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Preparation of halloysite nanotubes coated with Eudragit for a controlled release of thyme essential oil

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ABSTRACT: Active food packaging that releases active agents can extend the shelf-life and improve the quality and safety of food products. Essential oils have been used as natural food preservatives due to their antioxidant, antimicrobial, and anti-insect properties. However, one of the limitations of using essential oils as active agents is their high volatility. In this study, thyme essential oil, an active antioxidant agent, was encapsulated into halloysite nanotubes (HNTs) using a vacuum process to sustain the release rate and to solidify the thyme oil (TO) from a liquid state. Moreover, the TO-loaded HNT capsules (TO/HNT capsules) were coated with Eudragit EPO polymer to avoid burst release and to prolong the release time. The morphology of the prepared samples was characterized using SEM, TEM, and nitrogen adsorption–desorption analysis by BET method. Zeta potential and FTIR analysis were used to verify the encapsulation of the TO and the Eudragit EPO polymer coating of TO/HNT capsules. After Eudragit EPO polymer coating of TO/HNT capsules, the surface charge of the samples was converted from -17.5 ± 0.2 mV to $+19.4 \pm 1.5$ mV. The amount of encapsulated TO was determined using a GC-FID. Encapsulation efficiency and payload of TO/HNT capsules prepared using 26.7% (w/v) TO solution were 14.94% and 14.58%, respectively. The encapsulated TO was released in a sustained manner for 96 h. In addition, antioxidant activity of the samples was evaluated using a DPPH assay and a reducing power assay. In both two assays, the antioxidant activity of the TO/HNT capsules was increased along with the increasing concentration of TO/HNT capsules. © 2015 Wiley Periodicals, Inc. J. Appl. Polym. Sci. **2015**, *132*, 42771.

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INTRODUCTION

In recent years, active food packaging received a large amount of interest. Specifically, the controlled release system of active agents has attracted attention because of its ability to allow for the transfer of the active agent from the packaging materials to the food products. The most commonly used antioxidants in the food industry are chemically synthesized preservatives. However, as the consumer demand for the use of natural additives has steadily increased, essential oils have become popular alternative compounds for extending the shelf-life of fresh and processed food.[1,2] Most essential oils are known to have antimicrobial and antioxidant properties and are classified as generally recognized as safe (GRAS) by the U.S. Food and Drug Administration.[3–5] Thyme essential oil (TO), which is extracted from *Thymus vulgaris* L., is used in medical and pharmaceutical industries as well as in flavor and food industries. The major component of TO, thymol(2-isopropyl-5-methylphenol), which is a natural monoterpene phenol, has been extensively studied for food packaging applications due to its strong antimicrobial, antioxidant, and anti-insect properties.[6–10] Like most other essential oils, however, the use of TO as an active agent is limited due to its high volatility and liquid state at room temperature. However, encapsulation technology may potentially be used to reduce the volatilization of essential oils. Additionally, it is also possible to use encapsulation to protect oils from environmental factors.

The halloysite nanotube (HNT) is a two-layered aluminosilicate clay mineral that has the ideal formula of $Al_2Si_2O_5(OH)_4 \cdot nH_2O$, which is chemically similar to kaolinite. HNT is a member of kaolin group of clay minerals which are classified as GRAS for food packaging. HNTs are naturally abundant in countries such as China, USA, Brazil, and Korea. The HNT mostly displays a hollow tubular morphology formed by the rolling of layers. The length of an HNT is approximately 1 μm, with an inner diameter of 10–30 nm and an outer diameter of 50–70 nm. This tubular structure provides HNTs with a high surface area. HNTs have silicon dioxide external and aluminum dioxide internal surfaces. This structure results in a negatively charged outer surface and a positively charged inner lumen surface in a broad pH range (pH 2–8).[11–15] Due to their nanoporous structure,

HNTs have multiple applications for nanotechnology, such as nanoscale reactors for biomineralization, adsorbents for pollutants, and additives for polymer nanocomposites.[16–20] Furthermore, HNTs have attracted attention for their potential as a new type of nanocontainer for encapsulating various active agents such as drugs, marine biocides, antifouling paint, cosmetics, and other functional agents.[21–25] For example, Wei *et al.*[22] investigated the sustained release of antiseptics from HNTs. The release rate of antiseptics from HNTs was significantly more sustained when compared to pure antiseptics in water. However, it is not yet studied to encapsulate volatile compound into the HNTs and to investigate aerial release profile of the active agents from HNTs and polymer coated HNTs.

