ANALYSIS OF STRUCTURE AND ANTIMICROBIAL ACTIVITY OF CeO$_2$ AND Nd$_2$O$_3$ NANOPARTICLES

Hanif Yuliani$^{1,*}$, Rina Dewi Mayasari$^1$, Eryanti Kalembang$^1$, Yelvia Deni$^1$, Defi Rahma Santi$^2$, Putri Widya Pangestika$^2$, Setyo Purwanto$^3$, Bambang Sugeng$^4$, Suyanti$^4$, Moch. Setyadji$^4$

$^1$Pusat Teknologi Material, Badan Pengkajian dan Penerapan Teknologi, Gedung 224, Puspiptek, Tangerang Selatan, Banten 15314, Indonesia
$^2$Program Studi Fisika, Institut Teknologi Kalimantan, Jl. Soekarno-Hatta Km 15, Balikpapan, Kalimantan Timur 76127, Indonesia
$^3$Pusat Sains dan Teknologi Bahan Maju, Badan Tenaga Nuklir Nasional, Gedung 42, Puspiptek, Tangerang Selatan, Banten 15314, Indonesia
$^4$Pusat Sains dan Teknologi Akselerator, Badan Tenaga Nuklir Nasional, Jl. Babarsari, Yogyakarta 55281, Indonesia

*Corresponding Author Email: hanif.yuliani@bppt.go.id

ABSTRACT

Cerium oxide (CeO$_2$) and Neodymium oxide (Nd$_2$O$_3$) nanoparticles using local content have been synthesized by precipitation method. The CeO$_2$ and Nd$_2$O$_3$ nanoparticles were characterized by X-Ray Diffraction (XRD) and Fourier Transform Infrared (FTIR) to analyze the material phase and structure. The XRD spectrum shows that CeO$_2$ and Nd$_2$O$_3$ nanoparticles have face-centered cubic and hexagonal, and cubic, respectively. The anti-microbial activity of CeO$_2$ and Nd$_2$O$_3$ nanoparticles was analyzed by diffusion method using gram-negative bacteria (E. coli, S. aureus, P. aeruginosa), and gram-positive bacteria (S. entericatyphi, L. monocyogenes), and fungus (C. albicans). The result confirms that CeO$_2$ and Nd$_2$O$_3$ nanoparticles have the capability of microbial pathogen inhibition. The CeO$_2$ nanoparticles have the effective activities of inhibition for the microbial of S. aureus and S. entericatyphi, whereas Nd$_2$O$_3$ nanoparticles can inhibit the microbial of P. aeruginosa, S. entericatyphi, and L. monocyogenes.

Keywords: CeO$_2$, Nd$_2$O$_3$, face-centered cubic, hexagonal, antimicrobe
INTRODUCTION

Nowadays, nanotechnology, primarily nanomaterials research and development, has been widely used for various applications in daily life, such as medicine and food [1], sensors [2], energy [3], and others. In the medicine application, the nanomaterials developed for antibiotic or antifungal applications, mainly based on rare earth elements (REE) as the raw materials [4]. Indonesia has a lot of natural resources of cerium oxide (CeO$_2$) and neodymium oxide (Nd$_2$O$_3$) classified in REE [5]. The CeO$_2$ and Nd$_2$O$_3$ have great potential to produce new various antibiotics [6-8].

Several previous studies have reported anti-microbial activity on REE nanomaterials [6-7,9]. Parvathya and Venkatramanb have investigated the differences in the synthesis methods due to the antimicrobial activity, i.e., green synthesis (G-CeO$_2$) and chemical synthesis (C-CeO$_2$). The results showed that G-CeO$_2$ nanoparticles had higher activity than C-CeO$_2$ against Escherichia coli, Pseudomonas aeruginosa, Streptococcus pneumonia, and Proteus Vulgaris bacteria [7]. The CeO$_2$ nanoparticles also obtain a good antibacterial activity towards both gram-negative and positive bacteria because it has Ce$^{3+}$ ions and rich surface oxygen vacancies [8].

In this paper, we reported the materials properties and anti-microbial activities of CeO$_2$ and Nd$_2$O$_3$ nanoparticles. Anti-microbial activities of the nanoparticles were tested with six types of microbes, i.e., Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, Salmonella entericatyphi, Listeria monocytogenes, and Candida albicans. The results of this research are the preliminary study of REE research for antibiotic applications which are expected to be new potential antibiotics.

