Applied Chemistry Project

Project title Alginate Capsules for oil entrapment

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Program Bachelor of Science in Applied Chemistry

Academic year 2020

Faculty of Science, Chulalongkorn University

Alginate Capsules for Essential oil entrapment

by

Mr. Thawin Tangsirivanich

In Partial Fulfillment for the Degree of Bachelor of Science

Program in Applied Chemistry (International Program)

Department of Chemistry, Faculty of Science

Chulalongkorn University

Academic Year 2020

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By Mr. Thawin Tangsirivanich

Accepted by Department of Chemistry, Faculty of Science, Chulalongkorn University in Partial Fulfillment of the Requirements for the Degree of Bachelor of Science Program in Applied Chemistry (International Program)

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Abstract

In this study, the drying condition for dried the Kaffir lime oil loaded alginate gellan gum macrocapsules (KLO-AG) and Kaffie lime oil loaded alginate gellan gum/TOBC macrocapsules KLO-AGT macrocapsules were determined at 50 and 60°C by observing their %weight loss. The results showed that the minimum drying hour for the KLO-AG and KLO-AGT macrocapsules was 2 and 3 hrs, respectively. Moreover, the 4 types of chitosan coated capsules were preparing and testing their physical and chemical properties. The results indicated that the capsules coated 2% w/v chitosan solution gave the best results in which highest shell thickness, compression strength and KLO loading. However, the release profiles of those coated capsules were not clear and repeat experiment was necessary.

Keywords: Alginate; Capsule, Gellan gum, Chitosan, Kaffir lime oil

Acknowledgement

Firstly, I would like to thank to my advisor of this project; Associate Professor Pattara Thiraphibundet, Ph.D., for her advice, and for the support throughout the research. I would like to thank the member of report committee; Professor Dr. Sumrit Wacharasindhu. I am also would like to thank the co-advisor of the app chem project; Professor Dr. Paitoon Rashatasakorn for his advising, and a great support throughout my university life.

During the research time in the lab for months, I had been successfully accomplishing many tasks, thank you for many helps from my lab senior; P'Amp and P'Plameow. Also, thanks to my lab partner; Yok, for being my company throughout the project which really helps me from overthinking.

I would like to thank the atmosphere inside the university for boosting the work environment for me to complete this project. Without music, I would not have work half day straight throughout the lab. I really appreciate good music. Also, I appreciate delicious food around Chula for motivating myself to work harder.

Finally, I would like to thank to playing sports that increases my motivation and taught me how to never give up.

Thawin Tangsirivanich

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Chapter 1
Introduction

1.1 Introduction to the research problem and significance

The importance of the fabrication of macrocapsles are that macrocapsules has great roles in major industries of the world such as in pharmaceutical, biochemical, or even in food supplement industry which revolutionizes the boundary of material sciences.

Miss Aungsana Prajaksangsiri reported the achievement of fabrication the kaffir lime oil (KLO)-loaded alginate macrocapsules using a coaxial glass-tube [1]. She studied the various ratio of gellan gum and TEMPO-oxidized bacterial cellulose (TOBC) as the mixture component of shell solution with the aim of enhancement the mechanical property of capsules. Moreover, she coated the capsules using chitosan and determined the release profile of KLO.

To extend this study, the drying condition of capsules prepared from the mixture of alginate, gellan gum and TOBC was determined at 50 and 60°C to observe the suitable time for drying capsules. Moreover, the higher concentration of chitosan solution for coating capsules were preparing and evaluated their release profile storaged for 30 days by GC technique.

1.2 Research objectives

1.2.1 Prepare KLO-loaded alginate/gellan gum macrocapsules using a coaxial glass-tube and evaluate their physical property at 50˚C and 60˚C drying condition.

1.2.2 Prepare KLO-loaded alginate/gellan gum/TOBC macrocapsules using a coaxial glass-tube and evaluate their physical property at 50˚C and 60˚C drying condition.

1.2.3 Prepare chitosan-coated KLO-AGT macrocapsules using immersion method and determine their physical and mechanical properties.

1.3 Literature search

1.3.1 Alginate capsule

Alginate is a natural polymer extracted from brown algae. It has an egg box structure and widely used as stabilizer, thickener, gelling agent, and emulsifier in food industry (Fig.1,1) [2,3]. It is generally used for encapsulation for many kinds of materials, such as drug, gene, cell, enzymes, and lipids. Its properties are aqueous solubility, biocompatibility, non-toxicity, non-immunogenicity, and biodegradability [4].

Coacervation is one of the oldest and most widely used method for encapsulation. It is carried out by precipitation of shell materials around the active substance. Coacervation can be divided into two groups which are simple coacervation and complex coacervation. Simple coacervation is the use of one colloidal solution which in this case is alginate. Complex coacervation is the mixing of two oppositely charged polyelectrolytes for shell material to cover around an active substance.

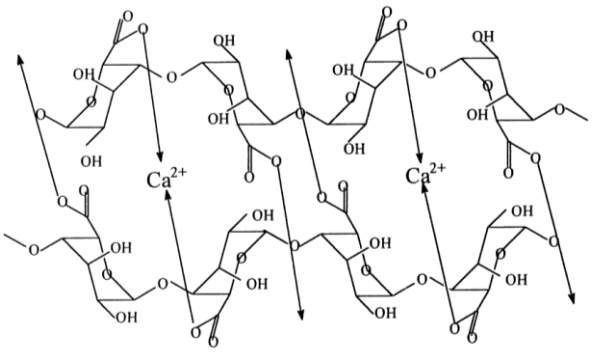


Figure 1.1 Structure of alginate during crosslinking with calcium cations

In 2020, Livia and colleagues [5] prepared encapsulation of black pepper essential oil (EO) using gelatin-sodium alginate complex coacervation. Black pepper was mixed with 40% (w/w) of tween 20 and then added to the gelatin/sodium alginate solution (shell) at different ratios (w/w). The mixture was homogenized at the stirring rate of 10 rpm for 30 min, the pH was adjusted for 4.0 using acetic acid and then $CaCl_2$ solution was added in to form the beads. The sample with core/shell ration of 1:2:1 showed lower encapsulation efficiency and loaded oil content which was 52% and 9.83% respectively.

In 2014, Ujwala and colleagues [6] studied encapsulation of eugenol using gelatin/alginate complex coacervation. Shell forming solution that contained of gelatin and alginate in ratio 4:1 was mixed with eugenol which was emulsified with encapsulating medium. The pH of mixture was adjusted by hydrochloric acid at 40°C until phase separation and then 37% formaldehyde solution was added and stirred for 2h at room temperature for crosslinking process.

Disadvantage of coacervation techniques was average size diameter is quite small which was 30-800μm [7], utilizes crosslinkers and low encapsulation efficiency [8]. Additionally, there are also limitations of active substance which is water-insoluble, therefore it is necessary to use emulsifiers to disperse in the shell material.

In 2014, Nittaya and colleagues [9] manufactured the olive-oil loaded alginate macrocapsules using electro-coextrusion by a high voltage power machine. The capsule construction is based on the pressure from the syringe pump. The diameter of capsule and shell thickness were 0.89-1.61 and 17.4-66.4 mm, respectively.

In 2015, Meghdad and colleagues [10] fabricated the n-nonadecane-loaded alginate capsule using melt coaxial electrospray. The core melted at 50°C before the injection. Later, extruded through the coaxial nozzle into $CaCl_2$ solution bath. The various concentrations of alginate solution and working distance had beed studied how they affected the capsule size. The result showed that reducing alginate concentrations and working distance decreased the capsules diameter (275-480 μm).

In 2017, Danish and colleagues [11] reported a microfluidic platform for generating alginate microcapsules with a combination of coaxial and T-junction (Fig. 1.2). At the coaxial, alginate and silicone oil were used as shell and core and were injected through their inlets to gain smaller particles. Smaller particles were mixed with aqueous calcium chloride droplets which flowed out from a T-junction. Alginate microcapsules diameter was in a range of 800-950 μm.

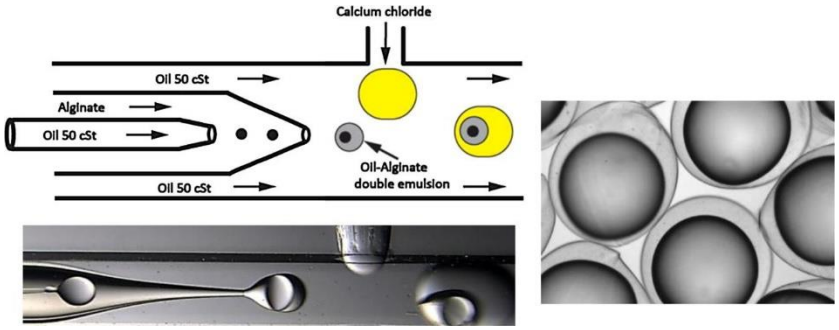


Figure 1.2 Droplet generation from coaxial and T-junction followed by merging for polymerization and oil core-alginate shell microcapsules by generation [11]

1.3.2 Additives for alginate capsule in this study

1.3.2.1 Gellan gum

Gellan gum is a bacterial exopolysaccharide produced by the bacterial Sphingomonas elodea. Gellan gum is a linear anionic polymer with a tetrasaccharide repeating sequence (Fig. 1.3) [12]. Structually a double helix, formed by two intertwined left-handed threefold helical chains. Gellan gum can form gels with the existence of cations because it has electrostatic interactions with the carboxylate groups of the polymer chains [13]. Gellan gum are used in variety of applications in the field of food, biomedicine, cosmetic, or even pharmaceutic due

to its thickening agent, gelling agent, nontoxic, flexibility, biocompatible, and biodegradable [14-16].

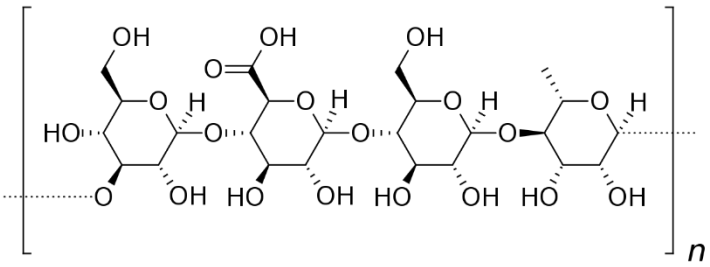


Figure 1.3 Structure of gellan gum

In 2015, Seongcheol and colleagues [17] prepared graphene oxide gellan gum sodium alginate nanocomposites (GO-GG-Alg) by a simple solution mixing-evaporation method and studied the mechanical properties of the blended nanocomposites. The results showed that mixing GO and GG in Alg solution improved the mechanical properties.

In 2019, Yukai and colleagues [18] successfully fabricated locust bean gum/gellan gum hydrogel (LBG/GG) by using borax as a crosslinking agent for biomedicine applications. The tensile strength of LBG/GG hydrogel containing 2 wt% of GG was 33.6 kPa, which is almost three times that of the pure LBG hydrogel. These results indicate that LBG and GG can complement each other to improve the mechanical properties of the hydrogels.

1.3.2.2 TEMPO-oxidized bacterial cellulose (TOBC)

TOBC is one of the modification bacterial cellulose (BC) via oxidation reaction to generate carboxylate groups on the surface of BC using 2,2,6,6-tetramethyl-1-piperidine-N-oxy radical (TEMPO), sodium bromide (NaBr) and sodium hypochlorite (NaClO) system (Fig. 1.4). Gram-negative bacteria Acetobacter xylinum produced BC generally. It is 3D nanoporous fiber networks with fiber diameters of 30-50 nm [19]. BC fiber are strong due to hydrogen bonding and has high degree of crystallinity, high mechanical strength, and high hydrophilicity [20,21].

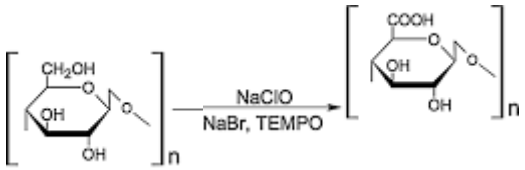


Figure 1.4 Synthesis reaction of TOBC

Carboxylate groups on the surface of TOBC increased the gaps between fibers due to the interaction of anionic charges, which resulted in a well-aqueous dispersed state [22]. TOBC and alginate shared similar chemical structure, which they both participate in Ca^{2+} crosslinking process.

