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α-Tocopherol-Loaded Polycaprolactone (PCL) Nanoparticles as a Heat-Activated Oxygen Scavenger

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ABSTRACT: A nanoencapsulation technique was applied to an oxygen-scavenging system, and thermal processing was investigated as an activator to trigger the oxygen-scavenging reaction. α-Tocopherol-loaded polycaprolactone (PCL) nanoparticles (NPs) were prepared using an oil-in-water emulsion solvent evaporation method. The influences of iron(II) chloride, water, and thermal processing on the oxygen-scavenging capability were investigated. NPs without iron(II) chloride, moisture, and thermal processing had no oxygen-scavenging effect. However, the oxygen content (%) in the cup headspace of 20.9% decreased to 20.4% when the oxygen-scavenging system contained NPs, water, and iron(II) chloride. The oxygen content (%) decreased further to 19.5% when water was eliminated from the mixture. In this research, NPs and iron(II) chloride with thermal processing had an oxygen-scavenging capacity of 6.44 cm^3 of O_2/g and an oxygen-scavenging rate of 0.21 cm^3 of O_2 g^{-1} day^{-1}. Results indicated that NPs and iron(II) chloride in an oxygen-scavenging system can be used as a heat-activated oxygen scavenger.

KEYWORDS: oxygen scavenger, nanoparticle, α-tocopherol, thermal processing, iron(II)

INTRODUCTION

Oxygen scavengers can reduce microbial growth, the development of off-flavors, color and flavor changes, and nutritional losses in many food products by removing oxygen in package headspace. During the past 30 years, various oxygen scavengers have been investigated by the food packaging industry.[1−3] Oxygen-scavenging sachets using iron powder have been traditionally used in the packaging industry.[4] However, there are several disadvantages to the oxygen-scavenging sachet. The sachets pose a potential risk of breakage/leakage of the scavenging material and localized scavenging capability. To confront these problems, oxygen scavengers could be incorporated into food packaging materials.[5]

Unlike oxygen-scavenging sachets, oxygen-scavenging films required an activation system to prevent the initiation of the oxygen-scavenging reaction with atmospheric oxygen prior to its intended use. Many patents have been issued using ultraviolet (UV) light as an activator to trigger the oxygen-scavenging reaction.[6−9] UV-triggered oxygen-scavenging films are usually comprised of an oxygen scavenger, a transition-metal catalyst, and a photoinitiator. However, one limiting factor with this type of activation system is a significant reduction in packaging line speed. In addition, there is a substantial cost increase because of the high cost of the photoinitiator and the operation and maintenance costs of the UV machine. Therefore, the development of a new oxygen-scavenging system that does not require a UV activation step should be valuable to the food packaging industry.

Oxygen-scavenging nanoparticles could have potential as an alternative activation system. Nanoencapsulation techniques have been widely used in the medical and food industries. Encapsulated materials can be protected from moisture, heat, light, and oxygen. Several biodegradable polyesters, such as polyglycolide, polylactide, and polycaprolactone, are used as particle wall materials. Polycaprolactone (PCL) is considerably cheaper than the others and has been investigated for drug-delivery applications for several years. It is non-toxic and non-mutagenic.[10−12]

Another key issue in oxygen-scavenging technology is the use of natural compounds as the basis for the oxygen scavenger. One potential compound is α-tocopherol. It has been incorporated into the polymer materials as a stabilizer[13] and as an antioxidant in controlled release packaging materials to reduce the oxidation in food products.[14,15] Byun et al.[16] have shown that α-tocopherol and iron(II) chloride has oxygen-scavenging capability. Oxygen free radicals are produced in the presence of iron(II) chloride, and these free radicals were eliminated by receiving electrons from α-tocopherol. Therefore, iron(II) chloride and α-tocopherol can scavenge oxygen by this successive chemical reaction.

