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SYNTHESIS AND ANTIMICROBIAL ACTIVITY OF CERIUM OXIDE/AG DOPES SILICA MESOPOROUS MODIFICATION AS NANOFILLERS FOR FOOD PACKAGING APPLICATIONS

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ABSTRACT

Nanofiller of cerium oxide/ silver doped modified mesoporous silica (Ce/Ag-MMS) had been successfully synthesized by the hydrothermal method. Based on the results of the XRD analysis using the Scherer method, nanofiller Ce/Ag-MMS crystal sizes were obtained between 9.68 – 12.44 nm. The morphology of Ce/Ag-MMS nanofiller based on the results of SEM analysis was spherical with a uniform size. The particle size of Ce/Ag-MMS from TEM analysis was around 3.4 – 4.5 nm. The antimicrobial properties Ce/Ag-MMS nanofiller increased become 86.85% compared to cerium/silver nanofillers when used to inhibit the growth of *Staphylococcus aureus* bacteria. This nanofiller had been one of the alternatives to be exploited and developed for applications in active packaging. The use of nanofiller as an active packaging needs further economic assessment for industrial-scale production.

Key words: Nanofiller, cerium oxide, silver, mesoporous silica, active packaging

INTRODUCTION

Plastic was one of the objects that contribute to environmental pollution. Plastics can contaminate soil, water, and even air. Plastic water pollution occurs when plastic was washed away in the ocean and can pollute marine biota. Plastics on the ground break down very long (Arora *et al*., 2010; Auras *et al*., 2005). While the burned plastic releases chemicals that were harmful to anyone who breathes. Although it was common knowledge that plastic pollutes the environment, the use of plastic remains high (Averous, 2008). Indonesia has a major plastic waste problem on its hands. At the moment, the country is second only to China when it comes to dumping plastic waste into the world's oceans. According to a study by the University of Georgia, an estimated 3.22 million metric tons of plastic waste is tossed annually into the ocean surrounding Indonesia, while another 8.82 million metric tons of China's plastic waste also makes its way into the ocean (Syamsiro *et al*., 2014).

Research on bioplastics has been in the spotlight for many years. Research on biodegradable plastics has begun in Indonesia. Biodegradable plastics can be made from cassava starch, jackfruit, and others (Murariu *et al*., 2011). These plastics are widely used for active packaging. However, bioplastics made have a weakness, namely poor mechanical thermal and barrier properties when compared with non-biodegradable plastics. Therefore, many researches have been conducted to improve the nature of bioplastics (Narayanan *et al*., 2012). One way was to use nanocomposite. Nanocomposite consists of two parts, nanofiller, and biopolymer. Nanofiller was a nano-sized material that can improve the surface area of the fillers. The more surface area of material the more the interfacial large between biopolymers and nanofillers (Panea *et al*., 2014). Nanofillers used as food packaging consist of several types, namely nanoparticles, nanofibrils, nanorods, and nanotubes. Nanofiller classification was organic nanofiller which consists of clay, chitosan (natural biopolymer), and Nissin (natural antimicrobial agent). Research on the use

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of organic nanofiller for food packaging applications namely clay (Pillin *et al*., 2006), chitosans (Imran *et al*., 2012), Nissin (Narayanan *et al*., 2012). The use of inorganic nanofiller was silver, copper, and gold. The metal oxide inorganic nanofiller consist of ZnO, TiO_2, MgO, and AgO (Realini *et al*., 2014).

Metal oxide which is currently being highlighted, was also known as cerium oxide. Cerium oxide has the advantage of being able to oxygen scavenger. This advantage that causes cerium oxide can be used as a nanomaterial for active packaging (Bing *et al*., 2012). In this study, the synthesis of cerium oxide nanofillers was expected to absorb oxygen properly so that it can increase food storage time without reducing nutritional quality. Doping of cerium oxide nanofillers with modified mesoporous silica was expected to increase thermal stability, mechanical properties and barrier properties of the nanofiller. It was also expected that doped cerium oxide nanofillers with silver nanoparticles have effective antimicrobial properties and increase the rate of crystallization kinetics of polylactic acid used as biopolymers. The economic aspects of the use of nanofillers from oxide nanoparticles require a more in-depth study of the advantages and disadvantages if they are produced on an industrial scale.