In 2015, Minsung and colleagues [22] produces TEMPO-oxidized bacterial cellulose (TOBC)-sodium alginate (SA) composites to improve the properties of hydrogel for cell encapsulation. Increasing TOBC content can enhance the compression strength of the composites. The compressive strength value with 20wt% of TOBC was the highest. On the other hand, the addition of more than 30wt% of TOBC deteriorated the compressive strength of the composites by inducing the slippage of TOBC nanofibers. The TOBC chains were not involved in crosslinking weakened the composite.

In 2016, Bhawna and colleagues [23] developed biodegradable nanocomposite films with enhanced mechanical and barrier properties by incorporating TEMPO-oxidized cellulose nanofibers (TEMPO-CNFs) into a chitosan matrix using an environmentally friendly casting method. The addition of higher TEMPO-CNFs content (20-25 wt%) increased both tensile stress and young's modulus from 17.5-18.7 MPa and 545-652 MPa which were much higher than the pure chitosan films.

In 2019, Luyao and colleagues [24] prepared fully biodegradable composites from a novel TEMPO-oxidized bacterial cellulose (TOBC) reinforced polylactic acid (PLA) for fused deposition molding (FDM). PLA mechanical properties and PLA/TOBC composites showed that the addition of TOBC could improve the mechanical strength and toughness of the materials. By adding 1.5% TOBC, the tensile strength, maximum bending strength and elastic modulus of the PLA were increased by 9.2%, 45% and 49%, respectively.

1.3.2.3 Chitosan

Chitosan is produced by partial deacetylation of chitin (Fig.1.5) and is the second most abundant biopolymers the found-on earth after cellulose. It is a linear cationic polysaccharide. Chitosan is hydrophilic because it has amino and hydroxyl functional groups in its repeating structural unit. In acidic conditions, the positively charged amine groups in chitosan enable electrostatic interactions between chitosan and other anionic biopolymers including sodium alginate [25,26].

Chitosan are commonly used in food, pharmaceutical, biomedical applications, and coating materials because it has good biocompatibility, biodegradability, and antibacterial properties [27,28]. Chitosan is divided into four types which are chitosan oligosaccharide, low

molecular weight chitosan, medium molecular weight chitosan, and high molecular weight chitosan. Different types of chitosan contain different amounts of surface charge and different mechanical properties.

Chitosan coating is one of the techniques to form the thin film on the capsule surface. Chitosan chains contain negative charges which can interact with the positive charges of alginate chains to form semi-permeable membrane. Chitosan gave some properties to the capsules such as reducing porosity, leakage of the substance inside capsule but it increases smoother surface for applications [29-31].

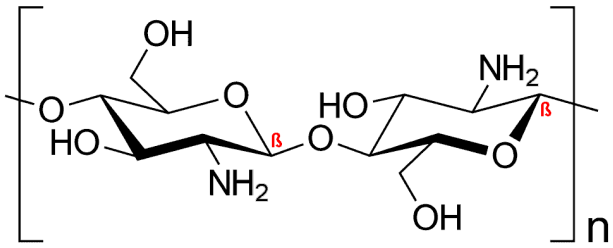


Figure 1.5 Structure of chitosan

In 2020, Xiaoxi and colleagues [25] studied the effect of molecular weight of chitosan to coating on alginate particles for improving the viability of Lactobacillus rhamnosus GG (LGG). Three types of molecular weight of chitosan have chitosan oligosaccharide (COS), low molecular weight chitosan (LMWC) and medium molecular weight chitosan (MMWC). SEM images of the surface of chitosan oligosaccharide (COS) coated alginate particles presented the smoothest followed by low molecular weight chitosan (LMWC) coated alginate particles. On the other hand, MMWC formed the roughest surface on the alginate particle. Besides, COS coated alginate showed a significantly higher hardness and springiness. This was expected because a smaller molecular weight of COS can form homogeneous, intact, and rigid layer outside the alginate particles. Increasing molecular weight of chitosan decreased the hardness of the particles.

In 2013, Ximeng and colleagues [32] prepared chitosan coated alginate/poly(N-isopropylacrylamide) beads (CAPB) for drug delivery field. It was transferred into the mixed solution containing of $CaCl_2$ and chitosan under gentle stirring. The spherical beads formed instantly were kept for 40 min to complete crosslink, the average size tended to increase with increasing chitosan solution. Not only the lower rate of released drug, but also higher stability than the uncoated beads. Increasing chitosan concentration increased drug loading efficiency

due to thicker chitosan shell on the surface of beads reduced the possibility of leaching out of the drug during the bead preparation.

In 2020, Samah and colleagues [33] fabricated paracetamol alginate beads using electrospray method and the additional coating with chitosan to improve the taste masking efficiency (Fig 1.6). Paracetamol powder was added into alginate solution and homogeneous suspension. Chitosan coating did not produce significant changes in the bead size and the bead shape was spherical as a result. Chitosan coating enhanced encapsulation efficiency to 50-76% due to electrostatic interactions between positively charged amine groups of chitosan and negatively charged carboxyl groups of alginates produce alginate-chitosan complex. This complex blocks up the large pore of calcium alginate-beads and forms a polyelectrolyte complex membrane on the surface of beads which ultimately reduced the diffusion of paracetamol.

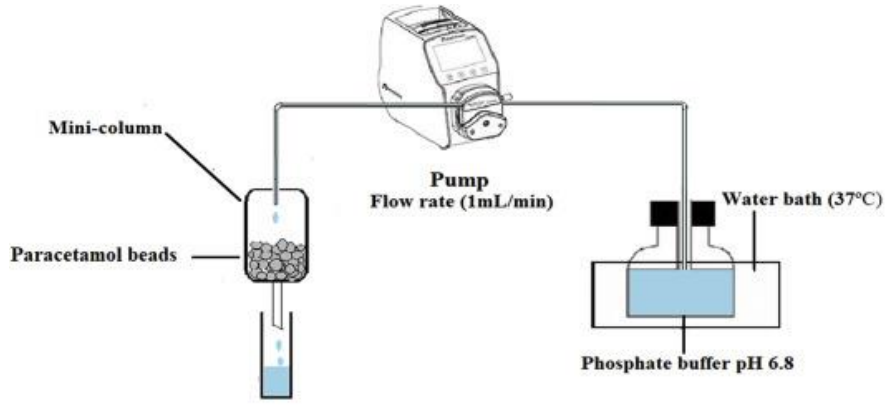


Figure 1.6 Taste making evaluation method [32]

As a summary, coacervation technique has several disadvantages such as it provided small average size diameter for macrocapsules in the range of 30-800 μm, utilizes crosslinkers, and resulted in low encapsulation efficiency. As the existing technology such as gelatin-sodium alginate complex coacervation reported in the introduction above, resulted in low encapsulation efficiency in the capsules fabricated. This came up to the research idea, which is firstly, the addition of TOBC because TOBC contains strong Hydrogen bond, high degree of crystallinity, high mechanical strength, and high hydrophilicity. Secondly, is the chitosan coating because chitosan has good biocompatibility, biodegradability, and antibacterial properties. Also, chitosan chain has the negative charge, which can interact with

the positive charge of alginate chains. Due to all these following properties, the mechanical properties of the macrocapsules can be improved.

Chapter II

Experimental

2.1 Chemicals and materials

Sodium alginate was purchased from Union Chemical 1986 Co., Ltd. (Thailand). Gellan gum was purchased from Siam Victory Chemicals. Calcium Chloride was purchased from Sigma-Aldrich (USA). Kaffir Lime Oil (KLO) was purchased from Thai-China Flavors and Fragrances Industry Co., Ltd. Chitosan was purchased from Bio21 Co., Ltd.

2.2 Experimental Procedure

2.2.1 General preparation of KLO-loaded alginate capsule using two coaxial glass-tube

The shell forming solution was prepared using the mixture of 1%w/v alginate and other polymers. The macrocapsules were fabricated using a set of equipment showing in Fig. 2.1 The shell forming solution and Kaffir lime oil (KLO) were loaded in syringes no.1 and no.2, respectively. Both substances dropped through a coaxial glass tube at various flow rates controlled by the syringe pumps into a solution of 5%w/v $CaCl_2$. The flow rate of the shell forming solution and the KLO were 10.5 mL/min and 1.7 mL/min, respectively. The capsules were soaked in the $CaCl_2$ solution and stirred for 10 minutes, subsequently, they were washed to remove the excess Ca^{2+} ion on the surface of capsules. The percentages of capsule formation efficiency (%CFE) was calculated by the following equation:

$$\%CFE = \frac{Number\ of\ oil\ loaded\ capsules\ in\ 60s}{Number\ of\ total\ drops\ in\ 60s}\ x100 \quad \text{..... Equation 1}$$

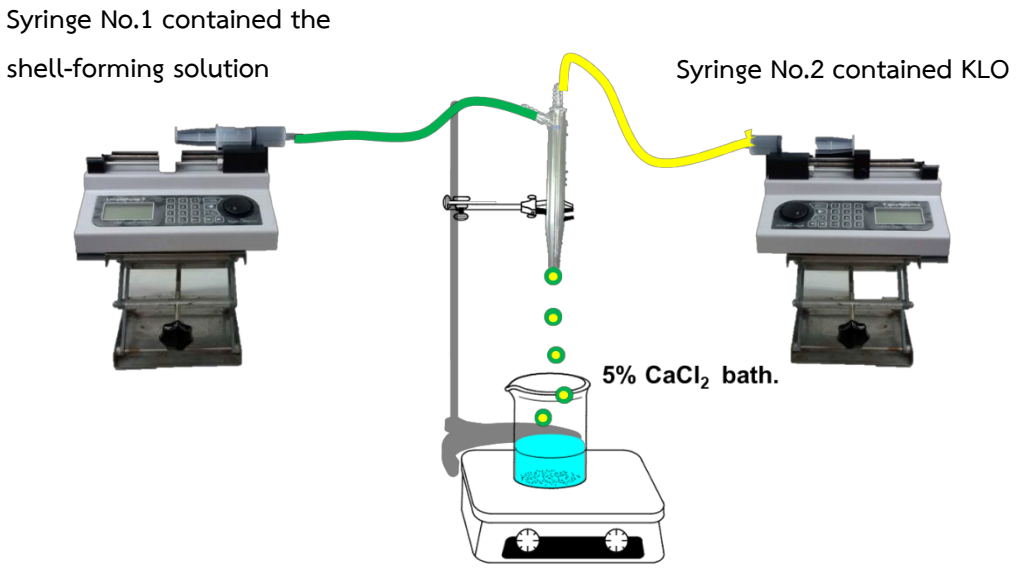


Figure 2.1 A set of equipment for fabrication KLO macrocapsules using a coaxial glass-tube

2.2.2 Preparation of KLO-loaded alginate/gellan gum macrocapsules

The KLO-loaded alginate/gellan gum (KLO-AG) macrocapsules were prepared by using the method mentioned in Section 2.2.1, in which the shell forming solution were 1%w/v of alginate and 1%w/v of gellan gum. The %CFE was calculated. Consequently, the fresh capsules were further oven-dried at 50 and 60˚C. The weight loss percentage of capsules was evaluated at every hour until the weight is constant. Moreover, the average capsule size of each time interval was determined (n=15).

2.2.3 Preparation of KLO-loaded alginate/ gellan gum/ TOBC macrocapsules

The KLO-loaded alginate/gellan gum/TOBC (KLO-AGT) macrocapsules were prepared by using the method mentioned in Section 2.2.1, in which the shell forming solution were 1%w/v of alginate, 1%w/v of gellan gum and 0.15%w/v of TOBC. The %CFE was calculated. Consequently, the fresh capsules were further oven-dried at 50 and 60˚C and examine their weight loss percentage.

2.2.4 Characterization of KLO-AG and KLO-AGT macrocapsules

2.2.4.1 Size and optical appearance

The size of microcapsule was measured using a digital electronic Vernier caliper micrometer (Mitutoyo, Tokyo, Japan). The optical appearance was taken by a digital camera at 40X magnification.