In this research, α-tocopherol-loaded PCL nanoparticles were prepared by an emulsion solvent evaporation method. Then, an oxygen-scavenging mixture including the α-tocopherol-loaded nanoparticles and iron(II) chloride were prepared. Breakage of the nanoparticles (activation of the oxygen-scavenging reaction) may be achieved by the application of heat because of the low melting point of PCL. The aim of this study was to develop a heat-triggered oxygen scavenger. The influences of water, heat, and iron(II) chloride on oxygen-scavenging capability were investigated.

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Table 1. Batch Compositions Used for Oxygen-Scavenging Mixtures[a]

code	nanoparticle (mg)	themal processing (°C)	water (μL)	iron(II) chloride (mg)
NTWI0-OS	200	77	50	
NTWI25-OS	200	77	50	25
NTWI50-OS	200	77	50	50
NTWI100-OS	200	77	50	100
NWI50-OS	200	NA	50	50
TWI50-OS	0	77	50	50
NTI50-OS	200	77	0	50
N-OS	200	NA	0	0

[a] N, α-tocopherol-loaded nanoparticle; T, thermal processing; W, water; I, iron(II) chloride; I0, 0 mg of iron(II) chloride; I25, 25 mg of iron(II) chloride; I50, 50 mg of iron(II) chloride; I100, 100 mg of iron(II) chloride.

■ MATERIALS AND METHODS

Materials. α-Tocopherol (96.2%; M_w, 430.7) was purchased from EMD Bioscience (San Diego, CA). PCL (M_n, 42 500; M_w, 65 000) was purchased from Sigma-Aldrich (St. Louis, MO). Polyvinyl alcohol (PVA) hydrolyzed 88% (M_w, 22 000) was purchased from Acros Organics (Fair Lawn, NJ). Iron(II) chloride tetrahydrate was purchased from J.T.Baker (Phillipsburg, NJ). High-performance liquid chromatography (HPLC)-grade methylene chloride (DCM) was purchased from J.T.Baker (Phillipsburg, NJ). Phosphate-buffered saline (PBS, 10× liquid concentrate) was purchased from EMD Bioscience (San Diego, CA). Retortable plastic cups with 3% EVOH barrier were provided from Printpack (Atlanta, GA). Lid materials for the cups, polyethylene terephthalate/nylon/aluminum foil/cast polypropylene (PET/foil/CPP), were provided from Fres-co System USA, Inc. (Telford, PA).

Formulation of Nanoparticles Containing α-Tocopherol. Nanoparticles were prepared using an oil-in-water emulsion solvent evaporation method together with an ultrasonification technique.[17] In this procedure, 300 mg of PCL was dissolved in 10 mL of methylene chloride containing 10 mg of α-tocopherol. A 2% PVA (w/v) solution was prepared in PBS solution. The PCL solution was added to 40 mL of the PVA solution. The total mixture was then placed in an ice bath and emulsified using a Branson Digital Sonifier (model 250, Danbury, CT) with 55 W of energy output for 3 min to obtain an oil-in-water emulsion. An additional 40 mL of the PVA solution was added to the emulsion. The final solution was stirred for 12 h on a magnetic stir plate to allow for the evaporation of methylene chloride and the formation of the nanoparticles. The suspension was then centrifuged at 4880*g* for 20 min. The pellet was resuspended in distilled water and centrifuged 3 more times at 1220*g* for 20 min each. The resulting nanoparticles were collected and frozen at −80 °C for at least 2 h and, subsequently, freeze-dried for 2 days. The freeze-dried nanoparticles were stored at 4 °C.

Scanning Electron Microscopy (SEM). The morphology of the nanoparticles was examined by SEM (S-4800 UHR FE-SEM, Hitachi High Technologies America, Inc.). Surfaces were prepared using platinum coating. SEM images were taken at 3 kV with 5000 magnification, and a 10 μm scale bar was used.