MATERIALS AND METHODS

Chemicals and reagents

Cerium (III) nitrate hexahydrate ($Ce(NO_3)_3.6H_2O$), silver nitrate and Tetraethyl orthosilicate used for a precursor of CeO_2, Ag, and Silica. Acetic acid, ammonium solution, 2-propanol, polyvinyl alcohol (PVA), glucose was purchased from Merck & Co.

Synthesis of nanofiller cerium/silver doped modified mesoporous silica

Nanocrystalline Cerium oxide and Ag nanoparticles were produced in previous research (Putri *et al*., 2019) mixed with modified mesoporous silica (MMS) then added slowly into isopropanol. After that, the mixture of solutions was distilled for 24 hours so that Ce-Ag-MMS suspension was formed. The suspension was filtered, and the solid of materials had obtained, then it was dried at 80°C for 5 hours, after that it was calcined at 550°C for 6 hours to obtain nanofiller Ce-Ag-MMS. The success of the synthesis process of nanofiller Ce-Ag-MMS showed from the results of TEM, XRD, and SEM-EDX.

Characterisation

The powder X-ray diffractogram of synthesized and calcined samples was recorded on a Rigaku Miniflex diffractometer with Cu Kα radiation between 1.5 and 10o (2θ) with a scanning rate of 1o/min. TEM micrographs of the samples were obtained with a JEOL 100CX microscope with 100 kV of acceleration voltage. SEM merk JEOL-JSM 6360 LA.

Antimicrobial activity

The activity test of nanofiller Ce-Ag-MMS was carried out to follow the AATCC 147-1998. The type of bacteria used was *Staphylococcus aureus*. The qualitative or antimicrobial activity testing microbial inhibitory test carried out in a way make a series of dilution test compounds (colloidal nanofiller Ce-Ag-MMS) with a variety of concentration was 25, 50 and 100%. The control was done on reagent media test compound. The inhibitory test was done by carried out with wetting sterile paper discs with nanofiller Ce-Ag-MMS solution colloid produced by the mixture, then put on Petri dishes containing *Staphylococcus aureus* bacteria was grown on *Mueller Hinton Agar* (MHA) media. The zone of inhibitory the test material was known by measuring the width of the zone of inhibition around the paper discs (in mm).

RESULTS AND DISCUSSION

XRD analysis

XRD pattern of Nanopartirkel Cerium oxide showed in Figure 1, (a), Ag nanoparticles (b), cerium-Ag nanoparticles (c), mesopore modified silica (d), and cerium oxide doped nanofiller modified mesoporous silica (e). In Figure 1e the diffraction peak was the result of XRD combined silica, cerium and silver compounds, from the results of the analysis it appears the peaks of Ag, Ce-Ag, SiO_2, and Si-Ce-Ag peaks indicate the interaction of the three reacted compounds (e). Based on calculations using the Scherrer equation, the crystal size is between 9.68 - 12.44 nm. From these results the nanofiller produced has nano size (<100 nm). The fillers with nano-size have the advantage of being able to increase the mechanical and barrier properties of the matrix of nanocomposites (Putri *et al*., 2018a).

