Properties analysis of crosslinked chitosan microcapsules by multiple emulsification method

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Introduction
Recently, as the quality of life has increased due to the improvement of the income level, the consumer’s desire for functional clothing is increasing day by day. In particular, to improve the comfort of clothing products, it is required to impart functionality such as antibacterial, deodorizing, temperature control, and moisture control to fabrics. However, existing functional clothing products use chemical-based finishing agents to impart functionality. Among them, synthetic antimicrobial agents are mostly irritating compounds and can be potentially harmful to the human body. Therefore, in this study, we tried to manufacture eco-friendly microcapsules that minimize the use of chemicals by using chitosan, a natural substance, as a wall material and basil oil as a core material.

Chitosan is a compound obtained by deacetylation of chitin (Muzzarelli 1977). Therefore, chitosan is a natural polymer suitable as a wall material of microcapsules because it can form a fine and dense network structure (Park & Lee 2005). In addition, due to its

Abstract
Chemical based finishing agents often affect human body and environment. Therefore, in this study, eco-friendly microcapsules were manufactured to minimize the use of chemicals by using chitosan, a natural substance, as a wall material and basil oil as a core material. First, the optimum manufacturing conditions were established through the shape and size of the synthesized microcapsules. Second, the synthesis and thermal stability of the prepared microcapsules were evaluated. Finally, the applicability to functional fiber processing was reviewed by measuring the release characteristics of the microcapsules. Consequently, using 2 wt% chitosan, Triton X-100 as a emulsifier and stirring at 6000 rpm were considered to be efficient in terms of capsule formation. FT-IR spectrum and Gas Chromatograph showed that microcapsules were stably synthesized. And microcapsule containing basil oil was given heat resistance by encapsulation. Lastly, microcapsules can be confirmed to have sustained-release, due to basil oil in microcapsules has a small amount of release up to 10 h, and is continuously released until after 60 h to release slowly. As a result, the manufactured microcapsules finishing agent may be applied to finishing of textile product.

Keywords: Encapsulation, Chitosan, Basil oil, Microcapsule

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biodegradable and environmentally friendly properties, it has been frequently used in textile processing studies (Ha et al. 1998; Han et al. 2008), and has been reported to have excellent antibacterial properties and biocompatibility to promote wound recovery (Bae et al. 2008). And also, chitosan is widely used in medicine, food, industry and agriculture because it maintains a positively charged state in a dissolved state and has excellent antibacterial properties, hygroscopicity, and affinity with living tissues (Jeon et al. 2003). Figure 1 shows chemical structure of chitosan.

The basil used as the core material of the microcapsules of this study is an annual plant of the Labiatae family of the Tubiflorae order. Currently, it is cultivated around the world and classified into sweet basil and exotic basil according to the content of the main components, linalool and methyl chavicol (Lee et al. 1999).

Sweet basil is mainly cultivated in Europe and the United States, and consists of 15–50% linalool and 15–25% methyl chavicol. It is mainly used in food. On the other hand, exotic basil is cultivated in Comares and Seychelles, and the content of methyl chavicol is more than 80%. And it is used in luxury perfumes, cosmetics, and soaps (Harborne 2001; Lee & Lee 2004; Mehdizadeh et al. 2016). Basil oil, produced by steam distillation of leaves and flowers, has antibacterial, antifungal, antioxidant, and anti-inflammatory properties, and has been used in various fields for flavoring, medicine, and food (Hercules & Chrissanthy 2017; Yoon & Park 2019).

On the other hand, as a method of attaching a functional material to the fiber, there are methods of attaching a microcapsule containing a functional material through a binder, mixing it into a yarn in a fiber spinning process and coating to fabric by the post-treatment. The method of mixing into the yarn in the fiber spinning process has a disadvantage in that the type of fibers or functional materials is limited. And the method of the post-treatment is poor in durability. On the contrary, the method of encapsulation a functional material can protect the core material from the external environment. Also, the thickness of the microcapsule wall film or the amount of the core material can be appropriately used to control the release of the functional material (Lee 2006). Microcapsules having these characteristics are used in various ways such as toners, pesticides, heat storage materials and fragrances, as well as drug delivery systems depending on the type and manufacturing method of core materials (Lee et al. 2008). Therefore, in this study, the method of encapsulation, which is considered to be the most suitable for functional processing of clothing products, was selected.