The aim of this work was to investigate the controlled release profile of TO incorporated with HNTs. For this purpose, TO-loaded HNT capsules (TO/HNT capsules) were prepared using a vacuum process. The TO/HNT capsules were coated with Eudragit EPO polymer (EPO) to sustain the release rate of TO and to solve the initial burst release problem. Since EPO is a positively charged polymer, it combines with the surface of the HNTs. We also evaluated the antioxidant activity of the TO/HNT capsules using a 1,1-di-phenyl-2-picrylhydrazyl (DPPH) radical scavenging assay and a reducing power assay.

MATERIALS AND METHODS

Materials

The HNTs used in this study were purchased from Sigma-Aldrich (St. Louis, MO). This clay had a specific gravity of 2.53 g/cm^3 and a pore volume of 1.26–1.34 mL/g. TO was purchased from Dotter-Line (Gunpo, Korea). Eudragit EPO (EPO) was purchased from Evonik Industries AG (Darmastadt, Germany) and used for HNT coating. The DPPH was from Sigma-Aldrich. Potassium ferricyanide was obtained from Kishida Chemical (Osaka, Japan). Trichloacetic acid was from BDH Laboratory Supplies (Poole, U.K.), and ferric chloride was from Kanto Chemical (Tokyo, Japan). All other chemicals used in this study were of analytical grade and used as received.

Preparation of TO/HNT Capsules

TO was loaded into HNTs according to the procedure described by Price *et al.*[26] with slight modifications. In brief, different amounts of TO (2 g, 3 g, 4 g, 5 g, and 6 g) were added to 15 mL of absolute ethyl alcohol and vortexed for 60 s. Final concentrations of each TO solution were 13.3%, 20.0%, 26.7%, 33.3%, and 40.0% w/v. Then, 4 g of HNTs was thoroughly dispersed into the solution with an ultrasonic homogenizer (VCX 750, Sonics & Materials, Newtown, CT) for 10 min to form a suspension. The homogenous suspension was kept in a vacuum chamber at 30°C. After 30 min, the vacuum was stopped and air was allowed into the chamber for 30 min. This process was repeated three times to increase the encapsulation efficiency (EE). The air in the lumen of HNTs was replaced with TO solution through the vacuum process. Next, TO/HNT capsules were separated from the solution by centrifugation using a high-speed centrifuge (Himac CR-21G Centrifuge, Hitachi, Japan) at 7000 rpm for 10 min and then washed with ethanol to remove any unloaded TO. Finally, TO/HNT capsules were dried overnight in a vacuum oven (VO-20X, JeioTech, Korea) at 40°C.

Preparation of Eudragit EPO-Coated TO/HNT Capsules (TO/HNT/EPO Capsules)

Throughout the coating process, the concentration of TO solution was maintained at 26.7% (w/v), because payload (PL) of TO was higher at this concentration than all other groups. Increasing the concentration of TO solution higher than 26.7% (w/v) would not significantly increase the PL. HNTs were thoroughly dispersed in the solution using an ultrasonic homogenizer for 10 min. The vacuum process also followed the above procedure. Next, 2 mL of EPO solution in ethanol was added to the suspension and then mixed using a vortex for 5 min. The EPO concentration in the suspension was 2% (w/v). TO/HNT/EPO capsules were separated, washed, and dried following the above procedure.

Scanning Electron Microscopy

The surface morphology of the pristine HNTs, TO/HNT capsules, and TO/HNT/EPO capsules was examined through field emission scanning electron microscopy (FE-SEM; Hitachi S4300, Hitachi, Japan) at an accelerating voltage of 15.0 kV. Before SEM observation, the samples were fixed on the sample holder and then coated with a thin layer of platinum using an ion sputter coater (Hitachi E1030, Hitachi, Japan).

Transmission Electron Microscopy

For transmission electron microscopy (TEM; TECNAI G2 F30, Philips-FEI, Holland) observation, pristine HNTs, TO/HNT capsules, and TO/HNT/EPO capsules were immersed in distilled water and dispersed with ultrasonic waves for 10 min. An aqueous dilute suspension of sample was dropped on the copper grid and left undisturbed for 1 min. Then, excess solution was removed using filter paper. Finally, the grids were treated with 2% (w/v) phosphotungstate solution to stain the sample for about 1 min and then dried overnight.

