products are classified into organic, inorganic and natural organic compounds. Organic antimicrobial agents raise health concerns and many of them do not have sufficient antimicrobial activity. Polymeric biocides can significantly reduce loss of antimicrobial activity associated with volatilization, photolytic decomposition, dissolution, and permeation[5]. On the other hand, inorganic antimicrobial agents employ Ag, Cu, and Zn compounds and are excellent in safety and antimicrobial activity. These agents are used in many types of household and medical products due to their good balance between antimicrobial activity and endurance. However, patients with metal allergy due to Cu or Zn have been reported[10]. Moreover, the regulatory process for antimicrobial agent approval is lengthy and expensive. Many active antimicrobial agents are commercially available but they are typically used in combination with one another because each antimicrobial agent is active against only a small number of fungal or algal strains. Rather than trying to develop a new, universal antimicrobial agents that would protect against a wide variety of microbial threats over a long period of time.

The interest in polymer nanocomposites with antimicrobial properties is continuously increasing due to the growing demand for healthy living. More and more such materials are produced using different technologies in order to achieve desired properties. Potential fields of application include, for instance, textile, industrial and food packaging or medical devices to prevent nosocomial infections. Recently, several natural and modified clay minerals have significant potential for antimicrobial materials[11-13] because of its large surface area and charged particles, which can result in strong interactions with negatively charged microbial. In this study, various poly(ethylene-*co*-vinyl alcohol) (EVOH) based nanocomposite particles were prepared by alkaline saponification using a suspension of nano-sized clay, titanium dioxide (TiO_2), pozzolan or anion release powder in poly(ethylene-*co*-vinyl acetate) (EVA)/toluene solution, and their antibacterial and antifungal activity were investigated. The prepared nanocomposite powders were mixed with aqueous sodium silicate (Na-silicate). The aqueous Na-silicate as a binder is used in various paints and coatings[14]. The morphology of nanocomposites filler dispersion in the Na-silicate matrix was also evaluated.

EXPERIMENTAL

Materials

Montmorillonit (MMT, Cloisite®Na^+, Southern Clay Products, Inc. TX, USA), anatase TiO_2 (A-TiO_2, NT-22, Nano Co., Ltd., Kyungnam, Korea), Pozzolan (Ge-Lite, DSG Co., Ltd., Kangwondo, Korea) and EVA (1159, Hanhwa Chemical Co., Ltd., Seoul, Korea) were used as received. Loess powder (HT-P100A), anion powder (RT-P20000) and rutile TiO_2 (R-TiO_2, YH-P100) were purchased by Shanghai Huzheng Nano Technology Co., Ltd. (Shanghai, China). EVA28 [1159 (density: 0.949 g/cm^3, vinyl acetate content: 28 wt%, melt flow index: 18 g/10 min at 190°C/2.16kg), Hanhwa Chemical Co., Ltd., Seoul, Korea] was used as received. EVA resin and nanofillers were pre-dried in a vacuum drying oven for at least 12 h at 40 °C to remove any moisture from the pellets before processing

Instrumentation

Scanning electron microscopy (SEM) observations of the samples were performed on a Hitachi S-4300 model (Tokyo, Japan). The surfaces of the specimens were prepared by using cryogenic fracturing in liquid nitrogen followed by a coating with platinum in an SPI sputter coater. The morphology was determined using an accelerating voltage of 15 kV. The surface sample composition was evaluated using SEM (Hitachi S-4300 model, Tokyo, Japan) equipped with an energy dispersive X-ray spectroscopy (EDX).

Film specimens were prepared by pressing the composites on a hot press at a plate press at 150 - 250 °C for 10 min under about 5 atm and quickly immersed into water. The sheet thus formed was free from any distortion problems. The films obtained were allowed to dry at 60 °C for 24 h. The final thickness of the dried films was in the range of - 0.5 mm.

Preparing of EVOH based nanocomposite powders

Various EVOH based nanocomposites were prepared with nanofiller loadings of 50 wt%. The EVA pellets (100 g) were first swelled in toluene (900 g) at room temperature for 12 h and followed by heating at

60 °C for 4 h. The required amount of nanofillers was each dispersed in the 10 wt% toluene solution of EVA at room temperature by mechanical premixing and bath sonication for 2 h. 20 ml of diluted suspensions (5.0 wt% in toluene) were saponification by dropwise addition to 200 ml of 0.5 M KOH in ethanol solution (1000 ml of ethanol/28.05 g of KOH solution). The heterogeneous solution was stirred at room temperature for ambient time, and then the solution was filtrated, and filtrate washed with methanol. The filtrate was dried under vacuum at 60 °C to a constant weight.

Shake flask test

The antibacterial activity of the nanocomposite powders was tested against *Staphylococcus aureus* (*S. aureus*, ATCC 25923) and *Escherichia coli* (*E. coli*, ATCC 25922) with the shake flask method. The bacteria were subcultured on nutrient broth and incubated for 20 h at 37 °C. The cells were suspended in 50 ml of phosphate-buffered saline (PBS) to yield a bacterial suspension of 1.70×10^7 - 1.16×10^9 colony forming units/ml (cfu/ml). The sample powder (0.5 g) was weighed and shaken in 20 ml of a bacterial suspension for 24 h. The suspension (25 wt/vol%) was serially diluted in PBS and cultured on nutrient broth at 37 °C for 24 h. The number of viable organisms in the suspension was determined by multiplication of the number of colonies with the dilution factor, and the percentage reduction was calculated on the basis of the initial count

Antifungal test

The antifungal activity of specimens was evaluated by the standard ASTM D 5590 test. The fungi used in this study were *Aspergillus niger* (*A. niger*, ATCC 6275) and *Penicillium funiculosum* (*P. funiculosum*, ATCC 11797). Cultures of *A. niger* and *P. funiculosum* were prepared by incubation at 28 °C in potato dextrose agar (PDA) for 72 h. By diluting with 5 ml of sterile distilled water, a culture containing about 1.2×10^6 cells/ml was prepared for each strain and used for the antifungal tests. The sterile Petri dish containing PDA was inoculated by this culture. 30 mm x 30 mm x 0.5 mm of hot-pressed EVOH nanocomposites film was sterilized by UV for 1 h and was placed in the center of the inoculated Petri dish. Thin coat of fungal suspension was applied to specimen using a sterile atomizer until the surface is covered. The agar dish was then incubated at 28 °C for 7 days. The growth rating was determined according to TABLE 1.

RESULTS AND DISCUSSION

Figure 1 shows the SEM micrograph of nanofillers. MMT and Loess nanoclays showed a dominant grain-size ranging from 1 to 5 μm, although agglomerated particles may be several time larger. The results of the EDX analysis of two nanoclays showed that the common elements are C, O, Al, Si, Mg, and Fe. Loess showed considerably low Mg and K contents (TABLE 2).