2.2.4.2 Weight loss percentage

Macrocapsules are weighted by using digital weighing scale in the unit of grams. These capsules are weighted each time to determine the weight loss percentage as shown in equation 2.

$$\%Weight\ loss = \frac{Wi - Wf}{Wi} x100 \text{..... Equation 2}$$

Wi = initial weight
Wf = final weight

2.2.4.3 Morphology analysis

The surface morphology and cross-section were observed by scanning electron microscopy (SEM, JSM-IT-500HR, JEOL, Japan). However, KLO in macrocapsules are needed to be removed before examination. Macrocapsules were freeze-dried, half-cut, and washed with acetone. Then, the macrocapsules were mounted on the metal stub by double sided adhesive tape, gold coated under vacuum and the images were taken at an acceleration voltage of 5 kV.

2.2.5 Preparation and characterization of chitosan-coated KLO-AGT macrocapsules

The chitosan coated macrocapsules were prepared by immersion method. Fresh fabricated KLO-AGT capsules were immersed in chitosan solution for 10 min in a magnetic stirring environment. Four different concentrations of chitosan solution in 12% w/v of 1M HCl, which were 0.5, 1.0, and 2.0, and 3.0% w/v, were studied. After immersion, excess chitosan solution was washed away and dried in an oven at 50°C for 4hrs. The Scanning Electron Microscope (SEM), compression strength and the release profile of all types of chitosan-coated KLO-AGT (C_x-KLO-AGT) macrocapsules were examined as followed.

2.2.5.1 SEM

The Scanning Electron Microscope (SEM) was done by freeze-dry different formulas of macrocapsules, then use the knife to separate each of the macrocapsules half. Then, put each of the microcapsule samples on carbon tape which then attached on the metal stand. After that, put the sample into the SEM for the analysis. The analysis of SEM is to analyze the cross-section and surface of each sample of macrocapsules to see the difference.

2.2.5.2 Compression test

Compression test was analyzed by a TA-XT2i (icon) Texture Analyzer (Stable Micro Systems, England). The instrument is equipped with a 30 kg load cell fitted with a TA-P/6 separating rod fixture. The P/6 has short rods of 6mm diameter each. The applied force is recorded as a function of 50% strain. Macrocapsules were mounted onto the pair of rods. The probe moves downwards at a speed of 2mm/sec until the capsules were broken for compression test. Compression test was reported as average values determined from fifteen specimens.

2.2.5.3 The KLO releasing study

The release profile was made in the interval of 1 month, which was separated into 0, 5, 15, 20, and 30 days. It was done to compare with the normal type of macrocapsule (KLO-AG and KLO-AGT macrocapsules) which was not coated with chitosan. The KLO can penetrate through capsule shell and lead to the loss of capsule weight. Thus, the KLO loss percentage was determined at the same time interval as KLO release profile evaluated by HS-SPME-GC.

- **KLO loss percentage**

Different KLO macromolecules were weighed approximately 0.15xx g (Wi) and then these capsules were weighed at the interval times, which consists of 5, 15, 20 and 30 days (Wf). The weight loss then converted to a percentage by using Equation 2.

- **Release profile study**

To evaluate the amount of KLO retentate in different macrocapsules, the citronellal content which is the major component in KLO was monitored by headspace solid phase micro-extraction gas chromatography (HS-SPME-GC). Capsules were placed in the 20ml open capped headspace glass vials and later taken to analyze at 5, 15, 20, and 30 days. There were 0.15xx g of capsule in each vial and the vial is non-reused after analysis. Each data was done in quadruplicate. (This study was done by the Center for Scientific and Technological Equipment, Suranaree University of Technology).

2.2.6 HS-SPME procedure

The SPME fiber coated with 65 μm of polydimethylsiloxane/ Divinylbenzene (PDMS-DVB) was used for the absorption of volatile compounds releasing from KLO macrocapsules. Firstly, SPME fiber was conditioned in the injector of the GC system at temperature 270˚C for 30 min. The capsules vial was sealed with PTFE/silicone and maintained an equilibrium time for 5 min at 35˚C. Subsequently, the SPME fiber was then exposed inside the vial to absorb volatile compounds in the headspace of the sample vial for 1 min. The SPME fiber was pulled out from the vial and immediately injected to the GC inlet with a spitless using thermal desorption at temperature 250˚C for 5 min.

2.2.7 GC-MS analysis

Volatile compounds were detected by an Agilent Technologies 7890 A & 7000 B GC/MS Triple Quad using an HP-5 MS capillary column (30m x 0.25mm i.d., 0.25μm film thickness;

J&W 322 Scientific USA). Helium of high purity (99.999%) was used as the carrier gas at a flow rate of 2ml/min. The GC oven temperature was programmed to increase from 40°C to 150°C at a rate of 3°C/min and 150°C to 250°C at a rate of 20°C/min. MS detection was performed with electron ionization (EI) at 70 eV, operating in the full-scan acquisition mode in the m/z range 35-300 Da. The transfer line and ion-source temperatures were 250°C and 230°C, respectively. Identification of each component was made by matching their recorded mass spectra with reference spectra in the data base (NIST) and compared their LRI and mass spectra with published data in the literature.

Chapter III
Results and discussion

3.1 Characterization of KLO-loaded alginate/gellan gum (KLO-AG) macrocapsules

The KLO-AG macrocapsules were successfully fabricated by using a coacervation method (see raw data in Appendix A). The %CFE of this experiment was 90.6±0.2%. Then, macrocapsules were dried at different temperatures (50 and 60˚C). Their average size and appearance were recorded as shown in Table 3.1. As expected, the average size of macrocapsules decreased depend on the drying time. Moreover, the loss of humidity in capsule shells made the appearance of capsule shell clearer.

The %weight loss was summarized in Table 3.2 and Figure 3.1. The %weight loss started to constant after 2 hours both 50 and 60˚C. This indicated that 2 hours was the minimum hour for drying these macrocapsules.

Table 3.1 The average size and appearance of the KLO-AG macrocapsules dried at 50℃ and 60˚C

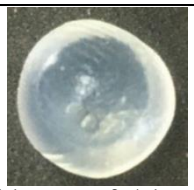
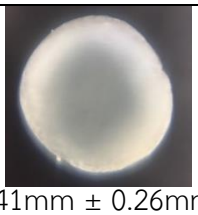
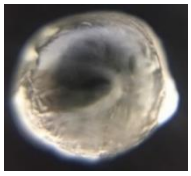
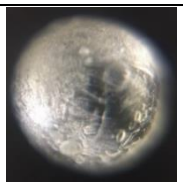
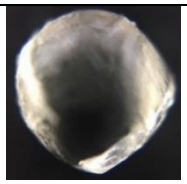
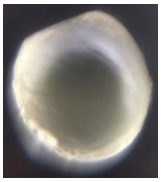
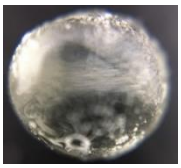
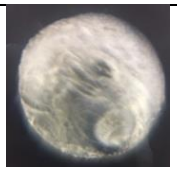
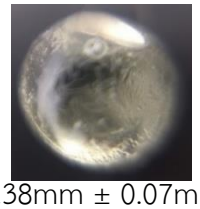
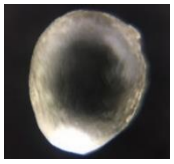
Time interval (hr)	Dried temp (˚C)	
	50	60
0	 5.46mm ± 0.24mm	 5.44mm ± 0.14mm
1	 3.65mm ± 0.15mm	 4.41mm ± 0.26mm
2	 3.26mm ± 0.03mm	 3.84mm ± 0.06mm
3	 3.36mm ± 0.12mm	 3.79mm ± 0.08mm
4	 3.36mm ± 0.07mm	 3.74mm ± 0.04mm
5	 3.38mm ± 0.07mm	 3.71mm ± 0.03mm

Table 3.2 The %weight loss of KLO-AG macrocapsules dried at 50 and 60˚C

Time interval (hr)	Dried temp (˚C)	
	50	60
0	0%	0%
1	62.8%	59.3%
2	83.4%	78.6%
3	85.1%	80.7%
4	85.9%	82.1%
5	86.9%	83.0%
10	87.1%	83.3%

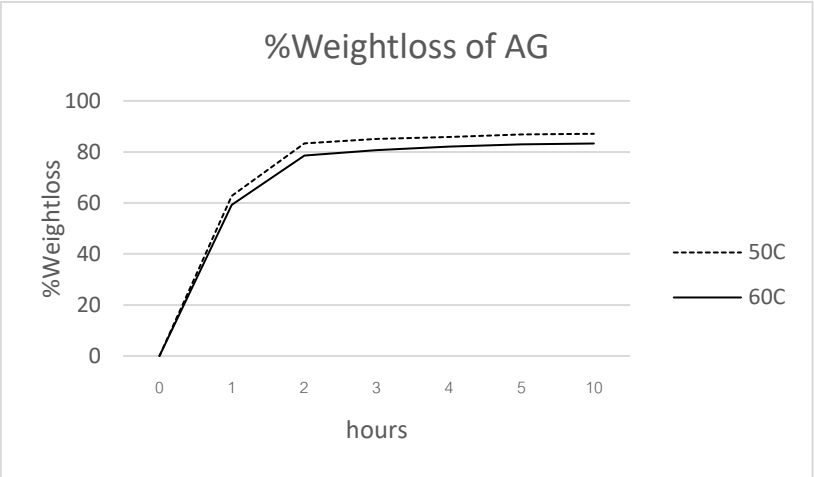
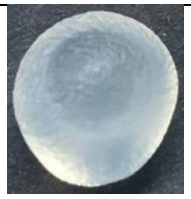
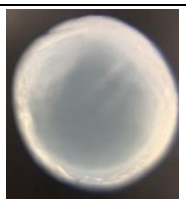
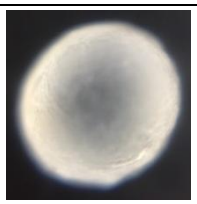
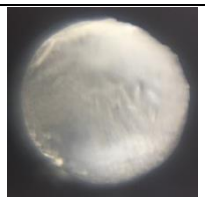
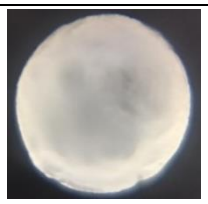
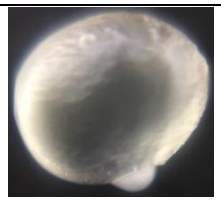
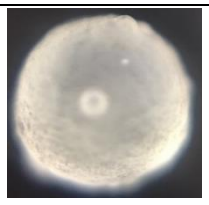
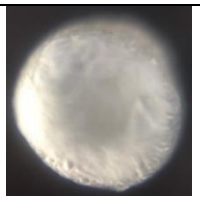
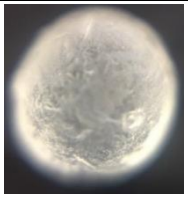
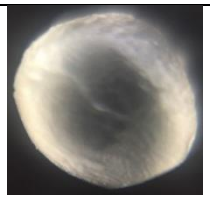


Figure 3.1 The %weightloss of KLO-AG macrocapsules dried at 50℃ and 60˚C.

3.2 Characterization of KLO-loaded alginate/gellan gum/TOBC (KLO-AGT) macrocapsules

TEMPO-oxidized bacterial cellulose (TOBC) with 0.1% of the degree of oxidation was added into the gellan gum and alginate solution with the aim of increase the properties of macrocapsules. Miss Aungsana Prajaksangsiri [1] had been studied the effect of various TOBC amount towards the mechanical properties of macrocapsules. In this study, the drying condition of the macrocapsules was evaluated (see raw data in Appendix B). The size decreased by the drying time as shown in Table 3.3. However, the addition of TOBC in the shell solution resulted in the more turbidity of shell appearance after longer drying compared to those of KLO-AG macrocapsules.