Most of the currently developed and commercially available microcapsule finished textile products used melamine formaldehyde microcapsules by in-sito polymerization. But to produce more environmentally friendly chitosan microcapsules O/W/O (oil/water/oil) emulsiﬁcation method was used.

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**Fig. 1** Chemical structure of chitosan
The O/W/O Multiple emulsification method is a method of manufacturing microcapsules by dispersing and crosslinking a water-soluble O/W emulsion into the oil phase once more. So, it is easy to control the release of core material (JO 2015). Prior studies have mainly given aromaticity to fabrics with microcapsules containing fragrance oils, but this study manufactured microcapsules processing agents synthesized from functional natural materials for application in the finishing of functional textile products. Therefore, this study was compared and analyzed the synthesis of microcapsules according to manufacturing conditions for the release of functional substances stably. And also examined the shape, properties, thermal characteristics, and release behavior of the prepared microcapsules. Lastly, fabric was treated with microcapsules agent and then antibiosis was evaluated, as a basic study applying to functional textile products.

**Methods**

**Materials**
In this study, the chitosan, a relatively low molecular weight of $8.0 \times 10^4$, was used as a wall material for microcapsule (Chembio Co., Ltd, Korea). Degree of deacetylation is 95%, and viscosity is 8.5 cps. the basil oil as a core material was purchased from Saero-queens, Korea. Acetic acid (Samchun Chemicals, Inc. 2002a) was used as a solvent for dissolving chitosan, Triton X-100 and Tween 20 (Samchun Chemicals, Inc. 2002a), Span 80 (Junsei, Japan) was used as an emulsifier. Viscosity of Triton X-100 is 240 mPa and Tween 20 is 400 mPa. Glutaraldehyde (Sigma, USA) as a crosslinking agent, N-hexane (Samchun Chemicals, Inc. 2002a) was used for washing the microcapsules, and all reagents were used more than grade 1.

**Manufacturing of microcapsules**
After adding 2 g of emulsifier Tween 20 or Triton X-100 to 20 g of 2 wt % chitosan solution (in 2 wt% acetic acid), it stirred for 10 min to 3000, 6000, 9000 rpm at 25 °C, pH 4.2 with a homogenizer (Ika, T25 digital ultra-turrax). And then, added 2 g of basil oil, and stirred for 10 min at 30 °C, pH 4.5 to prepare O/W emulsion. Thus, pH was adjusted to pH 10 by adding sodium carbonate. Next, O/W emulsion was added by dropwise to mineral oil and stirred for 10 min at 30 °C, pH 8.7 to make an O/W/O emulsion to synthesize microcapsules.

After that, 1.0 mL of glutaraldehyde was added dropwise, so crosslinking was performed while stirring for 1 h at 35 °C, pH 8.5. Finally, synthesized microcapsules were washed three times with N-hexane, filtered with an aspirator, and naturally dried.

Figure 2 shows manufacturing process of crosslinked chitosan microcapsules.

**Measurement of microcapsule properties.**
The forms of microcapsules according to manufacturing conditions was observed by using LV-SEM (Carl Zeiss, Merlin compact). Through comparison of surface, optical manufacturing condition was selected. Chitosan as a wall material, basil oil as a core material, and synthesized microcapsules were measured by FT-IR spectra using infrared spectrometers (Jasco, FT/IR-6300) to determine whether capsules were synthesized or not. Afterward, Gas Chromatography (Perkin Elmer, Clarus SQ8) separated basil oil and analyzed the mass spectrum of characteristic peaks to identify the main components.
Next, the average particle size of the synthesized capsule was calculated using a particle size analyzer (Malvern Korea, Mastersizer2000). The thermal stability of the microcapsules was evaluated by DSC-TGA (TA Instruments, Q600) in the range of 30–700 °C at a rate of 10 °C/min under a nitrogen atmosphere, and compared it is thermal stability to chitosan and basil oil.