A-TiO_2 and R-TiO_2 have a primary particle-size ranging from 15 to 35 nm. R-TiO_2 contained some impurities which are corresponding to Al and Si. A-TiO_2 also showed very low Al content. The pozzolan and anion nanopowders have a dominant grain-size ranging

TABLE 1 : Fungal growth rating

Observed growth on specimens	Rating
None	0
Traces of growth (<10%)	1
Light growth (10-30%)	2
Moderate growth (30-60%)	3
Heavy growth (60% to complete coverage)	4

TABLE 2 : EDX analysis results of the nanofillers[15].

Filler	Element (wt%)														
	C	O	Al	Si	Ti	Na	Mg	Fe	K	Ca	P	La	Ce	Nd	Th
MMT	2.8	67.8	7.2	17.5	-	2.3	1.0	1.4	-	-	-	-	-	-	-
Loess	2.3	69.0	12.2	13.9	0.6	-	0.3	1.5	0.2	-	-	-	-	-	-
A-TiO_2	4.2	50.9	0.2	-	44.7	-	-	-	-	-	-	-	-	-	-
R-TiO_2	0.9	41.6	1.1	0.8	55.6	-	-	-	-	-	-	-	-	-	-
Pozzolan	8.9	62.5	4.8	14.2	-	1.0	1.0	3.0	1.8	2.8	-	-	-	-	-
Anion	4.3	40.1	-	1.3	-	-	-	1.8	-	1.2	8.1	9.1	22.9	7.2	4.0

from 1 to 8 μm. The pozzolan is somewhat similar to Loess but it contains 1.0 wt% of Na and 2.8 wt% of Ca. The anion nanofiller can release the 2000-3000 negative ions/cm^3 at room temperature. Its main elements are C, O, P, La, Ce, Nd and Th.

The SEM image in Figure 2 provides an EVA/nanofiller coagulant surface prepared using ethanol/KOH solution after 6 h saponification time. The surface of the unsaponified EVA/nanofiller coagulant shows a dense skin layer, which appears to be nonporous. As the saponification time increased, the surface of the nanocomposite particles is eroded and the larger particles are delaminated and fractured into smaller particles due to changing matrix polymer polarity. After 6h saponification time, the particle shape recovered was almost the same as the parent particles (Figure 1).

Antibacterial activity of nanocomposite powders

The antibacterial activity of the nanofillers and nanocomposite powders was tested against *S. aureus*, and *E. coli* with the shake flask method. *S. aureus* and *E. coli* are two of the most common nosocomial pathogens[16,17] and they represent Gram-positive and Gram-negative bacteria, respectively. The percentage reduction of nanofillers and nanocomposite powders are compared in Figure 3 and Figure 4.

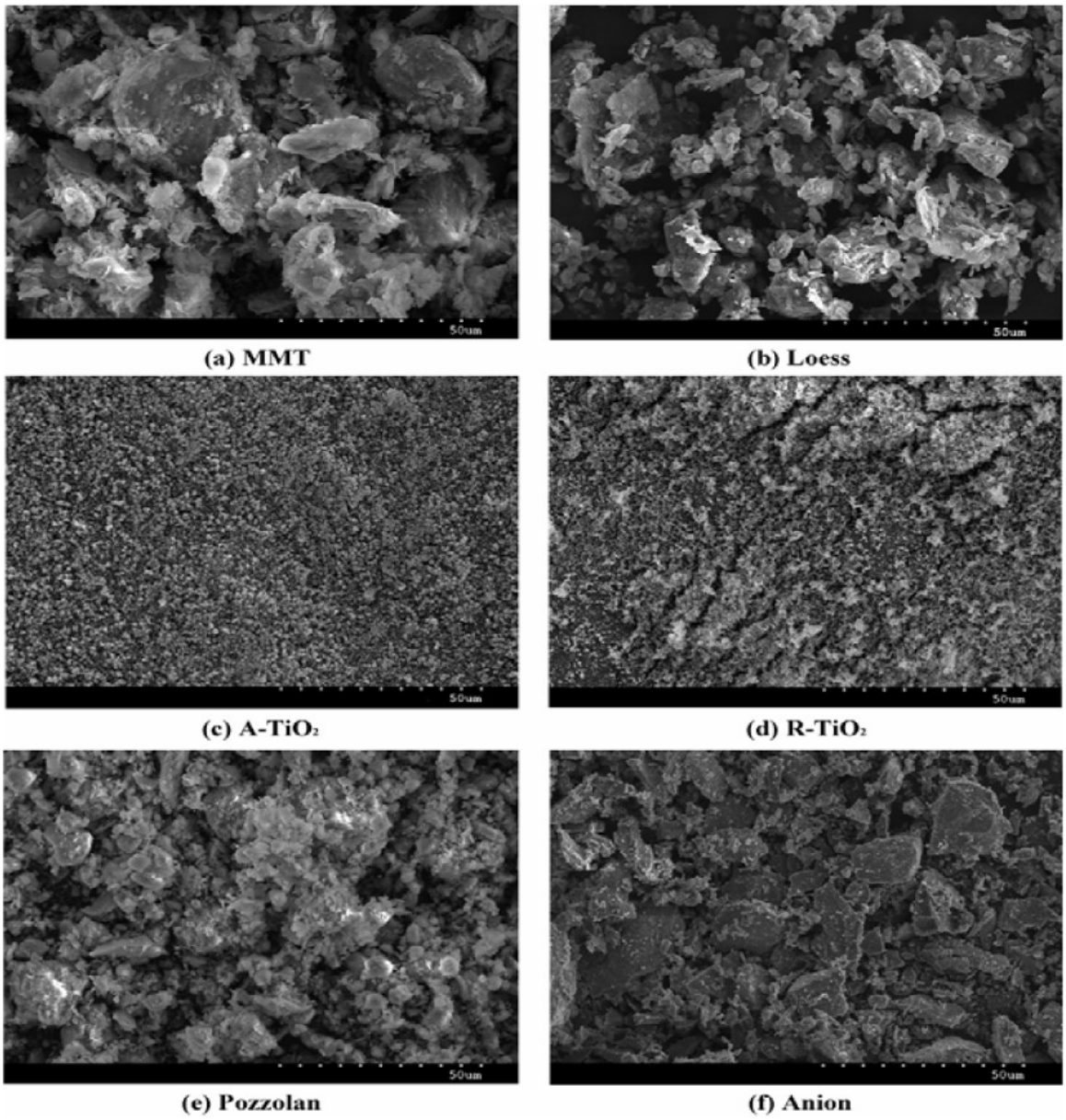

Figure 1 : SEM micrographs of the pristine nanofillers[15].