Sample Preparation. α-Tocopherol-loaded nanoparticles with differing amounts of iron(II) chloride (0, 25, 50, and 100 mg) and with or without water were placed inside a high oxygen barrier retortable cup containing 115 mL of ambient air (20.90% O_2). Then, the cup was sealed with PET/foil/CPP film with a lab sealer (Packaging Technologies, Davenport, IA). The sealing temperature was 240 °C with 40 kPa sealing pressure at a 1 s dwell time. A pilot-scale rotary, single-cage, water spray retort operating in static mode was employed in this research. After the retort reached a temperature set point of 77 °C, samples were thermally processed for 30 min at 77 °C in a Surdry model APR-95 rotary pilot retort (Stock America, Raleigh, NC). The temperature selected for this research was based on traditional "hot filling" applications.

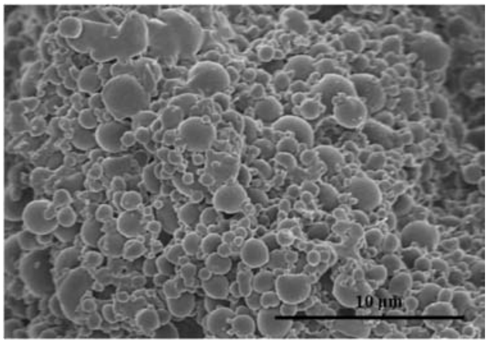


Figure 1. SEM image of the nanoparticle.

Oxygen Content Analysis. The oxygen content in the cup headspace was analyzed by a headspace oxygen/carbon dioxide analyzer (model 6600, Illinois Instrument, Johnsburg, IL). A sampling needle with a 0.45 μm polytetrafluoroethylene (PTFE) filter was inserted, and 15 mL of headspace gases was sampled through a septum. Calibration of the headspace analyzer was performed using ambient air after each sample measurement. All of the samples were measured in triplicate at days 0, 1, 4, 7, 14, and 30.

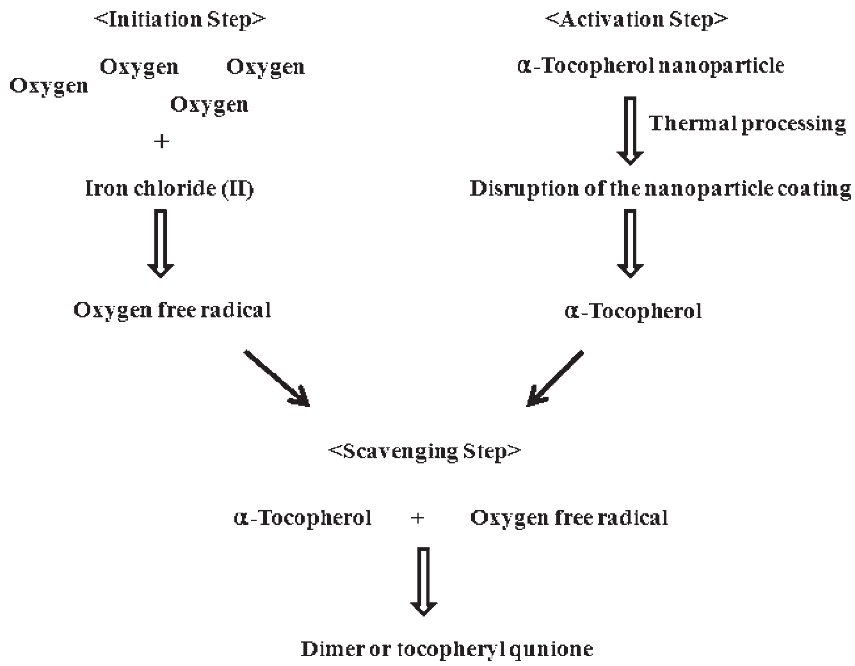
Statistical Analysis. Statistics on a completely randomized design were performed with the analysis of variance (ANOVA) using SAS (version 9.1, SAS Institute, Inc., Cary, NC), and differences among mean values were processed by Duncan's multiple range test. Significance was defined at a level of $p < 0.05$.