METHOD

Synthesis of CeO$_2$ and Nd$_2$O$_3$ Nanoparticles

The CeO$_2$ and Nd$_2$O$_3$ nanoparticles have been synthesized in the Laboratory of Center of Technology for Material BPPT by precipitation method using carbonate (NaHCO$_3$) and hydroxide (NH$_4$OH) precursors. Synthesis of nano-CeO$_2$ was carried out using 0.03 M cerium nitrate hexahydrate (Ce(NO$_3$)$_3$.6H$_2$O), 0.02 M NH$_4$OH, and 0.03 M NaHCO$_3$. Those solutions were mixed at the temperature of 55°C for 15 minutes and followed by drying at the temperature of 220°C for 2 hours and calcination at the temperature of 600°C for 3 hours [10]. Synthesis of nano-Nd$_2$O$_3$ used the same synthesis process as nano-CeO$_2$ with neodymium nitrate hexahydrate (Nd(NO$_3$)$_3$.6H$_2$O) precursor.

Material Characterization and Anti-microbial Test

The material structures were characterized by x-Ray diffraction (XRD) Rigaku and Fourier Transform Infra-red (FTIR) Thermo Scientific Nicolet iS50. The anti-microbial activities of the samples were analyzed in the Laboratory of Microbiology, Center of Technology for Pharmaceutical and Medical BPPT, using well diffusion method against and six pathogen microbes (Candida albicans, Staphylococcus aureus, Listeria monocytogenes, Salmonella
enterica typhi, Escherichia coli, Pseudomonas aeruginosa) from the collection of the Inter-
University Research Center (PAU) ITB. Kloramfenicol antibiotic was used simultaneously for
positive control.

RESULT AND DISCUSSION

Crystal Structure of CeO$_2$ and Nd$_2$O$_3$ Nanoparticles

FIGURE 1 shows the X-ray diffraction (XRD) pattern of the samples of CeO$_2$ and Nd$_2$O$_3$
nanoparticles. Analysis of the XRD profile for nano-CeO$_2$ (bottom) using Match and Rietveld
program reveals that the samples formed 58.5% CeO$_2$ phase (ICDD 98-002-8753) with face
center cubic structure and space group of Fm$\overline{3}$m (225). Besides, the formed minor phase is
41.5% thermonatrite compound (Na$_2$CO$_3$·H$_2$O) with an orthorhombic structure and space
group of Pca21 (29).

![XRD spectrum of CeO$_2$ and Nd$_2$O$_3$ Nanoparticles](image)

FIGURE 1. The XRD spectrum of CeO$_2$ and Nd$_2$O$_3$ Nanoparticles.

The data analysis of the XRD pattern for nano-Nd$_2$O$_3$ (top) indicates 2 (two) Nd$_2$O$_3$ phases,
i.e., 39.6% Nd$_2$O$_3$ with cubic structure (ICDD 98-064-5664) and space group of Ia$\overline{3}$ (206),
and 25.4% Nd$_2$O$_3$ with hexagonal structure and space group of P63/mmc (194) (ICDD 98-003-2514).
The residues are the impurities of 11.9% nitrate (NaNO$_3$) phase with a space group of
R$\overline{3}$c (167) (ICDD 98-006-4868) and 23.1% neodymium hydroxide with a space group of
P63/m (176) (ICDD 98-000-0398).
Functional Group Analysis of CeO\textsubscript{2} and Nd\textsubscript{2}O\textsubscript{3} Nanoparticles

FIGURE 2 shows the result of FTIR analysis for both CeO\textsubscript{2} and Nd\textsubscript{2}O\textsubscript{3} nanoparticles. The representation of FTIR absorbance peaks is summarized in TABLE 1. The FTIR result of nano-CeO\textsubscript{2} confirmed the XRD result in which the contained impurity is thermonatrite (Na\textsubscript{2}CO\textsubscript{3}.H\textsubscript{2}O) compound at a wavenumber of 1107.61 cm\textsuperscript{-1}, 1429.28 cm\textsuperscript{-1}, 2360.79 cm\textsuperscript{-1}, and 2978.71 cm\textsuperscript{-1} which represent the vibrational bond of C–O, C=O, and O–H, respectively. The CeO\textsubscript{2} compounds were detected at the wavenumber of 549.08 cm\textsuperscript{-1}, 616.97 cm\textsuperscript{-1}, and 864.20 cm\textsuperscript{-1}. The other FTIR spectrum of CeO\textsubscript{2} detected Ce–O stretching band at 475 cm\textsuperscript{-1}, 545 cm\textsuperscript{-1}, and 615 cm\textsuperscript{-1} [11].