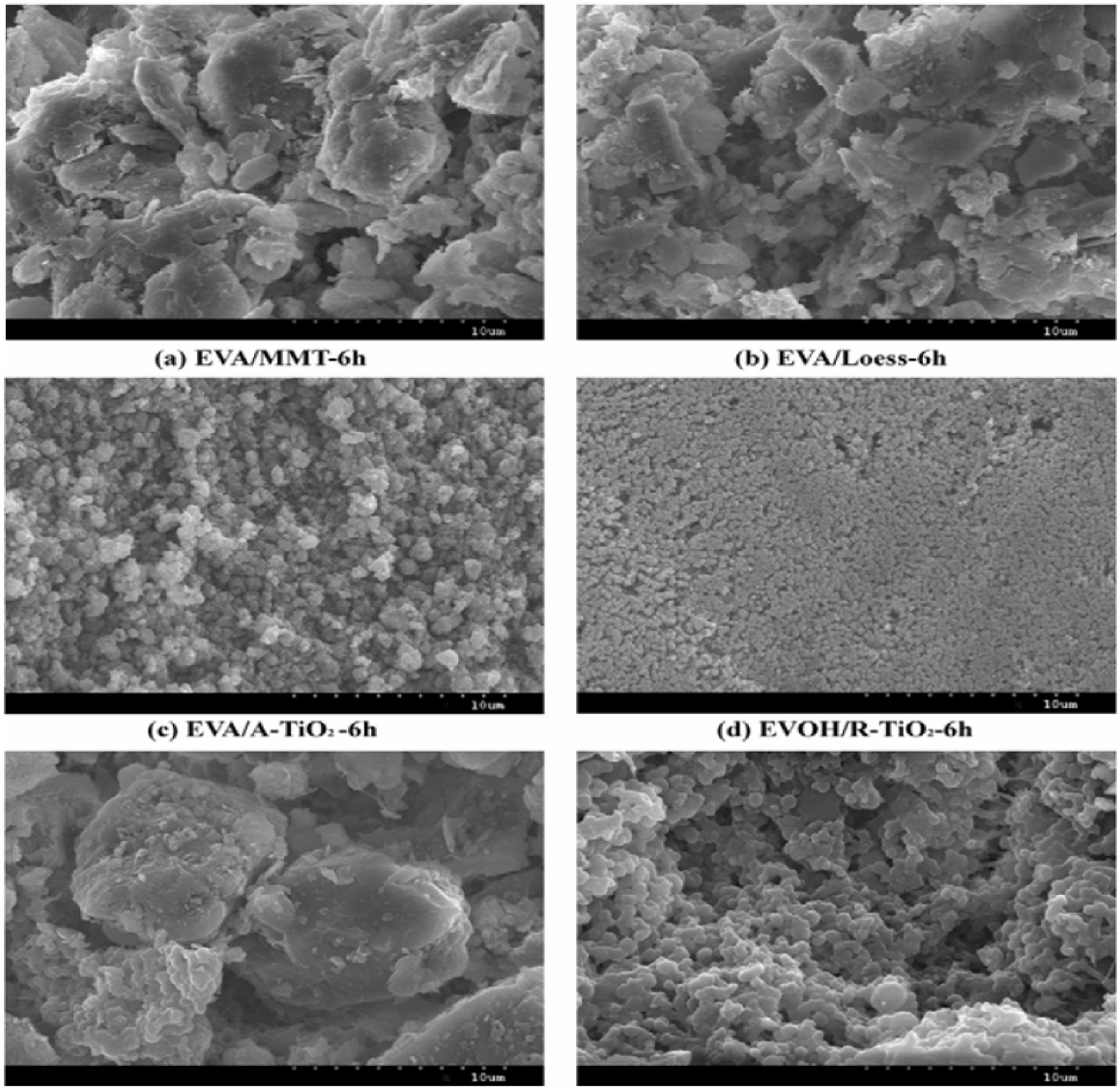

Figure 2 : SEM micrographs of the EVOH based nanocomposites (nanofiller content = 50wt%)[15].

At 25 wt/vol% in the bacterial suspension, A-TiO_2 reduces viable cell number of both *S. aureus* and *E. coli* significantly than other nanofillers indicating that A-TiO_2 has a potent antimicrobial agent than other nanofillers. A-TiO_2 extirpated 20.7 and 63.4 % of the viable cells of *S. aureus* and *E. coil*, respectively (Figure 3).

The photo-catalytic agent TiO_2 has been used extensively for killing different groups of microorganisms including bacteria, fungi and viruses, because it has high photo-reactivity, broad-spectrum antibiosis and chemical stability[18-21]. The photocatalytic activity of annealed TiO2 sturdily depends upon its existing phase, i.e., anatase, rutile, brokite. The anatase phase shows an indirect optical band gap of 3.2 eV, while the rutile phase has a direct band gap of 3.06 eV and an and an indirect one of 3.10 eV[22]. The killing mechanism involves degradation of the cell wall and cytoplasmic membrane due to the production of reactive oxygen species such as hydroxyl radicals and hydrogen peroxide. This initially leads to leakage of cellular contents then cell lysis and may be followed by complete mineralisation of the organism. Killing is most efficient when there is close contact between the organisms and the TiO_2 catalyst. The killing activity is enhanced by the presence of other antimicrobial agents such as Cu and Ag[23].

On the other hand, the R-TiO_2 and Anion had similar inhibitory behavior with *S. aureus* and *E. coli*. R-TiO_2 and Anion selectively inhibit towards *E. coli* compare with *S. aureus*. After 24 h of shaking, the R-TiO_2

showed the 4.7 and 58.9 % inhibition of the growth of *S. aureus* and *E. coil* whereas for Anion extirpated - 12.0 and 30.7 % of the viable cells of *S. aureus* and *E. coil*, respectively. It can be also found that except for the Loess the other nanofillers inhibited the growth of *E. coli* more than *S. aureus* (Figure 3).

The results of antibacterial activity of nanocomposites powders are presented in Figure 4. The EVA/MMT-50% nanocomposite powders showed stronger antibacterial activity against two strains than others. As the saponification time increased, the antibacterial activity towards *S. aureus* (Figure 5) and *E. coli* (Figure 6) of EVA/MMT-50% nanocomposite was gradually increased. It is noteworthy that 6h-saponified EVA/MMT-50% exhibits highest antibacterial activity against the two kinds of bacteria. After 24 h of shaking, the nanocomposite powder showed the 94.9 and 99.2 % inhibition of the growth of *S. aureus* and *E. coil*. The antibacterial behavior of saponified EVA/Loess-50% was the almost same as that of EVA/MMT-50%. However, EVA/Loess-50% showed more active on *S. aureus* than *E. coli* on up to 6 h saponification time. EVA/Loess-50%-6h exhibited 65.4 and 39.2 % inhibition of the growth of *S. aureus* and *E. coil*, respectively.