**Release properties of microcapsules**

After dissolving 0.2 g of the prepared chitosan microcapsules in 100 mL of artificial sweat (KS K ISO 105-E04), the absorbance at the maximum absorption wavelength (270 nm) of Estragole, the main component of basil oil, at a constant time interval at 34 °C, which is skin average temperature, is measured by an ultraviolet visible spectrophotometer (Jasco, V750) to evaluate the sustained-release properties of the core substance basil oil in the microcapsules. The release rate of basil oil was calculated by the following Eq. 1.

\[
\text{Release rate (\%)} = \frac{A_t - A_0}{A_0} \times 100
\]

where, \(A_t\): absorbance of released basil oil from microcapsules over time, \(A_0\): initial absorbance of basil oil released from microcapsules.

**Antibacterial activity of fabric treated with microcapsules**

Microcapsules finishing was processed in a fabric consisting of 94% tencel, 6% polyurethane. Microcapsule 5% (o.w.f.) and acrylic binder 5% (o.w.f.) are dissolved in water with the bath ratio 1:20, and then immersed in the fabric for 20 min. After taking the fabric out of the solution and pass through a mangle, heat-treated at 120 °C for 3 min. ratio 1: 20, and then immersed in the fabric for 20 min.
Bacteria reduction rate (%) = \( \frac{M_b - M_c}{M_b} \times 100 \)  

(2)

where, \( M_b \): the number of microbe in blank specimen incubated for 18 h, \( M_c \): the number of microbe in test specimen incubated for 18 h.

**Results and discussions**

**Forms of crosslinked chitosan microcapsules**

When manufacturing the O/W emulsion, the results of synthesizing microcapsules using emulsifiers Tween 20 and Triton X-100 are shown in Table 1. The viscosity of Tween 20 is 400 mPa and the viscosity of Triton X-100 is 240 mPa (Samchun 2002a, b) According to previous studies, the use of a surfactant having a high viscosity when manufacturing the O/W/O emulsion shows a homogeneous phase that is well dispersed. When manufacturing the O/W emulsion, it is preferable to use a surfactant having a low viscosity in order to be refined and dispersed (Jo 2015). In the process of manufacturing the O/W emulsion, microcapsules were prepared using Tween 20, which have lower viscosity of the Tween series of surfactants and Triton X-100 with a lower viscosity than Tween 20. SEM photographing showed that in case of Tween 20, average diameter is 7.03 µm and in case of Triton X-100 µm is 5.83 µm. Using the Triton X-100 can manufacture a round, even-sized microcapsules. Since synthesizing the capsules was made in the form of a stable and round sphere, it would be more advantageous than using Tween 20.

Table 2 shows the surface shape of the synthesized microcapsules according to the concentration of the chitosan of the wall material. Since the thickness of the wall material can be the main factor influencing the release of the core material in the manufacture of the microcapsules. First, when using 0.5 wt%, that is a low concentration of chitosan, it is possible to confirm the phenomenon of crushing without showing a stable sphere shape. At this time, the average diameter is about 6.88 µm. On the other hand, when a high concentration of 4 wt% was used, the excess chitosan did not take the form

**Table 1 Microscope images of synthesized microcapsules according to type of emulsifier**

| Type            | Triton X-100 | Tween 20 |
|-----------------|--------------|----------|
| Magnification   | ×1000        | ×1000    |

**Table 2 Microscope images of synthesized microcapsules according to type of emulsifier**

| Type          | Triton X-100 | Tween 20 |
|---------------|--------------|----------|
| Magnification | ×3000        | ×3000    |
of a capsule. When the chitosan concentrations were 1 wt% and 2 wt%, a relatively stable capsules were synthesized, and each diameter is 5.06 µm and 5.83 µm. However, in the previous study, it was confirmed that the release of the core material was delayed due to the increase in the thickness of the chitosan wall surrounding the core material (Han et al. 2008). Therefore, in fabric processing, since it is advantageous to release slowly after washing or for a long time, a concentration of 2 wt% was set as a manufacturing condition in order to slow the release rate by using a relatively high concentration of chitosan Table 3.

The optimum stirring speed may differ depending on the material used in the microcapsule manufacturing. Thus, in order to investigate the effect of the stirring speed when manufacturing the microcapsules, the surface shape of the synthesized microcapsules according to stirring speed is shown in Table 3 by using Triton X-100 as an emulsifier at a concentration of 2 wt% chitosan. First, when the stirring speed is 3000 rpm, the
particle size is uneven and cannot be considered to be stably manufactured. At this time, average diameter is 9.12 µm. It can be seen that a stable spherical microcapsule was synthesized in the case of manufacturing at 6000 rpm and 9000 rpm. Each average diameter is 5.83 µm and 5.87 µm. In the case of 9000 rpm, heat generation and color change
occurred during the synthesis process, so that 6000 rpm was set as the optimal manufacturing condition.