SEM analysis

Figure 2 showed the results of SEM analysis of various particles. Figure 2d showed that material produced shaped were spherical, uniform particle size, and cerium and silver material scattered throughout the surface of modified mesoporous silica. Silica nanoparticles have been reported to improve the mechanical and barrier properties of polymer matrices (Ahmed *et al*., 2002). Inter-molecular bonds from hydrogen bonds also increase, as well as the formation of C-O-Si groups, between

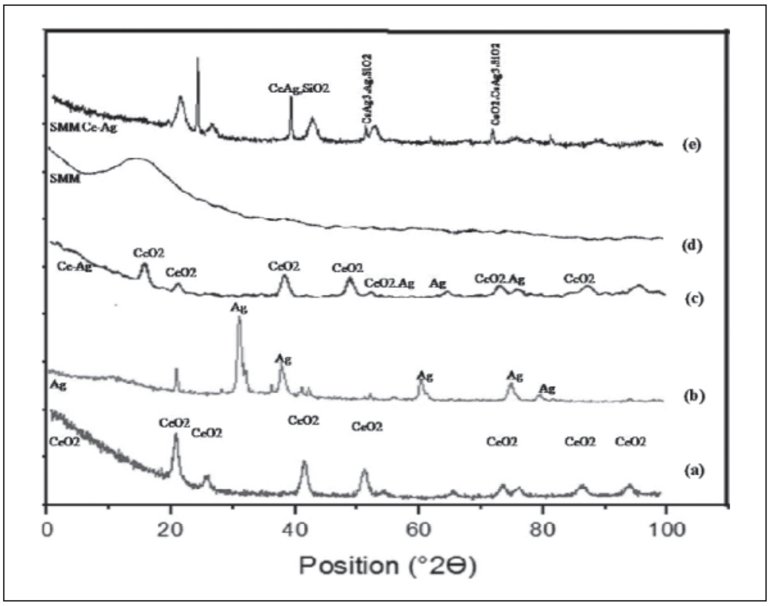


Fig. 1. XRD patterns of (a) cerium oxide nanoparticles (b) silver nanoparticles (c) cerium-silver nanoparticles, (d) modified mesoporous modification and (e) cerium-silver doped modified mesoporous modification.

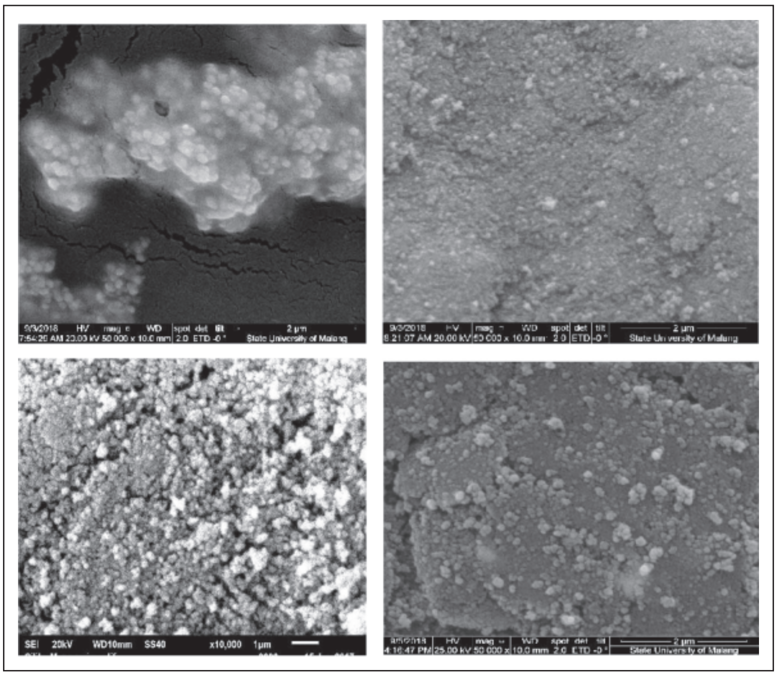
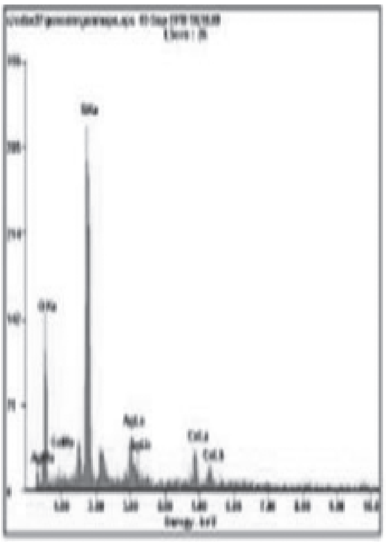


Fig. 2. SEM images of (a) cerium oxide nanoparticles (b) silver nanoparticles (c) modified mesoporous modification and (d) cerium-silver doped modified mesoporous modification.

silica nanoparticle (Rhim *et al*., 2013; Tian *et al*., 2013).