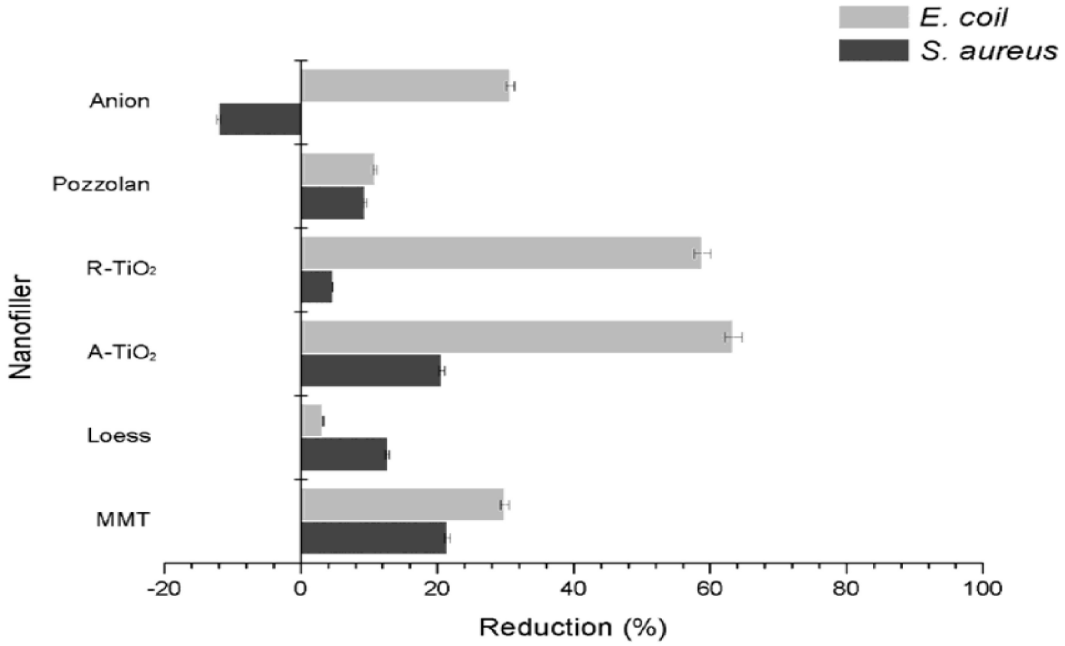

Figure 3 : Shake flask test results for the nanofiller.

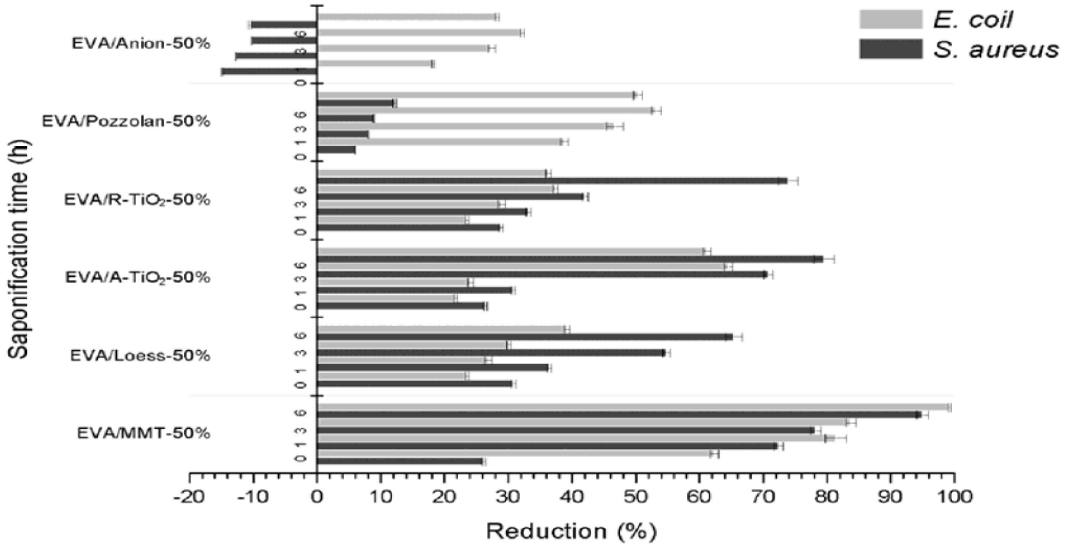

Figure 4 : Shake flask test results for the nanocomposites as a function of saponification time.

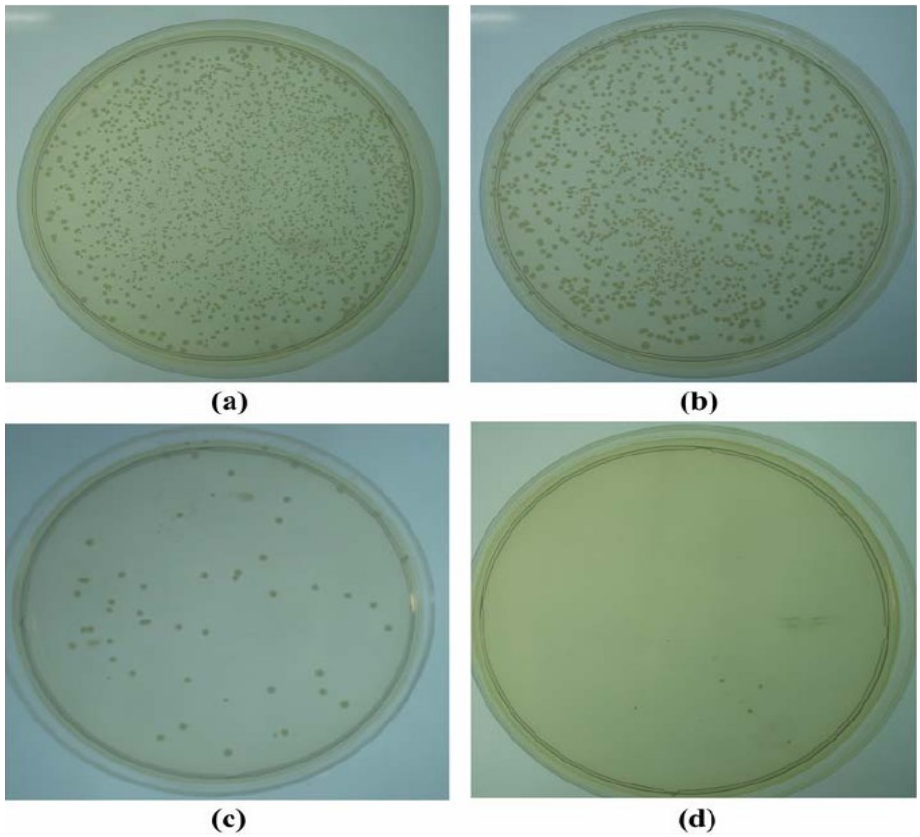

Figure 5 : Effect of EVA/MMT-50% [saponification time (a) 0h, (b) 1h, (c) 3h, and (d) 6h] on numbers of cfu of *S. aureus*.