Nitrogen Adsorption–Desorption Analysis

Nitrogen adsorption–desorption isotherm for pristine HNTs were obtained at 77 K using an ASAP 2010 micromertics porosimeter (ASAP 2010, Micromeritics). Before measurement, the pristine HNTs were dried at 110°C for 12 h. The specific surface area of pristine HNTs was determined by the Brunauer-Emmett-Teller (BET) method, and total pore volume was determined by the Barrett-Joyner-Halenda method.[27]

Zeta Potential Analysis

The surface charges of the pristine HNTs, TO/HNT capsules, and TO/HNT/EPO capsules were measured using a Nano-ZS nano size analyzer (Zetasizer Nano ZS, Malvern Instruments, UK). Briefly, 1–2 mg of each sample was suspended in 10 mL distilled water (pH 5.9), then mixed using a vortex for a few minutes. About 1 mL of the dispersion was added to a polystyrene latex cell, and the measurements were carried out at room temperature with a detector angle of 90°. Each sample was measured in triplicate to obtain the average values.

Fourier Transform Infrared Spectroscopy

Pristine HNTs, TO/HNT capsules, and TO/HNT/EPO capsules were also analyzed using the Fourier-transform infrared (FTIR; Varian 640-IR, Agilent Technologies, CA) at 25°C. Each sample was prepared using the potassium bromide (KBr) pellets

method. Briefly, each sample was ground and mixed with KBr, and prepared as a pellet by compression under a force of 10 tons in a hydraulic press. All FTIR spectra were obtained by 32 scans from wavelengths 400 to 4000 cm^{-1} with a resolution of 2 cm^{-1}.

Encapsulation Efficiency and Payload

TO loading was determined by dispersing accurately weighed amounts of sample (300 mg) in 10 mL of absolute ethyl alcohol. Then, the samples were broken using an ultrasonic homogenizer for 30 min. After centrifugation was carried out at 7000 rpm for 10 min to separate broken HNTs out of suspension, the supernatant was filtrated through a 0.2-μm membrane (Whatman), and the amount of TO was determined in triplicate for each sample using GC-FID. The GC-FID apparatus was an HP-6980 Agilent gas chromatograph equipped with a flame ionization detector (FID) with an SPB-608TM capillary column (30 mm × 0.25 mm ID, 0.25 μm film thickness). The GC temperature was programmed from 55 to 65°C, at 1°C/min (3 min hold); to 180°C, at 3°C/min (5 min hold); and finally up to 280°C, at 20°C/min (3 min hold). Quantification of TO was carried out using a calibration curve. EE and PL were calculated using the following equations:

$$\text{EE}\,(\%) = \frac{M_{\text{th}}}{M_0} \times 100$$

where M_{th} is the weight of the TO in the samples and M_0 is the initial weight of TO added.

$$\text{PL}\,(\%) = \frac{M_{\text{th}}}{M_s} \times 100$$

where M_{th} is the weight of the TO in the samples and M_s is the weight of the samples.

Release Studies

Release studies were performed in an oven at 25°C. Accurately weighed quantities of sample (300 mg) were incubated at 25°C oven. At specific time intervals (0, 1.5, 3, 6, 12, 24, 36, 48, 72, and 96 h), each sample was added to 10 mL of absolute ethyl alcohol to determine the amount of TO remaining after evaporation. The amount of TO was measured using the method described above. To investigate the release profile of TO in liquid state for control, the same amount (mg) of TO was incubated at 25°C and then added to 10 mL of absolute ethyl alcohol at same time intervals. Each experiment was repeated three times to obtain the average values. The percentage of TO released at each time point was calculated using the following equation:

$$\text{Thyme oil release}\,(\%) = \frac{M_i - M_t}{M_i} \times 100$$

where M_i is the initial amount of TO in the sample and M_t is the amount of TO in the sample at specific time.

Measurement of Free Radical Scavenging Activity

The free radical scavenging activity was investigated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. The DPPH assay was based on the methodology of Yamaguchi *et al.*[28] with slight modifications. Briefly, varying concentrations of TO/HNT capsules were added to 5 mL of ethanol and then broken using an ultrasonic homogenizer for 30 min. Next, the broken HNTs were separated from the suspension by centrifugation, and the resulting supernatant was used for the DPPH assay. To 2 mL of DPPH solution (0.25 m*M*, in ethanol), 2 mL of the supernatant was added. The mixture was shaken and incubated for 30 min in the dark at room temperature. Ethanol was used as a blank control. The resultant absorbance was recorded at 517 nm using a UV–Vis spectrophotometer (Optizen 3220UV, Mecasys, Korea). The DPPH radical scavenging activity was calculated using the following equations:

$$\text{DPPH radical scavenging activity}\,(\%) = \frac{A_b - A_s}{A_b} \times 100$$

where A_b and A_s are the absorption of the blank and sample solutions, respectively.