■ RESULTS AND DISCUSSION

Batch Composition and Characteristics of α-Tocopherol-Loaded Nanoparticles. This study was designed to assess the influence of mixture variables on the oxygen-scavenging capability of α-tocopherol-loaded nanoparticles and iron(II) chloride. Eight batches with varying compositions of scavenger mixtures were prepared (Table 1). The shape of the nanoparticles was spherical and uniform, as visualized in the SEM photographs (Figure 1). The nanoparticles had 91% encapsulation efficiency, a particle mean size of 368 nm, and a polydispersity of 0.27.[17]

Effects of α-Tocopherol-Loaded Nanoparticle, Iron(II) Chloride, Moisture, and Thermal Processing on Oxygen Scavenging. Transition metals react with oxygen and then produce oxygen free radicals.[18] However, encapsulated α-tocopherol was prevented from reacting with oxygen free radicals by

Scheme 1. Oxygen-Scavenging Reaction



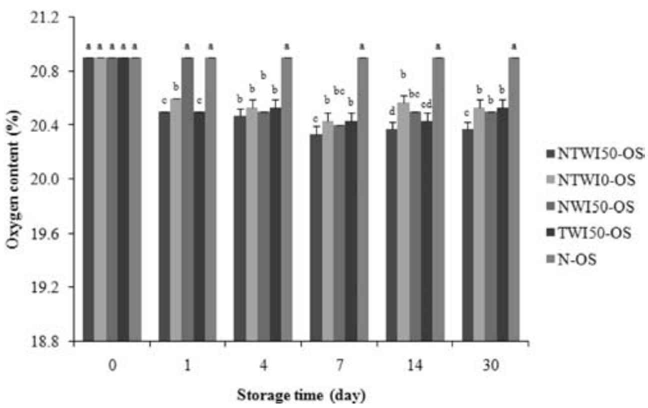


Figure 2. Influences of α-tocopherol, thermal processing, and iron(II) chloride on the oxygen-scavenging capability ($p < 0.05$).

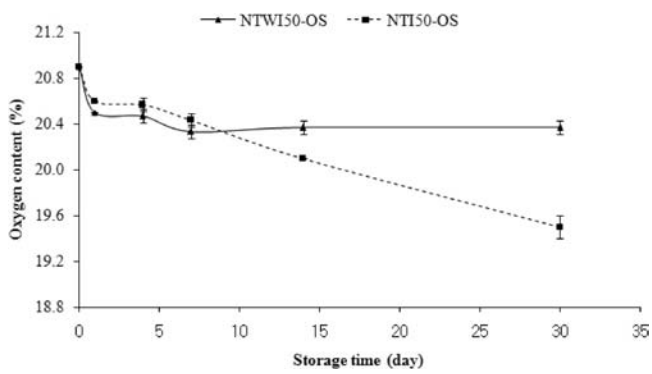


Figure 3. Influence of the moisture on the oxygen-scavenging capability.

coating the nanoparticles in this study. Without breakage of the nanoparticles, there was minimal opportunity for the oxygen free radicals to react with α-tocopherol. Disruption of the nanoparticle coating was achieved by means of thermal processing in this research (Scheme 1).

The effects of α-tocopherol-loaded nanoparticles, iron(II) chloride, and thermal processing on the oxygen-scavenging capability were investigated (Figure 2). The oxygen content (%) in the headspace of all batches, except batch N-OS, decreased during the storage time. The oxygen content (%) on batch NTWI50-OS was significantly lower than the other batch mixtures after day 14. α-Tocopherol-loaded nanoparticles with iron(II) chloride in the scavenger mixture coupled with thermal processing proved to be the most effective system for reducing the oxygen content (%) among all batch mixtures investigated. Consequently, the oxygen content in several batches was significantly higher than batch NTWI50-OS because of the fact that at least one of these three critical system components was excluded. Batch NTWI0-OS lacked iron(II) chloride. Batch NWI50-OS had no thermal processing step. Batch TWI50-OS did not include any α-tocopherol-loaded nanoparticles (Scheme 1). Therefore, α-tocopherol-loaded nanoparticles with iron(II) chloride and thermal processing provided the most effecting method to lower the overall oxygen content (%) in the container headspace. In addition, there was no reduction in the oxygen content (%) in batch N-OS throughout the entire storage time, demonstrating that α-tocopherol-loaded nanoparticles without iron(II) chloride, moisture, and thermal processing had no oxygen-scavenging effect. Therefore, the α-tocopherol-loaded nanoparticles should be stable for storage under ambient conditions prior to use.