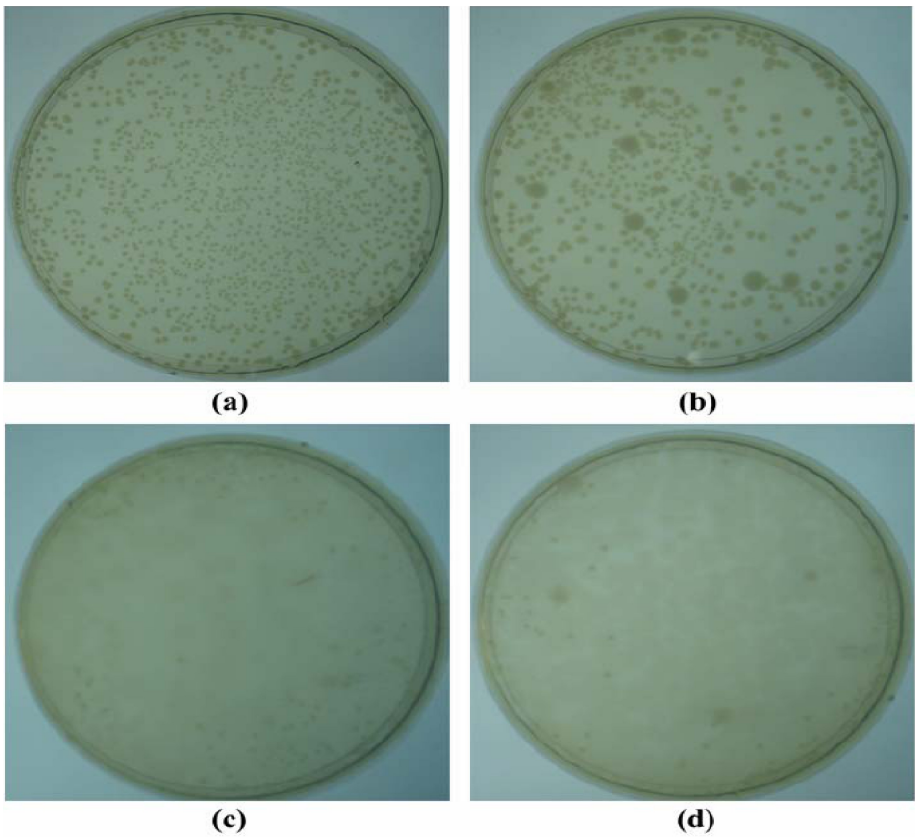

Figure 6 : Effect of EVA/MMT-50% [saponification time (a) 0h, (b) 1h, (c) 3h, and (d) 6h] on numbers of cfu of *E. coil*.

In case of EVA/R-TiO_2-50% and EVA/A-TiO_2-50%, the antibacterial activity against *S. aureus* was gradually increased with the saponification time. As expected, EVA/A-TiO_2-50%-6h and EVA/R-TiO_2-50%-6h exhibited 79.5 and 73.9 % inhibition of the growth of *S. aureus*, respectively. It can be also seen that EVA/A-TiO_2-50% and EVA/R-TiO_2-50% nanocomposites were more active on *S. aureus* than *E. coli* on up to 6 h saponification time.

The antibacterial effect of the saponified EVA/Pozzolan-50% was higher against *E. coil* regardless of the saponification time. In sharp contrast the EVA/Anion-50% had only antimicrobial activity on *E. coil*, but had no inhibitory effect on *S. aureus*.

It is noted that the antibacterial activity against *E. coil* of EVA/A-TiO_2-50%, EVA/R-TiO_2-50%, EVA/Pozzolan-50% and EVA/Anion-50% nanocomposites were increased up to 3 h saponification time and decreased thereafter. This indicated that there was an optimum VAc content for antibacterial activity against *E. coil*. EVA is a random copolymer consisting of ethylene and VAc as repeating units. VAc content has two fundamental effects that influence the properties of EVA copolymers. The first effect is to disrupt the crystalline regions formed by the polyethylene segments of the copolymer. The second overriding effect of VAc content results from the polar nature of the acetoxy side chain[24].

When the VAc group in polymer molecules converted to hydroxyl group through saponification, the total polarity of molecules is increased due to electronegativity of -OH group (2.68) is higher than that of VAc (2.56) one[25]. After saponification numerous hydroxyl groups in EVOH form strong hydrogen bond, both inter- and intra-molecular, which reduce the free volume of the polymer chains. In case of EVA copolymers, the degree of hydrogen bonding can be controlled by varying ethylene and VOH ratios in the copolymer. This is the polarization of the -OH bonds, because the oxygen atoms are more electronegative than hydrogen atoms. However acetoxy group cannot form inter- and intramolecular hydrogen bonds.

In order to inactivate or kill microbes, the nanocomposite particles must come close to or touch the microbes. Such interactions are either attraction or repulsion. As most bacteria carry a net negative surface charge[26], adhesion of bacteria is discouraged on negatively charged surfaces, while it is promoted on positively charged surfaces[27]. The increase in polarity of nanocomposites after saponification is reflected in the relative polar surface area, hydrogen bond donor, and hydrogen bond acceptor numbers, all of which increase substantially for antibacterial activity.

Antifungal activity of nanocomposite powders

The biocidal nanocomposites were tested for resistance to fungal growth in accelerated tests according to ASTM D5590, which is an accelerated test to determine the relative resistance of coating films to fungal growth. This is an agar-based method evaluating coated surfaces for their resistance to commonly occurring indoor and exterior fungi such as *A. niger*, *P. funiculosum* and *A. pullulans*. The antifungal activity of hot-pressed film of saponified nanocomposite is summarized in TABLE 3.

The *A. niger* and *P. funiculosum* grew on all surfaces of the unsaponified nanocomposite film. Among them, EVA/MMT-50% and EVA/A-TiO_2-50%showed very poor inhibition activity against *A. niger*. As the saponification time increased, the antifungal activity against *A. niger* and *P. funiculosum* of all nanocomposites was increased. After 7 days of incubation, saponified EVA/Loess nanocomposite film exhibited highest inhibitory ability against both strains. Except that the saponified nanocomposite films inhibited the growth of *P. funiculosum* more than *A. niger*. As shown in Figure 7, *A. niger* spores and their subsequent growth on the surface of EVA/MMT film completely inhibited after 6h saponification time whereas for *P. funiculosum* inhibited after 1h saponification time (Figure 8).