Measurement of Reducing Power

The reducing power of the TO/HNT capsules was determined according to the method described earlier by Yen and Chen with slight modifications.[29] Briefly, the supernatant of the samples was prepared using the method described above. Then, 2 mL of the supernatant was mixed with 2 mL of sodium phosphate buffer (0.2 *M*, pH 6.6) and 2 mL of potassium ferricyanide (1%, w/v). After incubation in a water bath at 50°C for 20 min, 2 mL of trichloroacetic acid solution (10%, w/v) was added to the reaction mixture. After centrifugation at 1000 rpm for 5 min, 2 mL of the supernatant was mixed with 2 mL of distilled water and 0.4 mL of ferric chloride solution (0.1%, w/v). The absorbance was measured at 700 nm using a UV–vis spectrophotometer. A blank sample was prepared without the TO/HNT capsules. Increased absorbance indicated an increase in reducing power.

Statistical Analysis

All values are described as mean ± SD and were calculated using the Statistical Package for the Social Sciences (SPSS, IL). Statistical data analyses were performed using one-way ANOVA for $n = 3$. $P < 0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

Encapsulation Efficiency and Payload

Different concentrations of TO solution were used to determine the optimum concentration for preparing TO/HNT capsules. As shown in Figure 1, it was found that the EEs significantly decreased from 17.20% to 10.62% as the TO concentration increased from 13.3% to 40.0% (w/v). Whereas PLs increased from 8.95% to 15.40% as the TO concentration increased. In addition, when the TO concentration was above 26.7% (w/v), the increase in PLs was less significant. Thus, the optimum concentration was 26.7% (w/v) when considering the EE and PL. At 26.7% (w/v) of TO solution, the EE and PL were 14.94% and 14.58%, respectively. When the TO/HNT capsules were prepared using a vacuum process, the air in the HNTs was replaced with the solution. Therefore, TO was loaded into the lumen of the HNTs. Loading of active agents into HNTs is based on the diffusion of molecules from the external solution into the pores of the HNTs due to the concentration gradient.[12] In previous studies,[13,14] different drugs were loaded into the HNTs with an

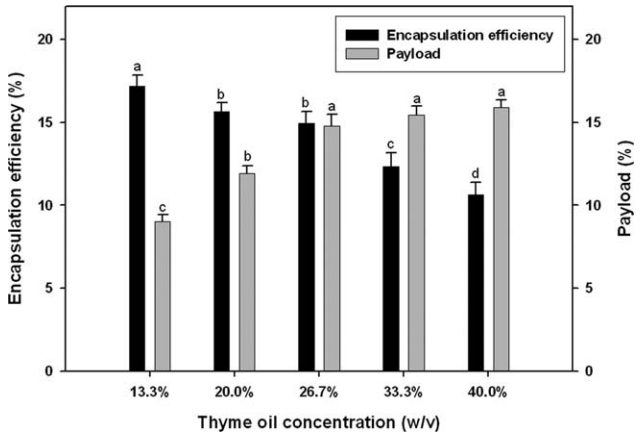

Figure 1. EE and PL of TO/HNT capsules prepared using TO solution at various concentrations. Each value is expressed as mean ± SD ($n = 3$).

efficiency of 10–15%. Similarly, the maximum PL was approximately 15% in this study.

Morphology Analysis

The surface morphology of each sample was investigated by SEM. The image of pristine HNTs is provided in Figure 2(a), showing a rod shape and indicating that HNTs have a length of approximately 1 μm. This result corresponds to those of previous studies.[11–15] Figure 2(b,c) show TO/HNT capsules and TO/HNT/EPO capsules, respectively. The TO/HNT capsules exhibited a smooth surface similar to the pristine HNTs. However, the TO/HNT/EPO capsules have a rough surface compared to that of pristine HNTs and TO/HNT capsules. This result demonstrates that TO did not affect the outer surface of the HNTs during the encapsulation process and also that EPO polymer combined with the surface of the HNTs, which have a negative charge, because EPO is a cationic polymer.

TEM was used to characterize the structural changes of the TO/HNT capsules before and after EPO polymer coating. Figure 3(a) shows the image of the TO/HNT capsule. This indicated that the HNTs have typical hollow structures with an outer diameter of 50–60 nm and a lumen diameter of 10–20 nm. Figure 3(b) shows that the outer wall of the TO/HNT capsules was coated with a EPO polymer. The outer diameter of the TO/HNT/EPO capsule was increased slightly from 50–60 nm to 75–85 nm. In addition, the TO/HNT/EPO capsule had a round shape at the end, whereas the TO/HNT capsule had a sharp end. This result was consistent with the SEM images.