EDX analysis

EDX analysis explains the elemental composition of the product of material. Figure 3 showed Si elements as much as 33.13, 37.03% O elements, 12.09% Ag elements, and 17.75% Ce. The most elemental composition was oxygen which was assumption came from silica oxide, cerium oxide, and silver oxide. The function of silica as a surface area for silver and cerium. The composition of Ag and Ce on the surface of modified mesoporous silica will strengthen the nanofiller properties produced



Element	Wt%	At%
OK	37.03	62.00
SiK	33.15	31.61
AgL	12.09	03.00
CeL	17.73	03.39
Matrix	Correction	ZAF

Fig. 3. EDX analysis of cerium-silver doped modified mesoporous modification.

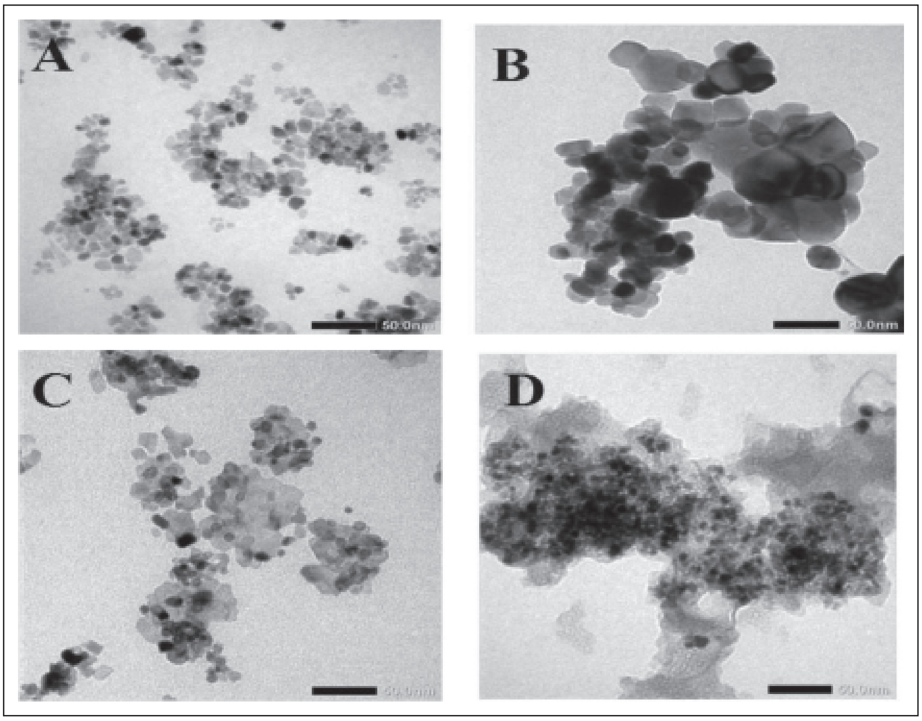


Fig. 4. TEM images of (a) cerium oxide nanoparticles (b) silver nanoparticles (c) cerium-silver nanoparticles and (d) cerium-silver doped modified mesoporous modification.

because Ag has antimicrobial properties and Cerium has the property of oxygen scavenging (Bing *et al*., 2012).