TABLE 3 : Antifungal activity for nanocomposite film

Sample	Antifungal activity							
	A. niger				*P. funiculosum*			
	0h	1h	3h	6h	0h	1h	3h	6h
EVA/MMT-50%	3	2	2	0	1	0	0	0
EVA/Loess-50%	1	0	0	0	1	0	0	0
EVA/A-TiO_2-50%	3	1	0	0	1	0	0	0
EVA/R-TiO_2-50%	1	1	1	0	1	0	0	0
EVA/Pozzolan-50%	1	1	0	0	1	0	0	0
EVA/Anion powder	2	1	1	0	2	0	0	0

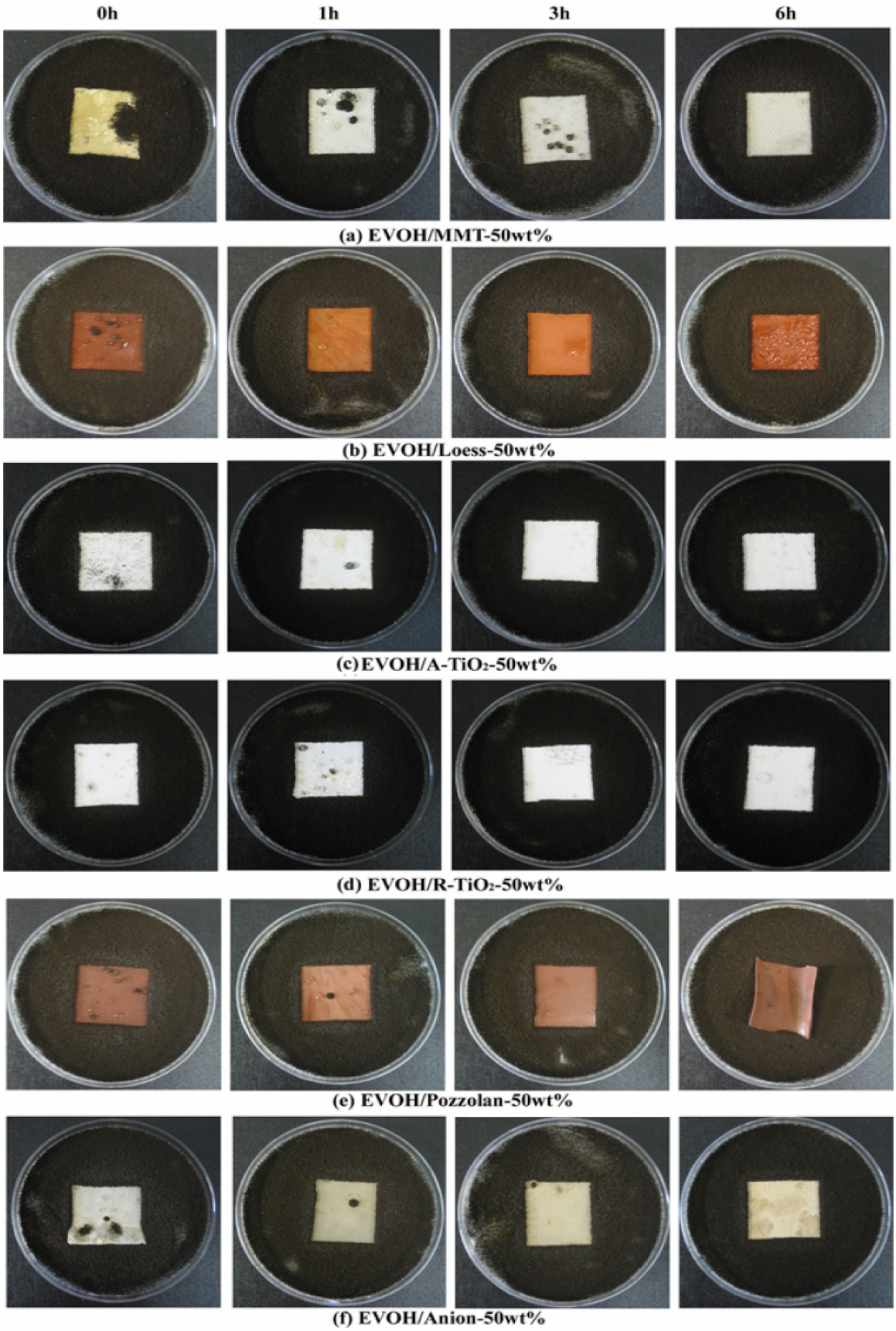

Figure 7 : Image of the saponified EVA/MMT nanocomposites after antifungal test for 7 days.

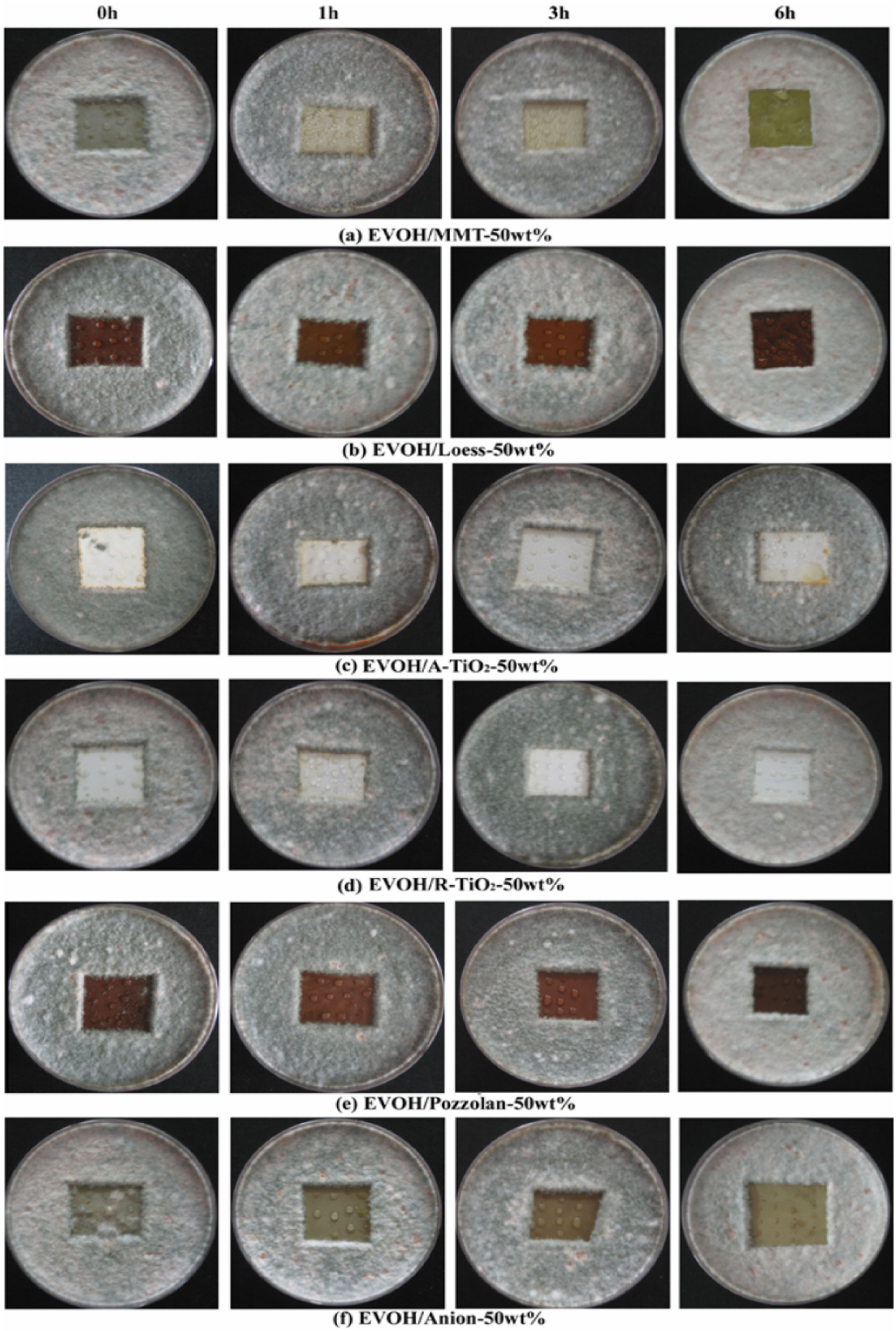

Figure 8 : Image of the saponified EVA/MMT nanocomposites after antifungal test for 7 days.