The effects of moisture on the oxygen-scavenging capability was also investigated (Figure 3). The oxygen content (%) of batch NTI50-OS was significantly lower than that of batch NTWI50-OS after day 14. The oxygen-scavenging capability was, therefore, increased by eliminating moisture from the oxygen-scavenger mixture. However, the oxygen-scavenging

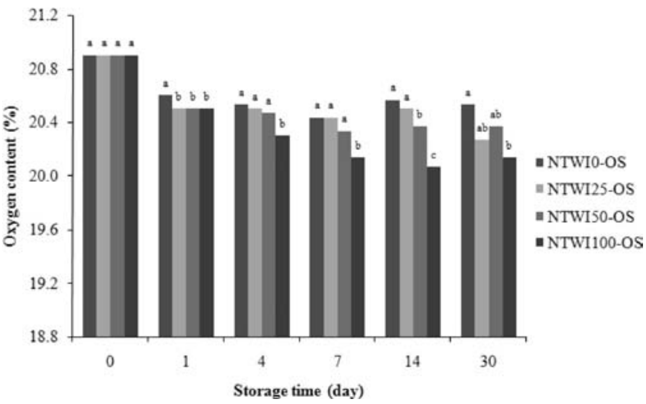


Figure 4. Influence of the amount of iron(II) chloride on the oxygen-scavenging capability ($p < 0.05$).

capability of batch NTWI50-OS was still higher than that of batch NTWI0-OS, NWI50-OS, and TWI50-OS at day 30 (Figure 2), indicating that the oxygen-scavenging mixture with moisture can still be an effective oxygen-scavenging system.

Effect of the Amount of Iron(II) Chloride. In addition, the effect of the iron(II) chloride quantity in the scavenger mixture on the oxygen content (%) reduction was investigated (Figure 4). The oxygen content in the container headspace (%) was decreased when the amount of iron(II) chloride in the scavenger mixture was increased from 0 to 150 mg during storage time. It was observed that batch NTWI100-OS containing 100 mg of iron(II) chloride showed the highest oxygen-scavenging capability. However, there was no significant difference in the oxygen content (%) between batch NTWI25-OS and NTWI50-OS at day 30.

Oxygen-Scavenging Capacity and Rate. Generally, the scavenging capacity of various scavengers has been observed to be as low as 1 cm^3 of O_2/g and with a scavenging rate of 0.1 cm^3 of O_2 g^{-1} day^{-1}.[19] The oxygen-scavenging capacity and rate for batch NTWI100-OS were calculated using the initial oxygen content at day 0 and oxygen content at day 30. α-Tocopherol-loaded nanoparticles with iron(II) chloride had a oxygen-scavenging capacity of 6.44 cm^3 of O_2/g and an oxygen-scavenging rate of 0.21 cm^3 of O_2 g^{-1} day^{-1}. These results demonstrate that the oxygen-scavenger mixture containing α-tocopherol-loaded nanoparticle and iron(II) chloride can be used as an effective heat-activated oxygen-scavenger system.

It is understood that the oxygen-scavenging capacity and rate of α-tocopherol-loaded nanoparticles with iron(II) chloride in this research were not as efficient as many commercial oxygen scavengers. However, this oxygen-scavenging system demonstrated the use of nanoencapsulation techniques in the development of a new oxygen-scavenging system not requiring a UV activation step. There are various other oxygen-scavenging compounds that could potentially be nanoencapsulated. The improvement of the oxygen-scavenging capacity using various oxygen scavengers and different encapsulation techniques will be a future work.

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