Table 3.3 The average size and appearance of the KLO-AGT macrocapsules dried at 50 and 60˚C

Time interval (hr)	Temp (˚C)	
	50	60
0	 5.62mm ± 0.15mm	 5.78mm ± 0.19mm
1	 4.49mm ± 0.26mm	 4.21mm ± 0.18mm
2	 3.57mm ± 0.10mm	 3.56mm ± 0.13mm
3	 3.57mm ± 0.10mm	 3.5mm ± 0.12mm
4	 3.47mm ± 0.05mm	 3.44mm ± 0.07mm
5	 3.44mm ± 0.05mm	 3.43mm ± 0.07mm

The %weight loss was summarized in Table 3.4 and Figure 3.2. The %weight loss started to constant after 3 hours at both 50 and 60˚ which was longer than that of KLO-AG macrocapsules. This might be the extra hydrogen bonding between TOBC chain and water molecules.

Table 3.4 The %weight loss of KLO-AGT macrocapsules dried at 50 and 60˚C

Time interval (hr)	Temp (˚C)	
	50	60%
0	0%	0%
1	53.7%	64.6%
2	55.4%	55.1%
3	14.4%	17.8%
4	5.7%	9.2%
5	4.9%	5.1%
10	2.9%	3.4%

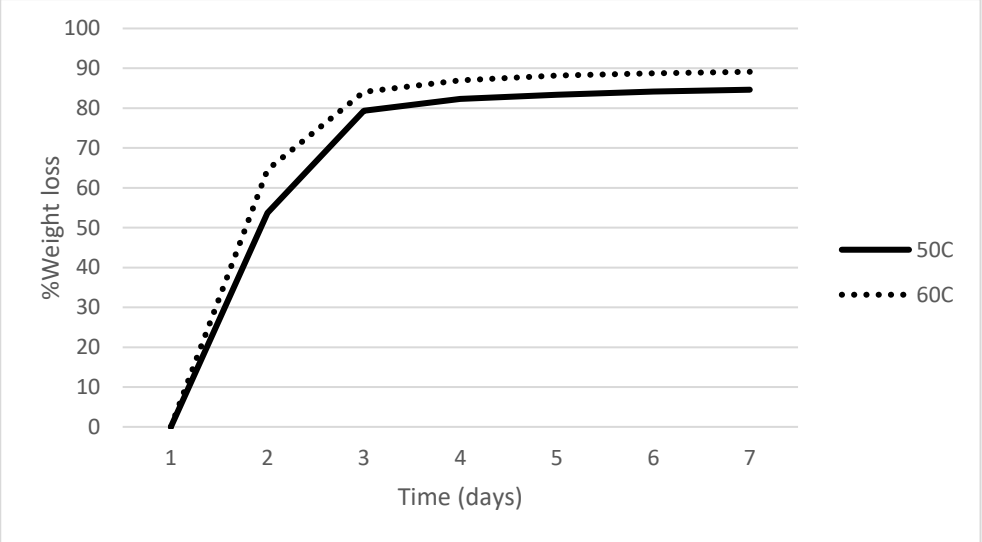


Figure3.2 The %weight loss of KLO-AGT macrocapsules dried at 50°C and 60˚C

3.3 Characterization of chitosan-coated KLO-AGT macrocapsules

The KLO-AGT macrocapsules were further coated by various concentrations of chitosan solutions (see raw data in Appendix C). Chitosan has been known as the effective coating substance for alginate particles. The electrostatic interaction between the positively charged amine groups (NH_3^+) of chitosan and the negatively charged carboxylate groups (COO^-) of alginate and gellan gum leads to the formation of chitosan layer on the capsule surface.

In this study, four concentrations of chitosan which were 0.5, 1.0, 2.0 and 3.0%w/v were used for coating fresh macrocapsules. The average size, shell thickness and appearance of those coated capsules were shown in Table 3.5. The average size of 4 capsule types was not significantly different. However, the larger thickness of capsule shell was the capsules which was coated by 2.0% chitosan. At the 3.0% chitosan, the thickness decrease which might be the repulsion of positive charges on chitosan chains.

The same trend of compression strength (Table 3.6 and Fig. 3.3) was observed in which the maximum value was the capsules coated by 2.0% chitosan solution.

The %weight loss after storing 20 days (Table 3.7 and Fig. 3.4) and the area of citronellal peak evaluated by GC (Table 3.8 and Fig3.5) were not in the same trend. The maximum loading of KLO was found in the capsules coated by 2% chitosan solution. After 5 days storing in the open condition, the KLO loading significantly decreased for all types of capsules. The reason of these results still unclear, and more repeat experiments were needed to confirm. The cause of this error possibly caused from the capsules are not well prepared due to the preparator, and the oil itself leaks out from the capsules due to that the capsules are brittle because of too much force are exerted on capsules during the experiment.

Table 3.5 The average size, shell thickness and appearance of the KLO-AGT-C macrocapsules

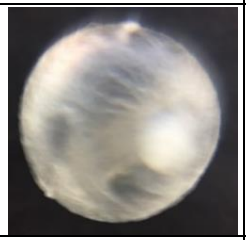
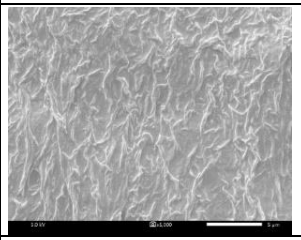
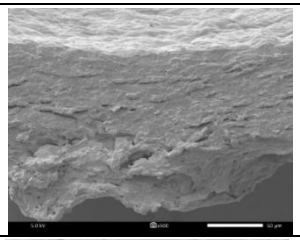
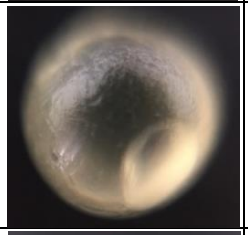
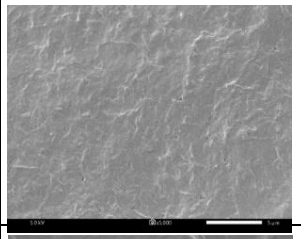
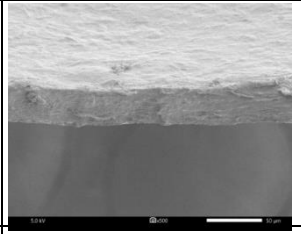
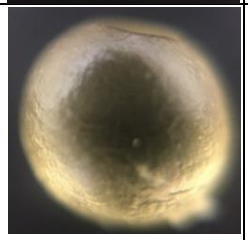
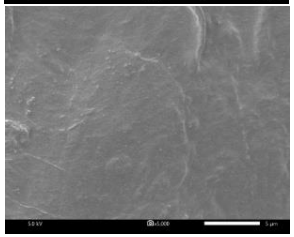
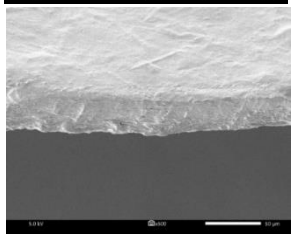
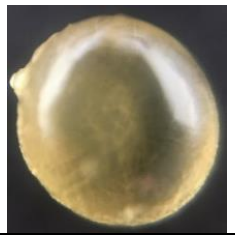
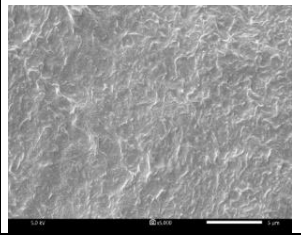
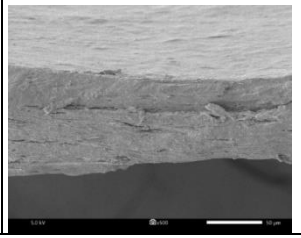
Capsules	Images of dry capsules by camera	Images of SEM		Average Size (mm)	Shell Thickness (μm)
		Surface	Cross-section		
KLO-AGT-C0.5				3.53 ± 0.28	95.08 ± 36.82
KLO-AGT-C1				3.67 ± 0.31	97.33 ± 33.10
KLO-AGT-C2				3.38 ± 0.29	94.09 ± 30.72
KLO-AGT-C3				3.57 ± 0.20	64.96 ± 15.41

Table 3.6 Compression strength data of chitosan coated KLO-AGT macrocapsules

Samples	Compression strength (KPa)
C0.5	339.73
C1	372.93
C2	444.77
C3	418.39

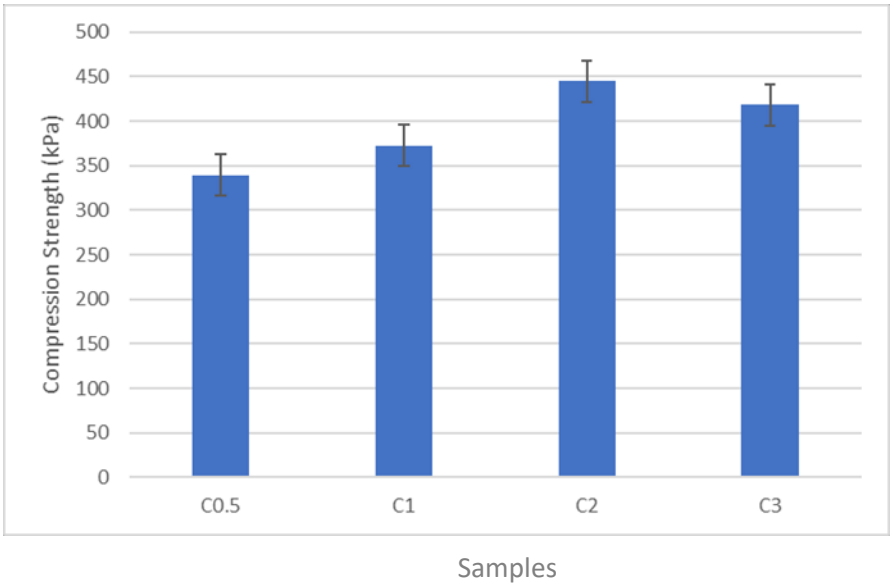


Figure 3.3 Compression strength data of chitosan coated KLO-AGT macrocapsules

Table 3.7 The %weight loss of KLO-AGT-C macrocapsules stored for 30 days

Time (days)	%Weight loss of capsules			
	C0.5	C1	C2	C3
0	0%	0%	0%	0%
5	2.42%	3.17%	2.82%	1.40%
10	4.06%	5.43%	5.02%	3.09%
15	5.34%	6.89%	5.76%	2.76%
20	6.05%	9.03%	9.20%	5.47%
25	6.11%	8.51%	8.45%	3.89%
30	8.99%	11.43%	13.92%	9.29%

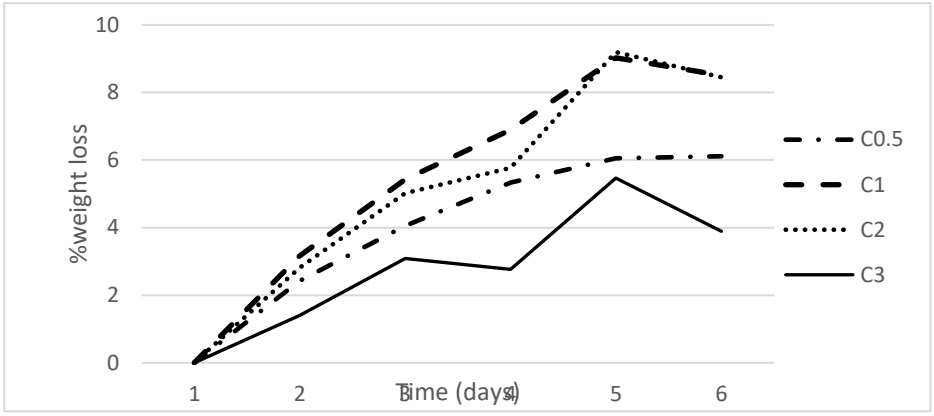


Figure 3.4 The %weight loss of KLO-AGT-C macrocapsules dried at 50°C

Table 3.8 The maximum peak area data of chitosan-coated KLO-AGT macrocapsules

Capsules	Area of citronellal peaks ($\times 10^8$) stored for... (day)			
	0	5	15	20
C0.5	15.2 ± 3.93	2.47 ± 0.66	4.79 ± 2.79	1.60 ± 0.52
C1	14.6 ± 5.30	4.25 ± 0.59	2.99 ± 4.17	0.87 ± 0.44
C2	49.1 ± 46.1	1.99 ± 0.84	1.42 ± 1.08	0.50 ± 0.17
C3	4.18 ± 1.03	4.17 ± 2.39	0.77 ± 0.82	2.14 ± 1.57