The FTIR result of nano-Nd\textsubscript{2}O\textsubscript{3} also validated the XRD result in which nano-Nd\textsubscript{2}O\textsubscript{3} has the same impurities, i.e., Sodium Nitrate (NaNO\textsubscript{3}) at the wavenumber of 1367.82 cm\textsuperscript{-1} and 1489.99 cm\textsuperscript{-1} which denote the vibrational bond of N–O. Furthermore, the wavenumber of 3606.11 cm\textsuperscript{-1} came from O–H bond of NdOH. Nd\textsubscript{2}O\textsubscript{3} compounds were spotted at the wavenumber of 534.29 cm\textsuperscript{-1}, 667.53 cm\textsuperscript{-1}, and 856.77 cm\textsuperscript{-1}. The similar result of the FTIR spectrum in Nd\textsubscript{2}O\textsubscript{3} have been reported by Yuvakkumar and Hong (2015) [8].

FIGURE 2. The FTIR spectrum of CeO\textsubscript{2} and Nd\textsubscript{2}O\textsubscript{3} Nanoparticles.
TABLE 1. The representation of the FTIR spectrum in the Nd$_2$O$_3$ and CeO$_2$ nanoparticles.

<table>
<thead>
<tr>
<th>Compound</th>
<th>Wavenumber (cm$^{-1}$)</th>
<th>Absorbance</th>
<th>Representation of functional group</th>
</tr>
</thead>
<tbody>
<tr>
<td>CeO$_2$</td>
<td>549.08</td>
<td>Ce – O</td>
<td>CeO$_2$</td>
</tr>
<tr>
<td></td>
<td>616.97</td>
<td>Ce – O</td>
<td>CeO$_2$</td>
</tr>
<tr>
<td></td>
<td>864.20</td>
<td>Ce – O</td>
<td>CeO$_2$</td>
</tr>
<tr>
<td></td>
<td>1107.61</td>
<td>C – O</td>
<td>Na$_2$CO$_3$·H$_2$O</td>
</tr>
<tr>
<td></td>
<td>1429.28</td>
<td>C = O</td>
<td>Na$_2$CO$_3$·H$_2$O</td>
</tr>
<tr>
<td></td>
<td>2360.79</td>
<td>O – H</td>
<td>Na$_2$CO$_3$·H$_2$O</td>
</tr>
<tr>
<td></td>
<td>2978.71</td>
<td>O – H</td>
<td>Na$_2$CO$_3$·H$_2$O</td>
</tr>
<tr>
<td>Nd$_2$O$_3$</td>
<td>534.9</td>
<td>Nd – O</td>
<td>Nd$_2$O$_3$</td>
</tr>
<tr>
<td></td>
<td>667.53</td>
<td>Nd – O</td>
<td>Nd$_2$O$_3$</td>
</tr>
<tr>
<td></td>
<td>856.77</td>
<td>Nd – O</td>
<td>Nd$_2$O$_3$</td>
</tr>
<tr>
<td></td>
<td>1489.99</td>
<td>N – O</td>
<td>NaNO$_3$</td>
</tr>
<tr>
<td></td>
<td>1367.82</td>
<td>N – O</td>
<td>NaNO$_3$</td>
</tr>
<tr>
<td></td>
<td>3606.11</td>
<td>O – H</td>
<td>NdOH</td>
</tr>
</tbody>
</table>

Anti-Microbial Activities of CeO$_2$ and Nd$_2$O$_3$ Nanoparticles

The results of the anti-microbial activity test of nano-CeO$_2$, nano-Nd$_2$O$_3$, and positive control were summarized in TABLE 2. From the result analysis, nano-CeO$_2$ and nano-Nd$_2$O$_3$ have higher inhibition ability than Control + to against bacteria (Gram + and Gram -) and fungi.

TABLE 2. Anti-microbial activities of CeO$_2$ and Nd$_2$O$_3$ nanoparticles.

<table>
<thead>
<tr>
<th>Types of Microbes</th>
<th>Inhibition Zone Diameter (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control (+)</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>0.34</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>26.13</td>
</tr>
<tr>
<td>Listeria monocytogenes</td>
<td>9.99</td>
</tr>
<tr>
<td>Salmonella enterica typhi</td>
<td>4.72</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>14.54</td>
</tr>
<tr>
<td>Pseudomonas aeruginosa</td>
<td>3.58</td>
</tr>
</tbody>
</table>

Babenko et al. have reported inhibitory activity of nano-CeO$_2$ towards Candida albicans, the interaction between nano-CeO$_2$ and fungi cell surface causes the irreversible change of cell structure and generate blocking capability for fungi enzymatic activity [11].