TEM analysis

Figure 4 explains the TEM results of cerium nanoparticles (a) silver nanoparticle (b) silver nanoparticles (c) silver-cerium nanoparticles (d), and cerium-silver modified mesoporous silica. Figures 4a and 4b showed silver nanoparticles and cerium appear to be round with a uniform size. Figure 4d was Ce-Ag-doped mesopore silica modification which shows a smaller size when compared to Cerium and silver nanoparticles. The measurement results with XRD characterization using the Debye Scherrer equation to calculate the crystal size. This

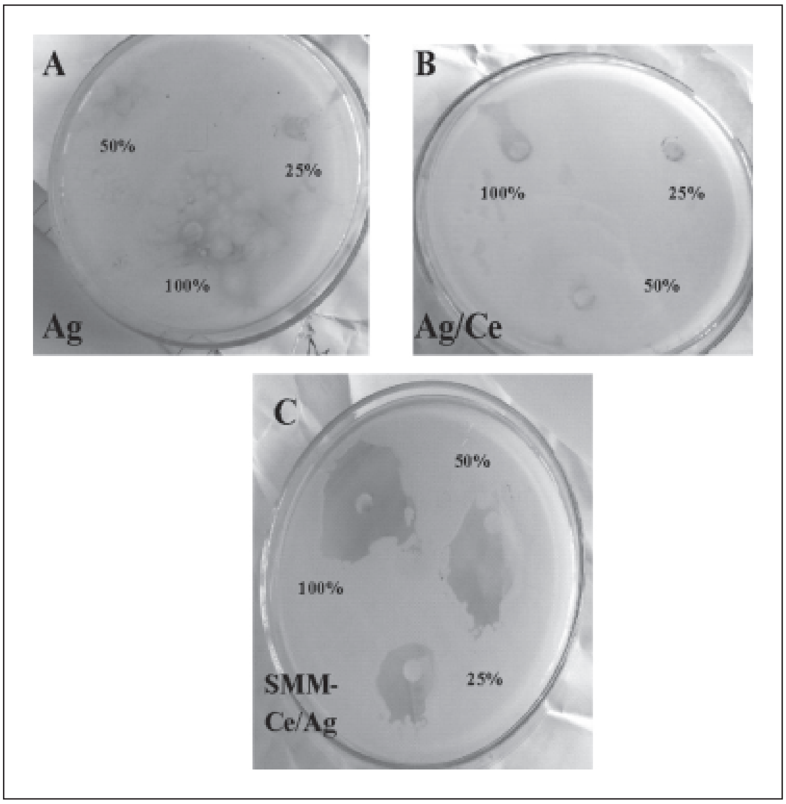


Fig. 5. Antibacterial activity of cerium-silver doped modified mesoporous modification nanoparticles against the growth of *Staphylococcus aureus* bacteria in (a) silver nanoparticles (b) cerium-silver nanoparticles and (c) cerium-silver doped modified mesoporous modification.

result was confirmed and strengthened by calculating the particle size of the material produced by using the J software image. The results of evaporation with the image of J software particle size of silver cerium doped modified mesoporous silica nanoparticles are ranging from 3.4 – 4.5 nm.

Antibacterial activity of cerium-silver doped modified mesoporous modification

Application of nanofiller Ce-Ag-MMS for antimicrobial activity was tested. Ce-Ag-MMS was tested against *Staphylococcus aureus* in 25, 50 and 100% dilutions.

Figure 5 showed silver nanoparticle colloid solution (a), silver-cerium colloid solution (b) and Colloidal Ce-Ag- MMS solution (c) which functions for inhibitory activity against *Staphylococcus aureus* bacteria with the formation zones of inhibition. The zone inhibition formed to explain the strength of the inhibitory power of nanoparticles against bacteria (Bahri *et al*., 2019; Zulkifli *et al.*, 2019). The wider the zone of inhibition generated shows, the stronger the inhibitory power of the compound to bacterial growth (Li *et al*., 2010; Sabri *et al.*, 2018). The width zone of inhibition formed using colloidal Ce-Ag- MMS solution.