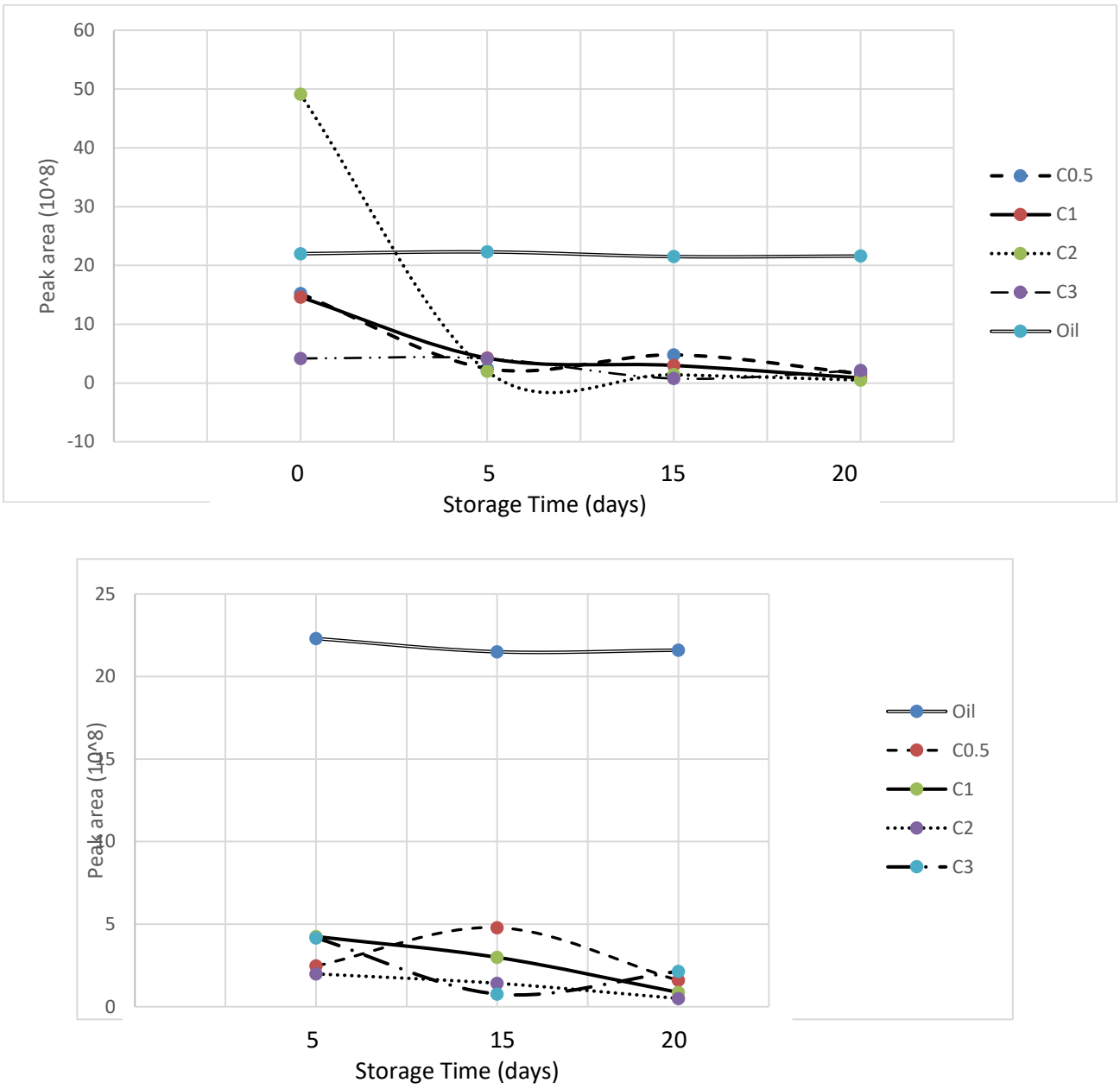


Figure 3.5 Area of citronellal peak released from 4 types of chitosan-coated KLO-AGT macrocapsules which open storage for 20 days (oil sample was tested as reference for injection error.

Chapter IV

Conclusion

In this study, the drying condition for dried the KLO-AG and KLO-AGT macrocapsules were determines at 50°C and 60°C by observing their %weight loss. The results showed that the minimum drying hour for the KLO-AG and KLO-AGT macrocapsules was 2 and 3 hrs, respectively. Moreover, the 4 types of chitosan coated capsules were preparing and testing their physical and chemical properties. The results indicated that the capsules coated 2% w/v chitosan solution gave the best results in which highest shell thickness, compression strength and KLO loading. However, the release profiles of those coated capsules were not clear and repeat experiment was necessary.

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Appendix

Part A: Experimental size data of KLO-AG macrocapsules

Table A1 Size data of KLO-AG macrocapsules which dried at 50°C at 0 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	6.45	5.46	5.76	5.17	5.10
2	5.71	5.42	5.14	5.27	5.28
3	6.72	5.74	5.22	6.89	6.12
4	5.48	5.03	5.66	6.48	5.12
5	5.72	5.49	4.59	5.29	5.28
6	6.27	5.21	4.64	5.36	6.12
7	5.55	6.03	6.04	4.74	5.32
8	5.91	5.29	4.77	5.09	5.05
9	5.87	4.41	6.19	5.58	5.17
10	6.30	5.27	4.79	5.56	5.65
11	6.09	5.42	5.62	4.78	4.75
12	6.29	5.24	5.28	5.65	5.46
13	5.09	5.13	5.48	6.23	5.16
14	5.90	5.91	5.98	5.18	5.12
15	6.28	5.25	5.31	5.17	5.29
16	5.46	5.29	5.50	5.98	5.52
17	5.65	5.02	4.67	5.92	5.41
18	5.81	5.12	5.30	4.54	5.42
19	5.71	6.34	6.40	5.27	4.62
20	5.29	4.77	4.92	4.74	4.18
Average	5.88	5.34	5.36	5.44	5.26

Average ± SD = 5.46 ± 0.24

Table A2 Size data of KLO-AG macrocapsules which dried at 50˚C at 1 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	4.87	3.2	3.44	3.31	4.28
2	4.54	3.85	3.82	3.83	2.93
3	3.61	3.65	3.2	3.82	3.28
4	3.74	4.37	3.42	4.04	3.8
5	4.33	3.51	4.63	3.58	4.48
6	3.65	2.97	4.34	3.23	4.18
7	3.32	4.07	3.07	2.77	3.15
8	3.3	3.87	4.61	3.49	3.72
9	3.7	3.46	4.09	4.14	3.26
10	3.28	3.35	3.5	3.91	4.21
11	3.92	4.02	3.24	3.81	3.02
12	4.33	3.48	3.21	3.73	3.46
13	4.13	3.31	4	2.94	3.07
14	4.01	3.31	3.56	3.1	3.02
15	4.7	3.77	3.25	3.96	3.03
16	3.48	3.75	3.86	3.5	3.71
17	3.31	3.58	3.92	3.11	3.54
18	3.54	3.24	4.17	3.73	3.09
19	3.99	3.34	3.99	3.77	3.01
20	3.08	3.88	4.08	3.26	3.74
Average	3.84	3.6	3.77	3.55	3.5

Average ± SD = 3.65 ± 0.15

Table A3 Size data of KLO-AG macrocapsules which dried at 50˚C at 2 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.32	3.42	3.08	2.14	3.29
2	3.15	3.60	3.55	3.60	3.07
3	3.16	3.37	2.69	3.17	3.08
4	3.41	3.20	3.51	3.26	2.94
5	3.46	3.08	3.39	3.88	3.24
6	3.36	2.89	3.16	3.78	3.33
7	3.34	3.19	3.35	3.55	3.50
8	2.91	3.29	3.04	3.21	2.96
9	3.10	3.07	3.30	3.24	3.65
10	3.42	2.47	3.04	3.22	3.32
11	3.55	3.38	2.84	2.96	3.48
12	3.42	3.42	3.48	2.50	2.88
13	3.28	2.95	3.38	3.58	3.52
14	3.30	3.15	3.11	3.27	4.15
15	3.46	3.29	3.71	3.49	2.65
16	3.46	3.47	3.12	3.35	3.36
17	3.37	3.36	3.40	2.88	3.41
18	3.24	3.50	2.96	3.27	3.08
19	3.60	3.24	3.37	3.17	3.26
20	3.06	3.41	3.34	3.39	3.46
Average	3.32	3.24	3.24	3.25	3.28

Average ± SD = 3.26 ± 0.03

Table A4 Size data of KLO-AG macrocapsules which dried at 50˚C at 3 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	2.88	3.15	3.14	3.37	2.97
2	3.56	3.14	3.53	3.66	3.49
3	3.62	3.38	3.68	2.63	3.15
4	3.30	2.81	3.09	2.04	4.05
5	3.03	3.28	3.22	3.63	3.65
6	3.34	3.29	3.71	3.66	3.36
7	3.26	3.78	3.07	3.54	3.28
8	3.01	2.83	3.95	3.77	3.35
9	3.38	2.70	3.31	2.89	3.12
10	3.30	3.29	3.77	3.51	3.02
11	3.55	3.60	3.45	3.35	3.69
12	3.48	3.07	3.79	3.18	3.90
13	3.67	3.08	3.51	3.79	3.38
14	4.06	3.46	3.57	3.28	2.92
15	3.84	3.42	3.54	3.36	3.15
16	3.36	2.72	3.46	3.17	3.51
17	3.40	3.30	3.12	3.36	3.39
18	3.32	3.42	3.16	4.01	3.74
19	3.50	3.06	3.76	3.40	3.35
20	3.67	2.57	3.79	3.54	2.95
Average	3.43	3.17	3.48	3.36	3.37

Average ± SD = 3.36 ± 0.12

Table A5 Size data of KLO-AG macrocapsules which dried at 50˚C at 4 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.47	3.50	3.94	2.97	3.32
2	3.30	3.52	3.55	3.12	3.31
3	3.18	3.48	3.47	3.60	3.63
4	3.07	3.64	3.82	2.72	2.98
5	2.76	3.72	3.09	3.31	4.22
6	3.41	3.37	3.71	3.80	3.33
7	3.14	3.47	3.28	3.72	3.31
8	3.33	3.83	3.43	3.47	3.48
9	3.06	3.34	3.22	3.25	3.06
10	3.34	3.23	3.57	3.17	3.06
11	3.35	2.93	3.38	3.63	3.93
12	3.12	2.88	2.88	3.18	3.16
13	3.30	3.27	3.65	3.47	2.57
14	3.22	3.61	4.01	3.96	3.08
15	3.23	3.51	3.47	2.91	3.81
16	3.05	2.88	3.17	2.98	3.04
17	3.28	3.12	2.92	3.82	2.80
18	3.93	3.30	3.62	3.58	3.40
19	3.00	3.02	3.61	3.53	3.01
20	3.93	3.53	3.49	3.33	3.58
Average	3.27	3.36	3.46	3.38	3.30

Average ± SD = 3.36 ± 0.07

Table A6 Size data of KLO-AG macrocapsules which dried at 50˚C at 5 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.36	3.56	3.79	3.57	3.26
2	3.53	3.45	3.52	3.13	3.66
3	3.24	3.30	3.40	2.96	3.25
4	3.44	3.49	3.36	3.27	3.56
5	3.09	2.94	3.67	3.87	3.34
6	3.33	3.48	3.79	3.84	3.62
7	3.00	3.44	3.17	3.16	3.48
8	3.01	3.09	3.58	3.08	3.82
9	3.24	3.05	3.70	3.42	3.96
10	3.39	3.37	3.21	3.53	3.45
11	3.81	2.94	3.42	3.47	3.13
12	3.49	3.96	3.24	3.16	3.51
13	3.37	3.27	3.74	3.00	3.38
14	3.44	3.20	3.30	2.91	3.16
15	3.83	3.40	3.12	2.70	3.51
16	3.90	3.38	3.38	3.25	3.38
17	3.86	3.22	3.60	3.27	2.98
18	3.32	3.39	3.80	3.54	2.23
19	3.20	3.26	3.43	3.80	2.93
20	3.52	3.56	3.45	3.43	2.84
Average	3.42	3.34	3.48	3.32	3.32