Full Paper

The nanoparticles prepared in this study also were designed for compatibility with a standard water-borne exterior coating formulation. For aqueous coating systems, a uniform and stable dispersion of fillers plays an important role. This requirement is especially critical when nano-sized inorganic fillers are involved. Their naturally hydrophilic surface not easily wetted by polymeric binder compositions which are generally hydrophobic. A further disadvantage of inorganic fillers is that they generally have associated with them a small, but significant, quantity of water[28]. EVOH copolymers have been widely used as food packages, biomedical and pharmaceutical industries due to their excellent gas barrier properties, high resistance to oils, good mechanical strength and harmlessness to health[29,30]. They also have significant potential for inorganic filler surface modifier due to their combined effect of hydrophilicity, as a consequence of the -OH side groups[31].

Figure 9 represents a fractured surface of the prepared Na-silicate/EVA/nanofiller-6h hybrid composites. 2 wt% of 6h-saponified nanocomposite powders were added into aqueous Na-silicate (98 wt%) and mixed to prepare a suspension of Na-silicate and nanocomposite powders. The antimicrobial coating agents made with nanocomposites particles were stable in the can for more than 6 months. These properties are required for ultimate commercialization; antimicrobial nanocomposites particles that can be added easily to an existing coating

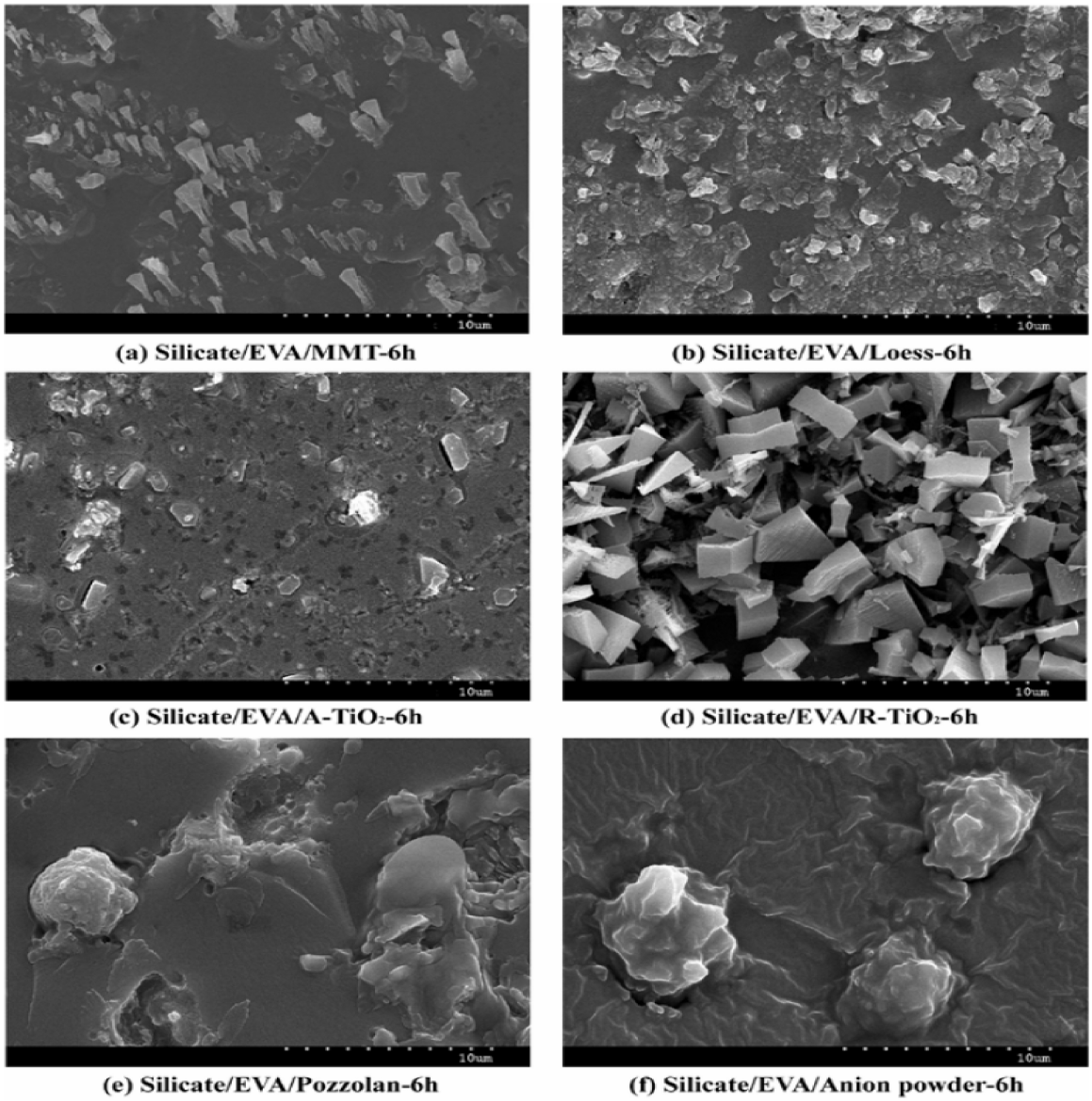

Figure 9 : SEM micrographs of the hybrid nanocomposites[15].

formulation without requiring significant changes in the formulation or interrupting the integrity of the formulation are much more likely to be considered. The suspensions were casted onto a PTFE film-supporting surface for 3 days. The films obtained were allowed to dry at 60 °C for 24 h. The final thickness of the dried films was in the range of 0.4 - 0.5 mm.

SEM analysis shows nanocomposites powder containing hybrid composites which are well distributed in the matrix with its morphology depending on the parent filler type. In the micrographs taken on the fracture surface of the Na-silicate/EVA/MMT-6h and Na-silicate/EVA/Loess-6h hybrid composites, thin plate-like structures with dimensions of 1 μm in Na-silicate matrix were observed.

In case of the EVA/A-TiO_2-6h and EVA/R-TiO_2-6h containing hybrid composites had hexagonal- or tetragonal-shaped crystal structure. In sharp contrast, the EVA/Pozzolan-6h and EVA/Anion powder-6h particles in the hybrid composite did not show any morphological change compared to their corresponding 6h-saponified particles.