The surface area and pore volume of the pristine HNTs were investigated by nitrogen adsorption and desorption analysis. As show in Figure 4, the isotherm shape of the pristine HNTs belongs to the type IV proposed by the International Union of Pure and Applied Chemistry (IUPAC), indicating mesoporosity.[30] This result was consistent with the microstructure investigated by TEM. In general, the specific surface area and pore volume of pristine HNTs reported by previous studies were 65 m^2/g and 0.08 cm^3/g, respectively.[27] In this study, the specific surface area and pore volume of pristine HNTs were calculated to 63.1 m^2/g and 0.14 cm^3/g, respectively. Therefore, the HNTs can be applied as a good container for active agent due to their structure, large specific surface area and pore volume.

Zeta Potential Analysis

The surface charges of pristine HNTs, TO/HNT capsules, and TO/HNT/EPO capsules were analyzed by measuring the zeta potentials. As shown in Table I, the surface charge of pristine HNTs was -17.8 ± 0.3 mV. This result corresponds to many previous studies.[12,13] In their natural form, the outer surface of pristine HNTs is negatively charged due to the Si—O—Si groups. Overall, the zeta potential of HNTs almost coincides with the zeta potential of silica. It is because measurement of the zeta potential is based on an electrophoretic kinetics measurement of the whole particle's movement, which is not essentially influenced by its internal charges.[15] The surface charge of the TO/HNT capsules was -17.5 ± 0.2 mV, similar to the value of pristine HNTs. It was evident that the surface of the HNTs was not affected by TO during the encapsulation process. However, the surface charge of TO/HNT/EPO capsules was converted from a negative charge to a positive charge with a value of 19.4 ± 1.5 mV. This indicated that the TO/HNT capsules were successfully enclosed within the EPO polymer. This can be attributed to the presence of dimethylaminoethyl methacrylate as a functional group of the EPO polymer. Dimethylaminoethyl methacrylate combined with the negatively charged surface of the HNTs due to electrostatic attraction. In previous research, HNTs were coated with the cationic polymer, polyethylenimine, to retard the release rate of glycerol from HNTs.[24]

FTIR Analysis

To confirm the structure of the HNTs and the existence of TO and EPO polymer, FTIR studies were conducted (Figure 5). In

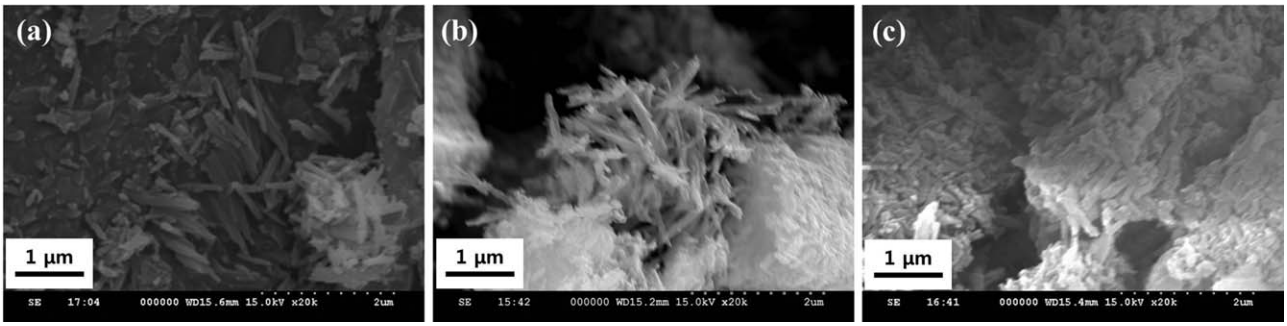

Figure 2. FE-SEM images of (a) pristine HNTs, (b) TO/HNT capsules, and (c) TO/HNT/EPO capsules. The bar is 1 μm.