Average ± SD = 3.38 ± 0.07

Part B: Experimental size data of KLO-AGT macrocapsules

Table B1 Size data of KLO-AG macrocapsules which dried at 60˚C at 0 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	5.57	5.68	5.27	5.03	5.51
2	5.41	5.72	5.48	5.15	5.89
3	5.86	5.42	5.31	5.25	5.42
4	5.92	4.73	4.80	5.82	5.15
5	5.01	5.41	5.42	5.61	5.77
6	5.52	5.47	5.72	5.08	5.35
7	5.98	6.11	5.31	4.64	5.87
8	5.28	5.62	4.63	4.93	4.92
9	5.59	4.87	5.89	5.66	5.56
10	5.99	5.14	5.18	5.47	5.71
11	5.46	5.19	6.19	4.75	5.48
12	5.27	5.01	5.86	4.90	5.68
13	6.12	5.37	5.37	5.12	4.97
14	5.48	5.06	5.11	5.06	5.09
15	6.26	5.79	5.51	4.97	5.47
16	5.28	5.41	5.18	5.03	5.89
17	5.59	5.47	6.19	5.15	5.42
18	5.99	6.11	5.86	5.25	5.15
19	5.46	5.62	5.37	5.82	5.77
20	5.27	5.79	5.72	5.61	5.35
Average	5.62	5.45	5.47	5.22	5.47

Average ± SD = 5.44 ± 0.14

Table B2 Size data of KLO-AG macrocapsules which dried at 60˚C at 30 min

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	4.90	4.32	4.88	5.85	5.54
2	4.60	4.25	4.54	4.93	4.67
3	4.79	4.77	4.85	5.30	4.80
4	4.58	5.2	4.25	4.09	5.03
5	4.46	4.35	4.40	5.73	5.28
6	4.74	4.52	5.02	4.73	5.04
7	4.47	4.61	4.96	4.90	4.22
8	4.42	4.44	5.24	5.02	5.13
9	5.14	4.48	4.81	4.71	5.46
10	5.12	4.38	4.10	4.04	5.45
11	4.39	4.46	5.56	4.51	4.58
12	4.42	4.67	4.92	4.70	4.73
13	5.73	4.72	4.81	5.45	5.08
14	5.13	4.53	4.33	4.33	4.67
15	5.03	4.64	4.76	4.97	4.61
16	4.58	4.44	4.40	4.51	4.58
17	4.46	4.48	5.02	4.70	4.73
18	4.74	4.38	4.96	4.71	5.08
19	4.47	4.46	5.24	4.33	4.67
20	4.42	4.67	4.81	4.97	4.61
Average	4.73	4.54	4.79	4.82	4.90

Average ± SD = 4.76 ± 0.14

Table B3 Size data of KLO-AG macrocapsules which dried at 60˚C at 1 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	4.01	4.59	4.85	3.98	4.90
2	4.13	4.41	3.81	4.29	5.06
3	4.32	4.60	4.24	4.93	5.01
4	4.55	4.27	4.88	4.69	4.91
5	4.54	4.18	4.69	4.35	4.57
6	4.20	4.38	4.31	4.46	4.47
7	4.28	4.73	4.18	3.95	4.43
8	4.59	3.69	4.11	4.33	4.78
9	4.92	3.95	4.12	4.50	5.01
10	5.66	4.21	3.91	5.05	4.32
11	4.67	4.79	4.29	4.09	4.54
12	4.27	3.44	4.73	4.26	4.93
13	4.06	4.73	4.78	0.30	5.09
14	5.15	4.69	4.04	3.94	5.03
15	4.28	5.01	3.79	4.01	5.47
16	4.01	4.59	3.91	4.69	5.09
17	4.13	4.41	4.29	4.35	5.13
18	4.32	4.60	4.73	4.46	4.97
19	4.06	4.27	3.79	3.95	4.57
20	4.20	4.18	4.04	4.03	4.37
Average	4.42	4.39	4.27	4.13	4.83

Average ± SD = 4.41 ± 0.26

Table B4 Size data of KLO-AG macrocapsules which dried at 60°C at 2 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.60	3.78	3.92	4.00	4.01
2	3.79	3.64	3.70	3.60	3.56
3	4.05	3.96	4.08	3.77	3.62
4	4.18	3.82	3.53	3.73	3.70
5	3.75	3.89	3.51	3.85	3.79
6	4.17	4.23	3.71	3.91	3.83
7	3.60	3.58	3.37	4.10	3.98
8	4.00	4.29	3.76	3.55	4.33
9	3.94	3.87	3.92	4.30	3.70
10	3.71	3.68	4.26	3.89	3.04
11	3.89	4.29	3.80	3.66	3.54
12	3.71	3.96	3.80	4.19	3.79
13	4.13	3.45	4.04	3.99	3.56
14	3.92	4.07	3.73	3.89	4.09
15	4.05	3.78	3.99	3.79	4.13
16	3.60	3.58	3.51	3.77	3.62
17	3.79	4.29	3.71	3.73	3.70
18	4.05	3.87	3.37	3.85	3.79
19	4.18	3.68	3.76	3.91	3.83
20	3.75	4.29	3.92	4.10	3.98
Average	3.89	3.90	3.77	3.88	3.78

Average ± SD = 3.84 ± 0.06

Table B5 Size data of KLO-AG macrocapsules which dried at 60˚C at 3 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	4.02	3.50	3.75	3.77	3.73
2	3.90	4.02	4.16	3.72	4.08
3	4.04	3.39	3.03	4.05	3.82
4	3.73	3.75	3.50	3.89	4.60
5	4.12	4.01	4.63	4.08	3.38
6	3.96	4.06	3.73	3.80	4.06
7	4.01	4.13	3.71	3.84	3.79
8	3.39	4.07	3.88	4.03	4.16
9	3.77	3.87	3.79	3.70	2.99
10	4.39	3.64	3.43	3.79	3.83
11	3.71	3.75	3.49	2.91	4.09
12	3.48	3.57	3.68	3.70	3.79
13	4.11	3.82	3.69	3.03	3.75
14	3.79	3.37	3.24	3.75	4.12
15	3.79	3.87	3.91	4.08	3.77
16	4.12	3.47	3.75	4.05	3.82
17	3.96	3.64	4.06	3.89	3.79
18	4.01	3.75	3.03	4.08	3.38
19	3.39	3.67	3.50	3.80	4.06
20	4.21	3.87	3.79	3.84	3.79
Average	3.90	3.76	3.69	3.79	3.84

Average ± SD = 3.79 ± 0.08

Table B6 Size data of KLO-AG macrocapsules which dried at 60°C at 4 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.65	3.69	3.78	3.92	3.99
2	3.80	4.03	3.77	4.18	3.42
3	3.85	4.01	3.84	3.81	3.46
4	3.92	3.60	3.81	3.95	3.86
5	3.80	3.90	3.63	3.91	3.12
6	3.75	3.73	3.66	3.95	3.55
7	3.50	3.83	4.46	3.95	3.62
8	4.07	3.75	3.62	3.68	3.83
9	3.52	3.85	3.44	3.82	4.11
10	3.59	3.86	3.39	3.81	3.64
11	3.58	3.69	3.90	4.00	3.87
12	4.06	4.04	3.90	3.55	3.86
13	3.58	4.02	3.79	3.11	3.98
14	3.60	3.87	3.35	3.67	3.91
15	3.92	4.14	3.40	3.27	3.81
16	3.81	3.67	3.59	3.49	3.62
17	3.64	3.49	3.49	3.69	3.79
18	3.71	3.57	3.67	3.71	3.81
19	3.54	3.59	3.57	3.59	3.90
20	3.59	3.67	3.71	3.67	3.61
Average	3.72	3.80	3.69	3.74	3.74

Average ± SD = 3.74 ± 0.04

Table B7 Size data of KLO-AG macrocapsules which dried at 60˚C at 5 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.59	3.85	3.68	3.53	3.76
2	3.77	3.84	3.83	3.87	3.95
3	3.72	3.67	4.08	4.05	3.60
4	4.02	3.63	3.74	3.92	3.70
5	3.37	3.43	3.83	3.28	4.10
6	3.69	3.91	3.77	3.58	3.79
7	3.46	4.17	3.50	4.25	3.69
8	3.85	3.58	4.14	3.60	3.49
9	3.97	3.56	3.80	3.65	3.59
10	3.21	3.81	3.87	3.78	3.60
11	4.12	3.95	3.65	3.83	4.08
12	3.86	3.85	3.30	3.77	3.88
13	3.56	3.55	3.65	3.72	3.86
14	3.54	3.84	3.78	3.82	3.83
15	3.53	3.54	3.59	3.53	3.84
16	3.67	3.37	3.67	3.59	3.59
17	3.52	3.54	3.34	3.67	3.67
18	3.61	3.56	3.56	3.52	3.39
19	3.51	3.67	3.27	3.59	3.71
20	5.67	3.51	3.67	3.39	3.52
Average	3.76	3.69	3.69	3.70	3.73

Average ± SD = 3.71 ± 0.03

Table C Weight data of KLO-AG macrocapsules which dried at 50˚C at 0,1,2,3,4,5, and 10 h

Time (hours)	Weight (g) of set no.						
	1	2	3	4	5	AV	%WL
0	2.7718	2.6897	2.7939	2.6977	2.644	2.7194	0.0
1	1.171	0.898	1.0575	1.0414	0.8888	1.0113	62.8
2	0.5242	0.4022	0.4842	0.3987	0.4501	0.4519	83.4
3	0.4651	0.3486	0.4319	0.3735	0.4108	0.4060	85.1
4	0.4404	0.2999	0.3916	0.3641	0.4221	0.3836	85.9
5	0.4115	0.2494	0.3312	0.3895	0.4009	0.3565	86.9
10	0.4101	0.2449	0.3213	0.381	0.3978	0.3510	87.1

Table D Weight data of KLO-AG macrocapsules which dried at 60˚C at 0,1,2,3,4,5, and 10 h.

Time (hours)	Weight (g) of set no.						
	1	2	3	4	5	AV	%WL
0	2.8407	2.7478	2.7374	2.6828	2.8983	2.7814	0.0
1	1.378	1.1114	0.973	0.9683	1.2257	1.1313	59.3
2	0.6175	0.593	0.5887	0.5481	0.6222	0.5939	78.6
3	0.5886	0.5526	0.5089	0.524	0.5117	0.5372	80.7
4	0.5656	0.5221	0.4615	0.498	0.4475	0.4989	82.1
5	0.5403	0.4961	0.4363	0.4755	0.418	0.4732	83.0
10	0.5419	0.5008	0.4051	0.4626	0.407	0.4635	83.3

Table E1 Size data of KLO-AGT macrocapsules which dried at 50°C at 0 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	5.80	5.78	5.32	6.04	4.93
2	6.09	5.18	5.33	5.41	5.43
3	5.41	5.27	5.90	6.18	5.87
4	4.94	6.35	5.15	5.05	5.69
5	5.15	6.38	5.12	5.87	5.12
6	5.55	6.21	6.43	5.69	5.53
7	5.23	6.14	5.12	5.12	5.83
8	5.07	5.51	6.43	5.53	5.60
9	5.92	5.59	5.59	5.83	5.97
10	5.86	5.53	6.10	5.97	5.22
11	5.59	6.72	5.16	5.17	5.81
12	5.47	5.12	5.45	5.43	5.72
13	4.94	6.43	5.61	4.90	5.64
14	5.66	6.82	5.45	5.32	5.40
15	5.56	5.32	5.36	5.53	5.22
16	5.69	6.21	5.29	5.87	5.87
17	5.07	6.14	5.12	5.69	5.69
18	5.92	5.51	6.43	5.12	5.12
19	5.86	5.12	6.43	5.53	5.53
20	5.16	6.43	5.59	5.83	5.83
Average	5.50	5.89	5.62	5.55	5.55