TABLE 3 shows the review of the inhibition test of nano-CeO$_2$ and nano-Nd$_2$O$_3$ against Gram + (S. aureus). The same result also has been researched by Reddy Yadaf et al. and Malleshappa et al. [13-14]. Meanwhile, TABLE 4 shows the inhibitory activity of nano-CeO$_2$ nano-Nd$_2$O$_3$
towards Gram – (E. coli). A similar result also has been obtained by Malleshappa et al. [14]. Moreover, the inhibitory activities of nano-CeO$_2$ and nano-Nd$_2$O$_3$ against Gram – (P. aeruginosa) have been reported by Ravishankar et al. [15], which have a comparable result of TABLE 5.

### TABLE 3. Anti-microbial activities of CeO$_2$ and Nd$_2$O$_3$ nanoparticles towards gram-positive (S. aureus) bacteria.

<table>
<thead>
<tr>
<th>S. aureus strain</th>
<th>Concentration (mg/mL)</th>
<th>Test Result</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>NCIM-5022</td>
<td>10</td>
<td>1.67</td>
<td>[12]</td>
</tr>
<tr>
<td>NCIM-5022</td>
<td>10</td>
<td>3.33</td>
<td></td>
</tr>
<tr>
<td>NCIM-5022</td>
<td>10</td>
<td>0.53</td>
<td></td>
</tr>
<tr>
<td>PAU ITB</td>
<td>10</td>
<td>1.47</td>
<td></td>
</tr>
<tr>
<td>PAU ITB</td>
<td>10</td>
<td>4.00</td>
<td>Sample CeO$_2$</td>
</tr>
<tr>
<td>PAU ITB</td>
<td>10</td>
<td>1.33</td>
<td>Sample Nd$_2$O$_3$</td>
</tr>
</tbody>
</table>

### TABLE 4. Anti-microbial activities of CeO$_2$ and Nd$_2$O$_3$ nanoparticles towards gram-negative (E. coli) bacteria.

<table>
<thead>
<tr>
<th>E. coli strain</th>
<th>Concentration (mg/mL)</th>
<th>Test Result</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>NCIM-5022</td>
<td>10</td>
<td>2.67</td>
<td>[13]</td>
</tr>
<tr>
<td>NCIM-5022</td>
<td>10</td>
<td>4.67</td>
<td></td>
</tr>
<tr>
<td>PAU ITB</td>
<td>10</td>
<td>0.67</td>
<td>CeO$_2$</td>
</tr>
<tr>
<td>PAU ITB</td>
<td>10</td>
<td>3.00</td>
<td>Nd$_2$O$_3$</td>
</tr>
</tbody>
</table>

### TABLE 5. Anti-microbial activities of CeO$_2$ and Nd$_2$O$_3$ nanoparticles towards gram-negative (P. aeruginosa) bacteria.

<table>
<thead>
<tr>
<th>P. aeruginosa strain</th>
<th>Concentration (mg/mL)</th>
<th>Test Result</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>NCIM-2242</td>
<td>10</td>
<td>3.33</td>
<td>[4]</td>
</tr>
<tr>
<td>NCIM-2242</td>
<td>15</td>
<td>3.57</td>
<td></td>
</tr>
<tr>
<td>NCIM-2242</td>
<td>20</td>
<td>4.50</td>
<td></td>
</tr>
<tr>
<td>PAU ITB</td>
<td>10</td>
<td>4.28</td>
<td>CeO$_2$</td>
</tr>
<tr>
<td>PAU ITB</td>
<td>10</td>
<td>3.67</td>
<td>Nd$_2$O$_3$</td>
</tr>
</tbody>
</table>

The anti-microbial mechanism of CeO$_2$ nanoparticles has been reported by Passos Farias et al. [4], microbial inhibition activity of CeO$_2$ nanoparticles is caused by the oxidative stress of microorganism cell membrane.

**CONCLUSION**

The CeO$_2$ and Nd$_2$O$_3$ nanoparticles have a high anti-microbial activity towards pathogen microbes, i.e., gram-positive bacteria (Staphylococcus aureus, Listeria monocytogenes), gram-negative bacteria (Pseudomonas aeruginosa, Escherichia coli, Salmonella entericatyphi), and fungi (Candida albicans).
ACKNOWLEDGMENT

We acknowledge to DIPA-BPPT for funding this research and to the Laboratory of Microbiology, Center of Technology for Pharmaceutical and Medical BPPT, for providing the facilities of the anti-microbial test.

REFERENCES