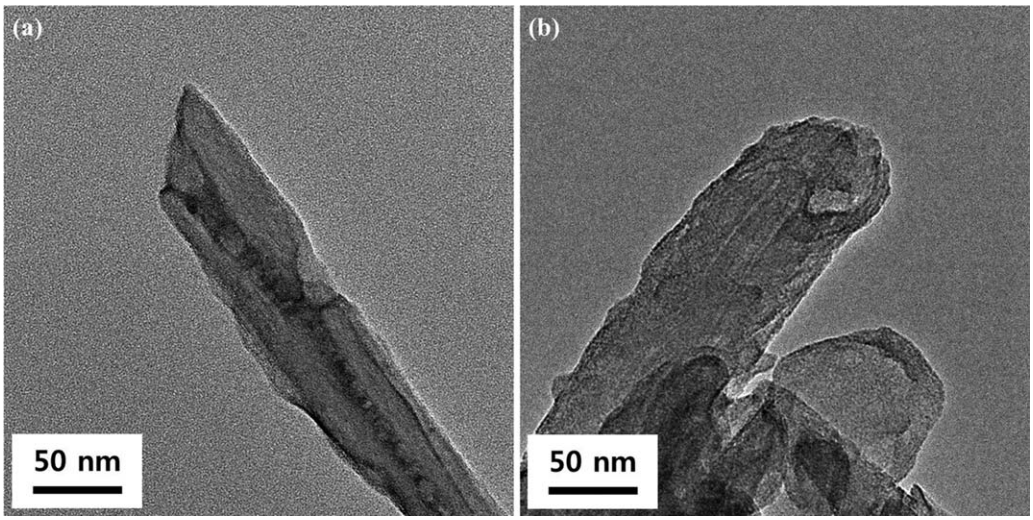

Figure 3. TEM images of (a) TO/HNT capsule and (b) TO/HNT/EPO capsule. The bar is 50 nm.

the FTIR spectrum of the pristine HNTs [Figure 5(a)], the peaks observed at 3696 and 3621 cm^{-1} are assigned to the OH stretching vibration of the inner Al—OH groups of the HNTs. The peaks at 1645, 911, and 470 cm^{-1} are assigned to the stretching vibrations of OH groups in absorbed water, the OH deformation vibration of the inner hydroxyl groups, and Si—O—Si deformation vibration, respectively. This assumption is in accordance with previous studies on HNTs.[31,32] Figure 5(b) displays the FTIR spectrum of the TO/HNT capsules. The absorption peak at 2964 cm^{-1} is assigned to the stretching vibration of the C—H bond from the CH_2 groups of the thymol, but it was absent the in case of pristine HNTs. It was evident that TO was loaded into the HNTs. Compared to the spectrum of the TO/HNT capsules, the FTIR spectrum of the TO/HNT/EPO capsules displays a new peak at 1734 cm^{-1} assigned to the C═O stretching band of the carboxylic ester of the EPO polymer, and the intensity of the peak at 2964 cm^{-1} has increased [Figure 5(c)].[33] This indicates that the TO/HNT capsules were coated with the EPO polymer. Figure 5(d) shows the FTIR spectrum of TO/HNT capsules after extracting TO. As you can see in this spectrum, a peak of TO/HNT capsules after extracting TO from the TO/HNT capsules was disappeared at 2964 cm^{-1} which is assigned to the stretching vibration of the C—H bond from the CH_2 groups of the thymol.

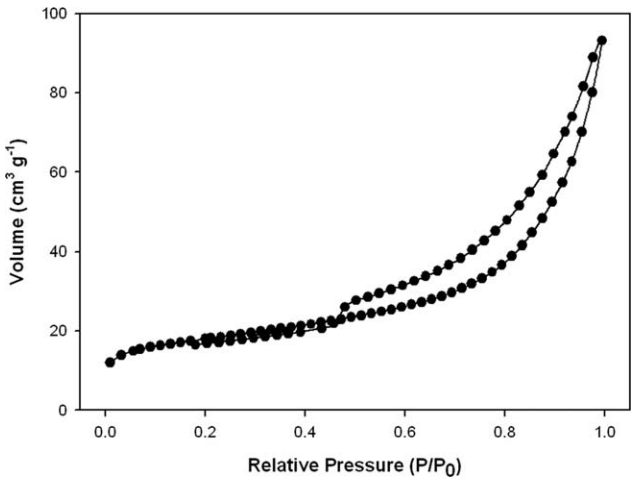

Figure 4. Adsorption–desorption isotherm of nitrogen for the pristine HNTs.