Average ± SD = 5.62 ± 0.15

Table E2 Size data of KLO-AGT macrocapsules which dried at 50°C at 1 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.86	4.30	5.11	4.14	4.88
2	4.71	5.19	4.54	3.66	4.67
3	4.38	3.81	4.60	4.16	4.16
4	3.54	4.98	4.86	5.10	4.56
5	3.53	4.36	4.27	3.09	4.97
6	4.04	5.20	5.17	4.10	4.49
7	3.65	5.50	3.99	4.73	5.24
8	4.48	4.06	4.55	5.69	5.01
9	3.76	5.14	4.06	5.44	5.02
10	4.69	5.03	3.99	4.18	4.49
11	4.22	4.10	4.52	3.74	3.86
12	4.44	5.96	3.61	4.42	4.23
13	4.45	4.44	3.80	4.05	4.96
14	4.25	4.43	4.11	4.95	4.06
15	4.06	3.94	3.59	5.36	4.45
16	4.04	5.20	5.17	4.10	4.49
17	3.65	5.50	3.99	4.73	5.24
18	4.48	4.06	4.55	5.69	5.01
19	3.76	5.14	4.06	5.44	5.02
20	4.69	5.03	3.99	4.18	4.49
Average	4.13	4.77	4.33	4.55	4.67

Average ± SD = 4.49 ± 0.26

Table E3 Size data of KLO-AGT macrocapsules which dried at 50°C at 2 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.65	4.09	3.27	3.28	3.36
2	3.88	3.40	2.95	3.67	3.61
3	3.72	3.85	3.18	3.53	3.56
4	3.56	3.45	3.50	3.79	3.66
5	3.46	4.22	3.34	3.19	2.88
6	3.57	4.24	3.00	3.22	3.55
7	3.26	3.46	3.20	3.56	3.36
8	3.30	2.96	3.51	3.24	2.87
9	3.48	3.91	3.08	3.24	3.64
10	3.25	3.33	3.96	3.89	3.17
11	3.79	3.51	3.39	3.48	3.46
12	3.27	3.90	3.32	3.91	3.83
13	3.01	2.74	3.38	3.35	4.24
14	3.61	3.47	3.34	4.01	3.49
15	3.48	3.83	3.70	3.49	3.68
16	3.88	3.40	3.87	3.53	3.56
17	3.72	3.85	3.75	3.79	3.66
18	3.98	3.98	3.34	3.98	3.97
19	3.68	4.22	3.68	3.69	3.55
20	3.98	4.24	3.87	3.56	3.89
Average	3.58	3.70	3.43	3.57	3.55

Average ± SD = 3.57 ± 0.10

Table E4 Size data of KLO-AGT macrocapsules which dried at 50°C at 3 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.56	3.56	2.81	3.23	3.64
2	3.67	3.45	3.70	3.55	3.80
3	3.41	3.63	3.69	3.51	3.91
4	3.36	3.24	3.16	3.80	3.75
5	3.65	2.95	3.35	3.39	3.83
6	3.35	3.32	3.45	4.33	3.64
7	3.52	3.66	3.56	3.56	3.51
8	3.56	3.46	3.74	3.62	3.65
9	3.32	3.52	3.36	3.41	3.16
10	3.46	3.43	3.59	3.20	3.46
11	3.72	3.49	3.59	3.03	3.50
12	3.78	3.38	3.15	3.59	3.21
13	3.52	3.35	4.51	3.54	3.63
14	3.43	3.64	3.29	3.33	2.94
15	3.65	3.32	3.17	3.19	3.51
16	3.65	2.95	3.35	3.39	3.83
17	3.35	3.32	3.45	4.33	3.64
18	3.52	3.66	3.56	3.56	3.51
19	3.56	3.46	3.74	3.62	3.65
20	3.32	3.52	3.36	3.41	3.16
Average	3.52	3.42	3.48	3.53	3.55

Average ± SD = 3.50 ± 0.05

Table E5 Size data of KLO-AGT macrocapsules which dried at 50°C at 4 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.52	2.78	3.31	3.46	3.69
2	3.32	3.29	3.76	3.08	3.34
3	3.69	3.41	3.50	3.45	3.35
4	3.46	3.97	3.34	3.14	3.05
5	3.13	3.49	3.40	3.01	3.27
6	4.01	3.75	2.89	3.52	3.32
7	3.38	3.74	3.34	3.49	3.12
8	2.92	3.19	3.64	3.16	3.41
9	3.56	3.27	3.96	3.22	3.28
10	3.55	3.46	3.66	3.47	3.83
11	3.44	3.36	3.46	3.80	3.24
12	3.09	3.57	3.47	2.07	3.11
13	3.43	3.29	3.13	3.90	3.18
14	2.95	3.24	3.81	4.18	4.16
15	3.74	3.40	3.45	3.49	3.46
16	3.79	3.49	3.40	3.01	3.69
17	4.01	3.75	3.69	3.52	3.52
18	3.59	3.74	3.69	3.68	3.62
19	3.69	3.69	3.64	3.56	3.61
20	3.56	3.59	3.96	3.68	3.69
Average	3.49	3.47	3.53	3.39	3.45

Average ± SD = 3.47 ± 0.05

Table E6 Size data of KLO-AGT macrocapsules which dried at 50°C at 5 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.73	3.20	4.50	3.53	3.36
2	3.36	2.63	3.56	3.44	3.47
3	3.59	3.77	3.88	2.46	3.35
4	3.31	3.58	3.31	3.58	3.78
5	3.28	3.36	3.16	3.16	2.92
6	3.39	3.56	3.82	3.84	3.32
7	3.61	3.42	3.81	2.98	3.44
8	2.74	3.23	3.81	3.13	2.92
9	3.26	3.49	3.05	3.92	3.58
10	2.97	3.57	3.12	3.01	3.83
11	3.62	3.26	3.59	3.32	3.75
12	2.74	3.66	3.36	3.21	3.26
13	3.35	3.68	3.02	3.48	3.39
14	3.49	3.37	3.12	3.48	3.53
15	3.27	3.49	3.51	3.42	2.98
16	3.62	3.66	3.59	3.42	3.75
17	3.74	3.66	3.36	3.61	3.76
18	3.35	3.68	3.62	3.78	3.79
19	3.49	3.67	3.12	3.78	3.53
20	3.67	3.49	3.51	3.62	3.67
Average	3.38	3.47	3.49	3.41	3.47

Average ± SD = 3.44 ± 0.05

Table F1 Size data of KLO-AGT macrocapsules which dried at 60°C at 0 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	5.73	5.11	6.01	7.22	5.75
2	6.61	5.84	5.83	4.70	5.24
3	6.31	5.90	6.04	5.58	6.35
4	6.00	5.56	5.52	6.06	6.02
5	5.69	5.89	5.69	5.84	5.60
6	5.90	5.09	5.44	6.35	5.26
7	5.24	5.40	5.98	6.05	5.36
8	4.75	6.49	5.92	5.25	5.62
9	5.82	6.41	6.40	6.33	5.75
10	6.33	5.96	6.08	6.01	5.33
11	5.97	5.28	6.24	6.58	5.57
12	3.53	6.05	5.86	5.95	5.28
13	5.60	5.72	6.17	6.39	5.86
14	4.53	5.94	6.26	4.46	6.39
15	4.87	6.23	5.72	5.26	5.11
16	5.24	5.40	5.98	6.05	5.36
17	4.75	6.49	5.92	5.25	5.62
18	5.82	6.41	6.40	6.33	5.75
19	6.33	5.96	6.08	6.01	5.33
20	5.97	5.28	6.24	6.58	5.57
Average	5.55	5.82	5.99	5.91	5.61

Average ± SD = 5.78 ± 0.19

Table F2 Size data of KLO-AGT macrocapsules which dried at 60°C at 1 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	4.98	4.31	3.32	3.38	4.67
2	3.22	4.30	3.98	3.74	4.05
3	4.53	4.46	4.25	4.20	3.89
4	4.49	3.99	4.88	4.66	4.88
5	3.90	3.97	4.49	4.79	4.00
6	4.60	3.53	3.31	4.07	3.80
7	4.56	3.88	4.16	3.43	4.10
8	4.59	3.86	3.60	4.21	3.99
9	4.42	3.99	3.71	4.85	3.78
10	4.55	4.73	4.42	3.36	3.79
11	3.97	3.97	3.63	4.57	4.36
12	4.79	3.37	4.62	4.97	4.52
13	4.25	4.38	4.89	4.28	3.02
14	4.80	3.75	3.47	4.91	4.02
15	5.38	4.59	3.48	4.81	4.45
16	4.56	3.88	4.16	3.43	4.10
17	4.59	3.86	4.23	4.37	4.09
18	4.42	3.99	3.71	4.85	3.78
19	4.55	4.73	4.42	4.38	4.29
20	4.56	4.78	4.49	4.57	4.36
Average	4.49	4.12	4.06	4.29	4.10

Average ± SD = 4.21 ± 0.18

Table F3 Size data of KLO-AGT macrocapsules which dried at 60°C at 2 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.31	3.26	3.93	3.66	3.92
2	3.24	3.26	3.70	3.49	3.75
3	3.16	3.01	3.66	3.57	3.94
4	2.98	3.24	3.88	3.84	3.59
5	3.74	3.88	3.43	3.76	3.50
6	3.52	3.81	3.47	4.19	3.44
7	3.53	3.36	3.78	3.77	3.68
8	3.79	4.12	4.49	3.48	3.79
9	2.69	3.56	3.34	3.14	3.40
10	3.18	3.10	3.62	3.96	2.82
11	3.29	3.41	3.78	3.90	3.17
12	3.69	3.99	3.56	4.22	3.13
13	3.30	3.24	3.82	3.22	3.31
14	3.45	3.81	3.51	3.23	3.36
15	3.62	3.15	3.66	3.90	3.07
16	3.74	3.88	3.43	3.76	3.50
17	3.52	3.81	3.47	3.74	3.44
18	3.53	3.36	3.78	3.77	3.68
19	3.79	3.97	4.49	3.48	3.79
20	2.69	3.56	3.34	3.14	3.40
Average	3.39	3.54	3.71	3.66	3.48

Average ± SD = 3.56 ± 0.13

Table F4 Size data of KLO-AGT macrocapsules which dried at 60°C at 3 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.06	3.75	3.47	3.50	4.07
2	3.17	3.38	3.58	3.45	3.40
3	3.25	4.49	3.74	3.56	3.34
4	2.99	2.65	3.88	3.85	3.58
5	3.44	3.69	3.80	4.04	3.61
6	3.54	4.21	3.95	3.06	3.24
7	3.34	3.32	3.51	3.17	3.25
8	3.84	3.57	3.31	3.86	3.33
9	3.20	3.39	2.76	3.62	3.24
10	3.28	3.24	3.69	3.26	3.50
11	2.63	3.66	4.62	3.70	2.71
12	3.29	3.73	4.51	3.50	3.59
13	3.65	2.95	3.62	3.12	2.98
14	3.66	3.25	3.57	3.87	3.28
15	3.54	3.57	3.34	3.95	3.54
16	3.44	3.69	3.80	4.04	3.61
17	3.54	4.21	3.95	3.06	3.24
18	3.34	3.32	3.51	3.17	3.25
19	3.84	3.57	3.31	3.86	3.33
20	3.20	3.39	2.76	3.62	3.24
Average	3.36	3.55	3.63	3.56	3.37

Average ± SD = 3.50 ± 0.12

Table F5 Size data of KLO-AGT macrocapsules which dried at 60°C at 4 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	2.91	3.56	4.16	3.50	3.53
2	3.61	3.96	3.64	3.42	3.84
3	3.85	3.75	3.57	3.82	3.31
4	3.29	3.34	3.30	3.25	3.67
5	3.07	3.72	3.64	3.21	3.23
6	3.44	3.00	3.42	3.36	3.44
7	3.39	3.18	3.06	3.12	3.59
8	3.91	4.03	3.24	3.40	3.56
9	3.68	3.15	3.68	3.28	3.59
10	2.62	3.36	3.41	3.73	3.30
11	3.46	3.57	3.36	3.30	3.50
12	2.79	3.28	3.25	3.33	3.02
13	3.43	3.85	3.63	3.02	3.87
14	3.28	3.04	2.79	3.32	3.83
15	2.89	3.15	3.68	3.36	3.72
16	3.39	3.18	3.06	3.12	3.59
17	3.91	4.03	3.24	3.40	3.56
18	3.68	3.15	3.68	3.28	3.59
19	3.69	3.36	3.41	3.73	3.69
20	3.46	3.57	3.36	3.74	3.50
Average	3.39	3.46	3.43	3.38	3.55