The resulting zone of inhibition was measured using callipers. The results of the measurement zone of inhibition colloidal Ce-Ag- MMS showed from Table 1. Colloid Ce-Ag- MMS solution can inhibit the growth of *Staphylococcus aureus* bacteria stronger than the inhibitory solution of silver nanoparticles and silver/cerium nanoparticles. Ag nanoparticle colloid (100%) 12.5 inhibition and Silver/Cerium nanoparticle colloid (100%) inhibitory power 17.34. Silver nanoparticles doped with Cerium nanoparticles showed an increase in inhibition of 25% with an increase in the area of zones of inhibition after doping with cerium nanoparticles as in (Putri *et al*., 2018b). Silver-

Table 1. The zone of inhibition used Ag-Ce-MMS nanoparticles against growth of *Staphylococcus aureus*

No.	Concentration colloid Ce-Ag-MMS solution (%)	Zone of inhibition (mm) *Staphylococcus aureus*
1	Control	0
2	25	13.95
3	50	23.3
4	100	32.40

Table 2. Example of Nanofiller had used in food packaging

Classification	Type of nanofiller	Benefits of nanofiller	References
Organic	Clay	Cost effective, increase biodegradation and safe as food packaging materials	Rhim *et al.*, 2011
	Chitosan	Antimicrobial properties	Martelli *et al.*, 2013
	Cellulosa	Biocompatibility, barrier properties, attractive appearance, non-toxicity and low cost	George *et al.*, 2014
Inorganic (Metal)	Silver	Antimicrobial properties	Martelli *et al.*, 2013
	Copper	Antimicrobial properties	Conte *et al.*, 2013
Inorganic (Metal oxide)	ZnO	Antimicrobial properties	Kanmani *et al.*, 2014
	TiO_2	Photocatalytic agents for disinfection ability for bacteria	Zhu *et al.*,2012
	MgO	Stable, low volatile at high temperature and Antimicrobial properties	Sanuja *et al.*, 2014
	CeO_2	Oxygen scavenger properties under UV-Vis light radiation	Putri *et al.*, 2018a

cerium nanoparticles doped with modified mesopore silica showed an increased in inhibition of 80% (32.4 mm)(Table 1) when compared with silver-cerium nanoparticles zone of inhibition (17.34 mm) (Putri *et al*., 2018b). It can be concluded that the ability to inhibit bacterial *S. aureus* growth Silver-cerium nanoparticles doped with modified mesopore silica was stronger than Ce-Ag nanoparticles and Ag nanoparticles.

Synthesis nanofiller using organic and inorganic material reported in the kinds of literature shown in Table 2 and the results of this research were found to be comparable to this report. Accordingly, the combination of nanofiller cerium- silver doped mesoporous silica was great and potentially used for applications in food packaging. However, the advantages of this nanofiller from the economic aspect bioplastic used for active packaging need further economic assessment which will be discussed in the next article.

CONCLUSION

Application of nanofiller nanomaterial oxide cerium oxide/silver doped modified mesoporous silica was very potential to be developed into nanocomposite with enhanced properties and performance of materials. Silica nanofiller can be improved mechanical, thermal, and barry properties, cerium has the property of oxygen scavenger and silver has antimicrobial properties. The combination of the three nanofillers will produce a nanofiller that has superior properties. Nanofiller Ce/Ag-doped mesopore silica modification was successfully synthesized by obtaining crystal size from the XRD results of 9.8 – 12.44 nm. Nanofiber particle size produced from TEM analysis results is between 3.4 – 4.5 nm. From the results of the SEM analysis, it was showed that nanofillers are spherical in a uniform size. The antimicrobial ability of nanofiller Ce/Ag modified mesoporous silica also increased by 86.85% compared to nanofiller Ce/Ag, which was shown from an increased zone of inhibition from 17.34 mm (Ce/Ag) to 25.95 nm (Ce-Ag-MMS). Further studies are crucial to investigate the effect of adding biopolymers such as PLA, PVA with nano-filler Ce-Ag -MMS to forming bio-nanocomposite can function and applications in food packaging. Economic aspect bioplastic used for active packaging needs further economic assessment which will be discussed in the next research.

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