CONCLUSIONS

Various EVOH based nanocomposite powders were successfully prepared by simple saponification using suspension of nano-sized clay, TiO_2, pozzolan or anion release powder in EVA/toluene solution. With the saponification time, the surface of the nanocomposite particles is eroded and the larger particles are delaminated and fractured into smaller particles due to changing matrix polymer polarity. After 6h saponification time, the particle shape recovered was almost the same as the parent particles.

The antibacterial activity of prepared nanocomposites was compared against *S. aureus* and *E. coli* with the shake flask method. The EVA/MMT-50% nanocomposite powders showed stronger antibacterial activity against two strains than others. The antibacterial activity against *E. coil* of EVA/A-TiO_2-50%, EVA/R-TiO_2-50%, EVA/Pozzolan-50% and EVA/Anion-50% nanocomposites were increased up to 3 h saponification time and decreased thereafter. This indicated that there was an optimum VAc content for antibacterial activity against *E. coil*. The increase in polarity of nanocomposites after saponification is reflected in the relative polar surface area, hydrogen bond donor, and hydrogen bond acceptor numbers, all of which increase substantially for antibacterial activity.

The biocidal nanocomposites were tested for resistance to fungal growth in accelerated tests according to ASTM D5590. The antifungal test results showed that the antifungal activity against *A. niger* and *P. funiculosum* of all nanocomposites was increased with the saponification time. After 7 days of incubation, saponified EVA/Loess film showed the highest inhibitory ability among six nanocomposite films tested. Since the nanocomposite powders prepared by this method have good antibacterial and antifungal activity, good dispersity in aqueous solution and easy processability they can be used for industrial applications such like functional filler for paint and coating materials in aqueous system, antimicrobial filler for polymer compounds and plastic film to make antimicrobial packaging.

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CONFLICT OF INTERESTS

This work does not have a direct financial relation with the commercial identities mentioned in the paper that might lead to a conflict of interests for any of the authors.

REFERENCES

[1] E.S.Park; J.Appl.Polym.Sci., **110**, 1723-1729 **(2008)**.
[2] H.S.Yang, E.S.Park; Macromol.Mater.Eng., **291**, 621-628 **(2006)**.
[3] J.H.Kim, E.S.Park, J.H.Shim, M.N.Kim, W.S.Moon, K.H.Chung, J.S.Yoon; J.Agric.Food Chem., **52**, 7480-7483 **(2004)**.
[4] E.S.Park, H.K.Kim, J.H.Shim, M.N.Kim, J.S.Yoon; J.Appl.Polym.Sci., **93**, 765-770 **(2004)**.
[5] E.S.Park, H.S.Kim, M.N.Kim, J.S.Yoon; Eur. Polym.J., **40**, 2819-2822 **(2004)**.

[6] W.S.Moon, K.H.Chung, D.J.Seol, E.S.Park, J.H.Shim, M.N.Kim, J.S.Yoon; J.Appl.Polym.Sci., **90**, 2933-2937 **(2003)**.
[7] W.S.Moon, J.C.Kim, K.H.Chung, E.S.Park, M.N.Kim, J.S.Yoon; J.Appl.Polym.Sci., **90**, 1797-1801 **(2003)**.
[8] E.S.Park, H.J.Lee, H.Y.Park, M.N.Kim, K.H.Chung, J.S.Yoon; J.Appl.Polym.Sci., **80**, 728-736 **(2001)**.
[9] E.S.Park, W.S.Moon, M.J.Song, M.N.Kim, K.H.Chung, J.S.Yoon; Int.Biodeter.Biodegr., **47**, 209-214 **(2001)**.
[10] H.Nakashima, N.Miyano, T.Takatuka; J.Health Sci., **54**, 390-399 **(2008)**.
[11] S.E.Haydel, C.M.Remenih, L.B.Williams; J.Antimicrob.Chemother., **61**, 353-361 **(2008)**.
[12] P.Herrera, R.C.Burghardt, T.D.Phillips; Vet. Microbiol., **74**, 259-272 **(2000)**.
[13] M.J.Wilson; J.Chem.Ecol., **29**, 1525-1547 **(2003)**.
[14] J.Hazziza-Laskar, G.Helary, G.Sauvet; J.Appl. Polym.Sci., **58**, 77-84 **(1995)**.
[15] E.J.Lee, E.S.Park; Recent Res.Devel.Appl.Pol.Sci., ISBN: 978-81-308-0520-7, **5**, 35-73 **(2013)**.
[16] E.Kenawy, F.I.Abdel-Hay, A.El-Raheem, R.El-Shanshoury, M.H.El-Newehy; J.Polym.Sci., Part A: Polym.Chem., **40**, 2384-2393 **(2002)**.
[17] J.Hazziza-Laskar, G.Helary, G.Sauvet; J.Appl. Polym.Sci., **58**, 77-84 **(1995)**.
[18] V.Nadtochenko, N.Denisov, O.Sarkisov, D.Gumy, C.Pulgarin, J.Kiwi; J.Photochem.Photobiol., A: Chem., **181**, 401-407 **(2006)**.
[19] A.-G.Rincón, C.Pulgarin; Appl.Catal., B: Environ., **51**, 283-302 **(2004)**.
[20] J.C.Yu, W.Ho, J.Lin, H.Yip, P.K.Wong; Environ.Sci. Technol., **37**, 2296-2301 **(2003)**.
[21] J.-S.Hur, Y.Koh; Biotechnol.Lett., **24**, 23-25 **(2002)**.
[22] W.F.Zhang, M.S.Zhang, Z.Yin, Q.Chen; Appl.Phys. B, **70**, 261-265 **(2000)**.
[23] Foster, A.Howard, Ditta, B.Iram, S.Varghese, A.Steele; Appl.Microbiol.Biotechnol., **90**, 1847-1868 **(2011)**.
[24] J.J.George, A.K.Bhowmick; Nanoscale.Res.Lett., **4**, 655-664 **(2009)**.
[25] S.G.Bratsch; J.Chem.Edu., **62**, 101-103 **(1966)**.
[26] B.A.Jucker, H.Harms, A.B.Zehnder; J.Bacteriol., **178**, 5472-5479 **(1996)**.
[27] A.H.Hogt, J.Dankert, J.Feijen; J.Biomed.Mater. Res., **20**, 533-545 **(1986)**.
[28] E.J.Lee, J.S.Yoon, M.N.Kim, E.S.Park; Carbon Nanotubes - Polymer Nanocomposites, S.Yellampalli, (Ed), InTech, Croatia, Chap 6, **(2011)**.
[29] S.Ramakrishnan; Macromolecules, **24**, 3753-3759 **(1991)**.
[30] A.Lasagabaster, M.J.Abad, L.Barral, A.Ares; Eur.Polym.J., **42**, 3121-3132 **(2006)**.
[31] E.J.Lee, J.S.Yoon, E.S.Park; J.Appl.Polym.Sci., **125**, E691-E704 **(2012)**.