Properties of crosslinked chitosan microcapsules

Figure 3 shows the results of analyzing the FT-IR spectrum of microcapsules, chitosan, and basil oil to confirm whether the chitosan microcapsules are synthesized or not. Since the chitosan characteristic peak is a stretching vibration peak resulting from NH binding at around 3400 cm$^{-1}$, and a C=O binding peak of the amide group at 1640 cm$^{-1}$, is also present in the microcapsules, it can be determined that chitosan was used as a wall material. On the other hand, the characteristic vibration characteristic peak of 1650 cm$^{-1}$, C=C coupling vibration peak, CH bending vibration peak at 1450 cm$^{-1}$, CO elastic vibration peak at 1025 cm$^{-1}$, is not shown in the microcapsule. It can be seen that the substance basil oil is stably incorporated into the capsule to synthesize the microcapsules.

Basil oil is divided into sweet basil and exotic basil according to the ingredients. Sweet basil is mainly composed of linalool, and exotic basil is mainly composed of methyl chavicol, that is, Estragole (Lee et al. 1999). The chemical structural formula of linalool and Estragole is shown in Fig. 4. As a result of separating the components of the basil oil used in the study through gas chromatography (Fig. 5), peaks appear around 12.87 and 14.53 min. Looking at the mass spectrum at 14.53 min, it shows a peak similar to the mass spectrum of Estragole. Therefore, the basil oil used in this study is confirmed to be an exotic basil based on Estragole as a main component, and it is thought that it can be quantified by determining whether or not the Estragole is released and when it is produced in a microcapsule containing basil oil as a core material.

Particle distribution of crosslinked chitosan microcapsules

The following Fig. 6 is a curve showing the particle size and distribution of microcapsules manufacturing at 6000 rpm for 1 h in the optimal manufacturing condition, which is using Triton X-100 and 2 wt% chitosan. The synthesized chitosan microcapsules were added in a small amount of the dispersant Span 80 due to the nature of aggregation in an aqueous solution state and measured by preventing precipitation and aggregation through ultrasonic injection. As a result of the measurement, it appeared to be close with around 5 μm, and the average particle size is 5.83 μm. It can be said to be within the range of 0.5–50 μm and the microcapsules belonging to the range of 1–10 μm size account for 82.3% of the total volume. The size of the microcapsules is usually 1 to several hundred micrometers, but it is common to use a size of 1–10 μm (Kim et al. 1995).

![Fig. 4] Chemical structures of main component of basil oil. a Linalool and b Estragole
It can be an appropriate size of microcapsule for use in functional processing because it is considered that it will not affect the texture or shape during textile processing.

**Thermal stability of crosslinked chitosan microcapsules**

Figure 7a shows the difference in heat flow according to the temperature of chitosan microcapsules containing chitosan, basil oil, and basil oil using DSC. Basil oil showed a peak around 165 °C, but the synthesized microcapsules showed a peak around 300 °C, showing a low melting temperature. This is believed to be the result of the release of the core material, basil oil, due to the destruction of the capsule structure.

In the TGA curve of Fig. 7b, in the case of chitosan, weight loss occurs gradually due to the moisture contained, and sudden weight loss is shown from 275 °C. In the case of basil oil, sudden weight loss is shown from 80 °C and volatilized before 200 °C.
characteristics. However, microcapsules containing basil oil have a weight loss due to the destruction of moisture and microcapsule structure, but they have a weight of more than 15% at 320 °C, so they can be said to have durability against heat by encapsulation.