Average ± SD = 3.44 ± 0.07

Table F6 Size data of KLO-AGT macrocapsules which dried at 60°C at 5 h

Samples	Size (mm) of set no.				
	1	2	3	4	5
1	3.89	3.17	3.41	3.98	3.63
2	3.43	3.35	3.60	3.32	3.91
3	3.90	3.72	3.95	3.14	3.46
4	3.44	3.44	2.95	3.39	3.33
5	3.31	3.18	3.40	3.71	2.96
6	3.08	3.09	4.26	3.93	3.05
7	3.47	3.33	3.69	3.55	2.96
8	3.74	2.96	2.76	3.30	3.05
9	3.20	3.45	3.72	3.57	3.93
10	3.80	3.22	3.49	3.39	3.18
11	3.53	4.16	3.30	4.03	3.68
12	3.07	2.92	3.58	3.43	3.39
13	3.18	3.41	3.72	3.45	3.07
14	3.49	4.10	3.47	3.31	3.41
15	3.31	3.64	3.34	3.51	3.74
16	3.20	3.45	3.52	3.57	3.41
17	3.61	3.22	3.49	3.39	2.92
18	3.53	3.59	3.30	3.43	3.05
19	3.07	2.92	3.21	3.43	3.43
20	3.18	3.41	3.37	3.45	3.18
Average	3.42	3.39	3.48	3.51	3.34

Average ± SD = 3.43 ± 0.07

Table G Weight data of KLO-AGT macrocapsules which dried at 50°C in the time interval of 0,1,2,3,4,5, and 10 h

Time (hours)	Weight (g) of set no.						
	1	2	3	4	5	AV	%WL
0	2.3673	2.457	2.2169	2.3056	2.2478	2.31892	0
1	1.0947	1.1629	0.7643	1.2097	1.1399	1.0743	53.6724
2	0.452	0.5424	0.4026	0.4968	0.5001	0.47878	79.35332
3	0.3987	0.4379	0.3674	0.4125	0.432	0.4097	82.33229
4	0.3769	0.4024	0.3476	0.3862	0.4184	0.3863	83.34138
5	0.3532	0.3686	0.3315	0.3787	0.4041	0.36722	84.16418
10	0.352	0.365	0.3089	0.3781	0.3787	0.35654	84.62474

Table H Weight data of KLO-AGT macrocapsules which dried at 60°C in the time interval of 0,1,2,3,4,5, and 10 h

Time (hours)	Weight (g) of set no.						
	1	2	3	4	5	AV	%WL
0	2.715	2.6414	2.7764	2.6206	2.6835	2.68738	0
1	1.1895	0.9031	0.7854	0.9453	0.9357	0.9518	64.5826
2	0.4122	0.4107	0.4519	0.4442	0.4156	0.42692	84.1139
3	0.3641	0.3503	0.3654	0.3524	0.3229	0.35102	86.93821
4	0.3162	0.3127	0.3307	0.3256	0.3077	0.31858	88.14533
5	0.2982	0.2931	0.3149	0.3072	0.2988	0.30244	88.74592
10	0.2974	0.2901	0.315	0.2778	0.2808	0.29222	89.12621

Table I1 Size data of chitosan-coated KLO-AGT macrocapsules which dried at 50˚C at 10 h

Samples	Size (mm) of set no.			
	B 0.5	B1	B2	B3
1	3.6	4.2	3.62	3.94
2	3.25	3.73	3.77	3.38
3	3.7	3.83	3.08	3.52
4	3.5	3.73	3.34	3.96
5	3.81	3.63	3.2	3.81
6	3.88	3.81	2.83	3.51
7	3.86	3.86	2.9	3.38
8	3.69	3.94	3.88	3.57
9	3.46	3.64	3.44	3.5
10	3.25	3.59	3.37	3.5
11	3.83	3.78	3.41	3.41
12	2.88	3.67	3.57	3.56
13	3.41	3.9	3.56	3.41
14	3.5	3.28	3.49	3.33
15	3.36	2.86	3.28	3.76
Average	3.532	3.6966667	3.3826667	3.5693333

Average ± SD = 3.55 ± 0.12

Table I2 Compression data of chitosan-coated KLO-AGT macrocapsules which dried at 50˚C at 10 h

Formula	Max Force Average (kg)	SD max force
B0.5	0.665729	0.46632
B1	0.800519	0.379554
B2	0.799416	0.346238
B3	0.837291	0.392032

Experimental Data

3.1 Size and standard deviation data of KLO-AG and KLO-AGT macrocapsules at 50˚C and 60˚C

Temp (˚C)	Size data of AG					
	0	1	2	3	4	5
50˚C	5.46 ± 0.24	3.65 ± 0.15	3.26 ± 0.03	3.36 ± 0.12	3.36 ± 0.07	3.38 ± 0.07
60˚C	5.44 ± 0.14	4.41 ± 0.26	3.84 ± 0.06	3.79 ± 0.08	3.74 ± 0.04	3.71 ± 0.03

Temp (˚C)	Size data of AGT					
	0	1	2	3	4	5
50˚C	5.62 ± 0.15	4.49 ± 0.26	3.57 ± 0.10	3.5 ± 0.05	3.47 ± 0.05	3.44 ± 0.05
60˚C	5.78 ± 0.19	4.21 ± 0.18	3.56 ± 0.13	3.5 ± 0.12	3.44 ± 0.07	3.43 ± 0.07

3.2 %Weight loss data of KLO-AG and KLO-AGT macrocapsules at 50˚C and 60˚C

Temp (˚C)	%Weightloss of AG						
	0	1	2	3	4	5	10
50˚C	0	62.8	83.4	85.1	85.9	86.9	87.1
60˚C	0	59.3	47.5	9.6	7.1	5.2	2.1

Temp (˚C)	%Weightloss of AGT						
	0	1	2	3	4	5	10
50˚C	0	53.7	55.4	14.4	5.7	4.9	2.9
60˚C	0	64.6	55.1	17.8	9.2	5.1	3.4

3.3 %Weight loss data of chitosan-coated KLO-AGT macrocapsules at 50˚C

Time	%Weight loss of capsules			
(days)	B 0.5	B 1	B 2	B 3
0	0	0	0	0
5	2.420809	3.165109	2.81527	1.40067
10	4.056142	5.425901	5.020371	3.0856
15	5.335437	6.894124	5.763129	2.763311
20	6.047515	9.025728	9.195646	5.466031
25	6.111898	8.508975	8.449027	3.888746
30	8.992403	11.42863	13.91544	9.29483

GCMS Data of chitosan-coated KLO-AGT macrocapsules

0day

	1	2	3	4	Average	SD
Oil	2.20E+09	2.15E+09	2.00E+09	2.02E+09	2.09E+09	97766729.17
C-0.5	1.18E+09	1.83E+09	1.89E+09	1.18E+09	1.52E+09	393361580.9
C-1	2.23E+09	1.28E+09	1.03E+09	1.28E+09	1.46E+09	529937103.2
C-2	8.89E+09	8.91E+09	7.48E+08	1.08E+09	4.91E+09	4612718215
C-3	3.09E+08	3.86E+08	4.20E+08	5.56E+08	4.18E+08	103203278.4

5days

	1	2	3	4	Average	SD
Oil	1.91E+09	2.03E+09	1.98E+09	1.91E+09	1.96E+09	58523499.55
C-0.5	2.58E+08	2.80E+08	1.52E+08	2.99E+08	2.47E+08	65672800.05
C-1	4.31E+08	3.58E+08	4.08E+08	5.01E+08	4.25E+08	59410997.86
C-2	2.97E+08	9.27E+07	2.05E+08	2.01E+08	1.99E+08	83553710.27
C-3	2.30E+08	4.64E+08	7.36E+08	2.37E+08	4.17E+08	238983088.7

15 days

	1	2	3	4	Average	SD
Oil	2.48E+09	2.48E+09	2.25E+09	2.28E+09	2.37E+09	124733048.3
C-0.5	2.65E+08	8.88E+08	3.49E+08	4.15E+08	4.79E+08	279328212
C-1	9.22E+08	5.42E+07	1.26E+08	9.30E+07	2.99E+08	416501652.7
C-2	1.05E+08	5.37E+07	2.99E+08	1.10E+08	1.42E+08	107763425.3
C-3	3.43E+07	7.30E+06	7.29E+07	1.93E+08	7.69E+07	81963665.73

20days

	1	2	3	4	Average	SD
Oil	2.20E+09	2.23E+09	2.15E+09	2.16E+09	2.19E+09	36968455.02
C-0.5	1.43E+08	1.54E+08	1.10E+08	2.32E+08	1.60E+08	51668010.7
C-1	1.12E+08	1.34E+08	3.67E+07	6.52E+07	8.70E+07	44118505.19
C-2	6.42E+07	6.34E+07	4.17E+07	2.87E+07	4.95E+07	17347238.01
C-3	1.62E+07	1.59E+08	3.34E+08	3.47E+08	2.14E+08	157309493.3

Compression test data of chitosan coated KLO-AGT macrocapsules

Formula	Max Force Average (kg)	SD max force	
B0.5	0.665729	0.46632	339.7318
B1	0.800519	0.379554	372.9335
B2	0.799416	0.346238	444.7692
B3	0.837291	0.392032	418.3908

Shell thickness data of chitosan coated KLO-AGT macrocapsules

Formula	Average	SD
B0.5	95.0826	36.81674
B1	97.3276	33.10125
B2	94.09027	30.72333
B3	64.95767	15.40551

Table 3.7 The maximum peak area of chitosan-coated KLO-AGT macrocapsules during the profile release

0day

	1	2	3	4	Average	SD
Oil	2.20E+09	2.15E+09	2.00E+09	2.02E+09	2.09E+09	97766729.17
C-0.5	1.18E+09	1.83E+09	1.89E+09	1.18E+09	1.52E+09	393361580.9
C-1	2.23E+09	1.28E+09	1.03E+09	1.28E+09	1.46E+09	529937103.2
C-2	8.89E+09	8.91E+09	7.48E+08	1.08E+09	4.91E+09	4612718215
C-3	3.09E+08	3.86E+08	4.20E+08	5.56E+08	4.18E+08	103203278.4

5days

	1	2	3	4	Average	SD
Oil	1.91E+09	2.03E+09	1.98E+09	1.91E+09	1.96E+09	58523499.55
C-0.5	2.58E+08	2.80E+08	1.52E+08	2.99E+08	2.47E+08	65672800.05
C-1	4.31E+08	3.58E+08	4.08E+08	5.01E+08	4.25E+08	59410997.86
C-2	2.97E+08	9.27E+07	2.05E+08	2.01E+08	1.99E+08	83553710.27
C-3	2.30E+08	4.64E+08	7.36E+08	2.37E+08	4.17E+08	238983088.7

15days

	1	2	3	4	Average	SD
Oil	2.48E+09	2.48E+09	2.25E+09	2.28E+09	2.37E+09	124733048.3
C-0.5	2.65E+08	8.88E+08	3.49E+08	4.15E+08	4.79E+08	279328212
C-1	9.22E+08	5.42E+07	1.26E+08	9.30E+07	2.99E+08	416501652.7
C-2	1.05E+08	5.37E+07	2.99E+08	1.10E+08	1.42E+08	107763425.3
C-3	3.43E+07	7.30E+06	7.29E+07	1.93E+08	7.69E+07	81963665.73

20days

	1	2	3	4	Average	SD
Oil	2.20E+09	2.23E+09	2.15E+09	2.16E+09	2.19E+09	36968455.02
C-0.5	1.43E+08	1.54E+08	1.10E+08	2.32E+08	1.60E+08	51668010.74
C-1	1.12E+08	1.34E+08	3.67E+07	6.52E+07	8.70E+07	44118505.19
C-2	6.42E+07	6.34E+07	4.17E+07	2.87E+07	4.95E+07	17347238.01
C-3	1.62E+07	1.59E+08	3.34E+08	3.47E+08	2.14E+08	157309493.3

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