Release Studies

Volatile compounds, such as essential oils, can migrate to foodstuff without direct contact in the gaseous state. For this reason, release studies were carried out in a 25°C oven for 96 h to investigate the release behavior in the gaseous state, and the percentage of TO released is shown in Figure 6. The TO was released very quickly from the original liquid state to the air. The amount of TO released from the original liquid state was 79.73% at 12 h and was almost completely released within 24 h. However, the release rate of TO slowed down significantly after encapsulation in HNTs and was extended even more after the EPO was coated onto the TO/HNT capsules. The amount of TO released from the TO/HNT and TO/HNT/EPO capsules was 61.76% and 45.27% at 24 h, respectively. The release of TO from each capsule was sustained up to 96 h. In the case of the TO/HNT capsules, however, an initial burst release was noticed. TO/HNT capsules without a coating exhibited an initial burst release of 47.96% within 12 h, while the TO/HNT/EPO capsules showed significantly retarded TO release. Because the outer surface of the HNTs was negatively charged, the cationic groups of the EPO polymer were easily attached to the surface of HNTs via electrostatic interactions. The EPO acted as a barrier to migrating TO into air. Therefore, the EPO coating was effective at slowing down the initial release rate of TO. Coating of the HNTs with polyethyleneimine to extend the glycerol release rate

Table I. Zeta Potential of Pristine HNTs, TO/HNT Capsules, and TO/HNT/EPO Capsules Dispersed in Distilled Water at pH 5.9

Material	Zeta potential (mV)
Pristine HNTs	−17.8 ± 0.3
TO/HNT capsules	−17.5 ± 0.2
TO/HNT/EPO capsules	19.4 ± 1.5

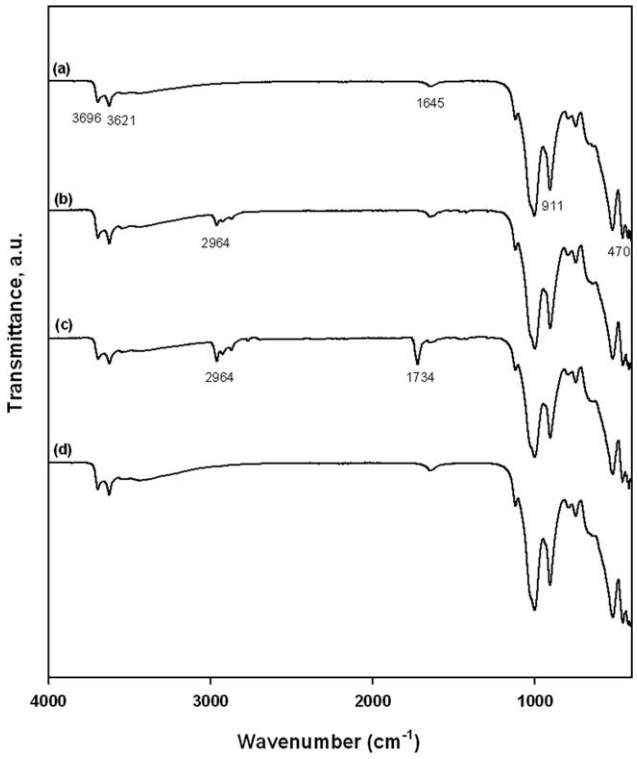

Figure 5. FTIR spectra of (a) pristine HNTs, (b) TO/HNT capsules, (c) TO/HNT/EPO capsules, and (d) TO/HNT capsules after extracting TO.

has been previously studied.[24] However, polyethyleneimine is unsuitable for food packaging because it has very strong toxicity. In contrast, the use of the EPO polymer in drug delivery systems as a pharmaceutical polymer has been widely studied.[34,35]

Antioxidant Activity of TO/HNT Capsules

DPPH radical scavenging and reducing power properties were assessed to determine whether the TO retained its antioxidant capacity during encapsulation into the HNTs. A freshly prepared DPPH solution is characterized by a deep purple color

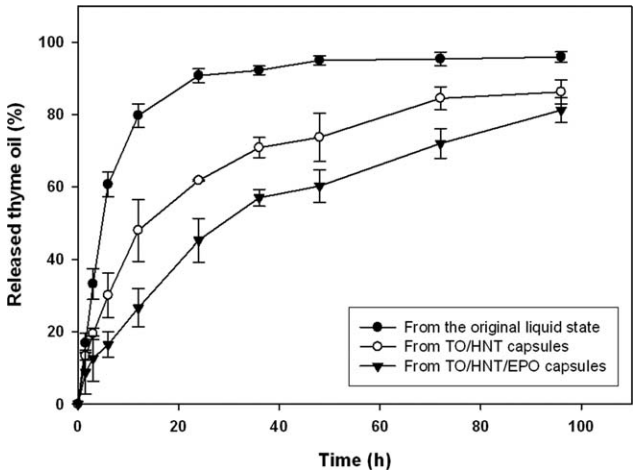

Figure 6. Cumulative release of TO from (a) the original liquid state, (b) TO/HNT capsules, and (c) TO/HNT/EPO capsules into the air in a 25°C oven. Each value is expressed as mean ± SD ($n = 3$).