Release property of crosslinked chitosan microcapsules
Microcapsule processed fabrics are required to evaluate the release characteristics of the core material in the synthesized microcapsules over time, since sustained release of the material in the microcapsules is continuously required even after use and washing. Estragole of basil oil was detected as the main component by GC–MS. According to the previous study, the maximum absorption wavelength of Estragole is around 270 nm. Thus, in order to evaluate the release in an environment similar to the sweat emission phenomenon of the human body, solvent was used in two artificial acidic sweat solution, alkaline sweat solution prescribed in KS K ISO 105: E04. After dissolving the microcapsules in acidic sweat solution, the release of basil oil after 60 h from immediately after dissolution is shown in Fig. 8 and in the case of alkaline sweat solution is shown in Fig. 9. Basil oil in microcapsules is somewhat more releasing up to 5 h after dissolving both acidic and alkaline sweat. After 12 h, it was confirmed as the absorbance increased slightly, basil oil was released in small amounts. Although more release occurred in the

![Fig. 7 a DSC and b TGA curves of microcapsules, chitosan, and basil oil](image-url)
acid solution, the tendency between the two solvents was similar. Since human sweat is acidic, it is thought that the result of release from acidic artificial sweat will be more important. Therefore, it can be judged to have sustained-release by being released slowly and continuously until after 60 h. In general, the reason for encapsulation is to increase the efficiency of the core material. It can be said that it is advantageous to slowly release core material for application to fibers. (Kim & Park 2007). With current technology, it is possible to control the emission according to the purpose of use by adjusting the thickness of the chitosan wall. Recently, using Melamine Formaldehyde, PMMA resin, etc., light-sensitive and temperature-sensitive capsules are being developed (Jung et al. 2017). Application technologies should be developed in a more eco-friendly way, and follow-up studies to improve laundry durability and functionality will be conducted.

**Antibacterial activity of fabric treated with chitosan microcapsules**

Table 4 shows the results of evaluating the antibacterial activity of fabrics treated with chitosan microcapsules by using determination of viable count using Staphylococcus aureus and Klebsiella pneumoniae. Especially, it is important to confirm for reduction of Klebsiella pneumoniae, because pneumococcus enters the human body through
In the case of untreated fabrics, the *Staphylococcus aureus* reduction rate was 20.3% and *Klebsiella pneumoniae* 45%. In the case of treated fabrics with chitosan microcapsules, the *Staphylococcus aureus* reduction rate was 99.9% and *Klebsiella pneumoniae* 99.9%. After 10 times washing, bacteria reduction rate of treated fabric was 99.9% for each bacteria. Therefore, our results have confirmed the basic antibacterial activity for environmentally friendly functional finishing on the fabrics by using chitosan microcapsules.

![Graph](image)

**Fig. 9** Release of basil oil in the chitosan microcapsules dissolving alkaline sweat solution by over time (a) in 5 h, (b) in 60 h

| Treatment   | No. of washing | *Staphylococcus aureus* | *Klebsiella pneumoniae* |
|-------------|----------------|-------------------------|------------------------|
| Untreated   | 0              | 89.0                    | 70.3                   |
| Treated     | 0              | 99.9                    | 99.9                   |
|             | 10             | 99.9                    | 99.9                   |

Table 4 Antibacterial activity of fabrics according to microcapsule treatment
Conclusion

This study is a basic research to apply functional fiber processing by using microcapsules consisting of natural substances. The chitosan microcapsule containing basil oil was manufactured as raw materials for functional fiber processing. And then the properties, thermal stability and release characteristics were examined. Finally, after fabric was treated with microcapsules agent, antibacterial activity was evaluated.

When preparing chitosan microcapsules, the concentration of chitosan was 2 wt%, and the emulsifier for producing O/W emulsion was confirmed to be efficient in terms of capsule formation using Triton X-100 and stirring at 6000 rpm. As a result of analyzing the FT-IR Spectrum of the chitosan microcapsule, the basil oil was incorporated into the microcapsules and the microcapsules were stably synthesized. Also, it was found that the manufactured microcapsule by protecting the basil oil, which is easily volatile by encapsulation, with chitosan having relatively thermal stability, was given heat resistance. As a result of evaluating the release of microcapsules, basil oil in microcapsules has a small amount of release up to 10 h, and is continuously released until after 60 h to release slowly. Also, bacteria reduction rate of fabric treated with microcapsules was 99.9%.

Therefore it can be expected it is possible to apply to functional fiber processing by confirming thermal stability, release property and antibacterial activity as a basic function.

Authors’ contributions
SJR participated in the experimental process and analyzed the results, and HSB designed experimental and reviewed the results. All authors read and approved the final manuscript.

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Availability of data and materials
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Competing interests
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