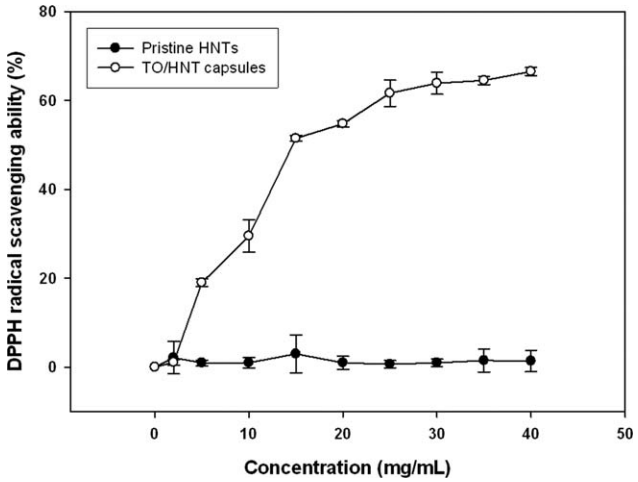

Figure 7. DPPH radical scavenging ability of pristine HNTs and TO/HNT capsules. Each value is expressed as mean ± SD ($n = 3$).

with a maximum absorption at 517 nm. The antioxidants quench DPPH free radicals and convert them into a colorless product, resulting in reduction in the absorbance at 517 nm.[36] The free-radical scavenging capacities of HNTs with and without TO are shown in Figure 7. The pristine HNTs did not exhibit any radical scavenging activity while the DPPH radical scavenging activity of the TO/HNT capsules correlated well with the increasing concentration of TO/HNT capsules.

Figure 8 shows the reducing power of pristine HNTs and TO/HNT capsules. The reducing power was determined based on the following chemical reaction: Fe(III) → Fe(II). The formed Fe(II) can react with ferric chloride to form a ferric ferrous complex that has an absorption maximum at 700 nm.[36] A higher absorbance at 700 nm indicates a stronger reducing power. The absorption of the TO/HNT capsules at 700 nm increased along with the increase in the concentration of TO/HNT capsules. This result corresponds with the results from the DPPH assay, indicating that TO was effectively encapsulated into the HNTs and that the antioxidant activity of the TO/HNT

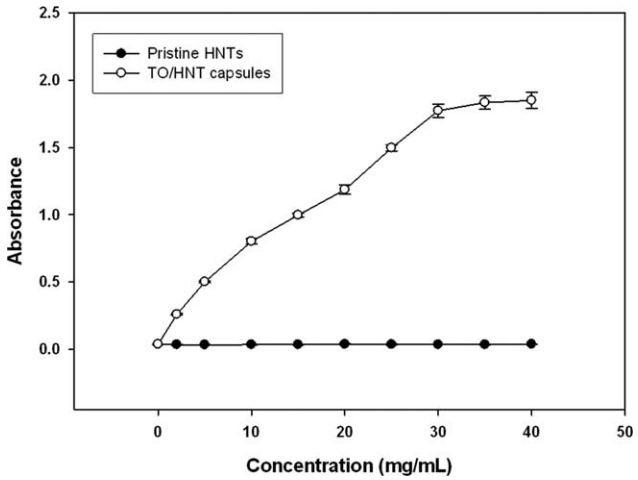

Figure 8. Reducing power ability of pristine HNTs and TO/HNT capsules. Each value is expressed as mean ± SD ($n = 3$).

capsules was pronounced. The antioxidant activity of TO, caused mainly by thymol, a natural phenolic compound, has been demonstrated in many previous studies.[7,37,38]

CONCLUSION

In this study, TO/HNT capsules were prepared using a vacuum process. The encapsulation of TO was performed with an EE of 14.94% and a PL of 14.58% at a concentration of 26.7% (w/v) of TO solution. The release of TO from the TO/HNT capsules was sustained up to 96 h. Furthermore, TO/HNT capsules were coated with EPO polymer to slow the initial burst release of TO from the HNTs. Pristine HNTs exhibited no antioxidant activity, while TO/HTN capsules exhibited excellent antioxidant activity due to the natural phenolic compound of TO. In addition, HNTs are environmental friendly and low cost materials that are very easy to handle. Therefore, HNT capsules containing essential oils have great potential for application in the food packaging industry.

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