

LSP18

Comparative study on physical properties of κ -carrageenan extracted from *Eucheumacottonii* in Tawau, Sabah and commercial κ -carrageenan

S.W Chan¹, S.H. Mirhosseini¹, S.T. Farah², C.P. Tan^{1,*}

¹Department of Food Technology, Faculty of Food Science and Technology, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia

²Department of Process and Food Engineering, Faculty of Engineering, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia.

*Corresponding author's email: tancp@putra.upm.edu.my

Keywords: *Eucheumacottonii*, κ -carrageenan, physical properties, syneresis, and water holding capacity

Introduction. *Kappaphycusalvarezii*, or commercially known as *Eucheumacottonii* are a group of red algae that mainly cultivated on the inner side of coral reefs around Philippines, Indonesia and other island coasts in the East Africa [6,15]. They are the main source of κ -carrageenan. The extract of *Eucheumacottonii* contains solely κ -carrageenan with less than 10% of ι -carrageenan [10]. Its high content of κ -carrageenan makes it one of the economically important red seaweed [10]. The world production of *Eucheumacottonii* for commercial κ -carrageenan extraction showed that Southeast Asian region contributed 96.5% of the total production, of which 55% comes from the Philippines, followed by Indonesia (38%) and Malaysia (2.5%) [5]. Yearly, Southern Philippines and Malaysia produces about 100,000 tonnes of *Eucheumacottonii*.

κ -carrageenan is a linear polysaccharide comprising of repeating galactose units and 3,6-anhydrogalactose (3,6 AG), which joined alternatively with α -(1,3) and β -(1,4) D-glycosidic linkages as shown in Fig. 1 [6,16,17]. In food industry, κ -carrageenan is typically used as gelling, thickening, stabilizing, and water-binding agents in various food products such as instant products, dessert, sauces, milk, yogurt, and meats [2,12]. More recently, κ -carrageenan was explored for its potential use as controlled release carrier in the encapsulation system [3,9]. κ -carrageenan encapsulant can exist in many forms such as matrix, bead, microcapsule, and microgel.

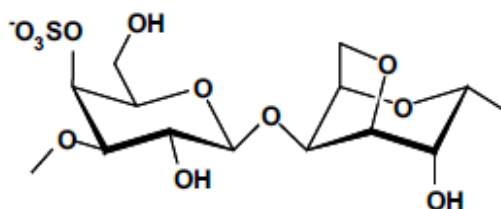


Fig. 1. Basic structure of κ -carrageenan repeating unit [13]

Due to its wide range of application, carrageenan industry has been growing rapidly during the past 15 years at an annual rate of 8% [4]. Carrageenan industry represents a multi-million dollar market worldwide that grows continuously every year. Malaysia has taken great interest on *Eucheumacottonii*, the κ -carrageenan producing red seaweed species. In Malaysia, *Eucheumacottonii* is largely cultivated in east coast of Sabah since the last three decades [10]. In the early 1970, *Eucheumacottonii* was manufactured as pet canned food. By the early 1980, the quality of alkali

treated flour had been improved on colour, odour, and taste, which it is exported as food grade κ -carrageenan [10]. The first food grade natural κ -carrageenan in Malaysia was produced in Tawau, Sabah. Tawau, which is located near to the world largest seaweed producing areas (Tawi-Tawi) at Philippines is a suitable place for *Eucheumacottonii* cultivation. κ -carrageenan extracted from *Eucheumacottonii*, Tawau may be a good substitute for imported commercial κ -carrageenan.

To the best of our knowledge very little information available on κ -carrageenan isolated from *Eucheumacottonii*, Tawau, Sabah. Therefore, a preliminary study was conducted to examine the physical properties of κ -carrageenan extracted from *Eucheumacottonii* in Tawau, Sabah and to compare with the commercial κ -carrageenan. This information is essential for better application κ -carrageenan from Sabah in food industry.

Materials and Methods

Materials. κ -carrageenan samples used in this study were obtained as gift from the following suppliers: TA150 (Tacara Sdn. Bhd., Tawau, Malaysia), SeaKem CM611, Gelcarin GP812, Gelcarin GP911 NF (FMC Biopolymer, Philadelphia PA, USA), and Grindsted[®] carrageenan CL220 (Danisco, Copenhagen K, Denmark). All the solvents and chemicals used for the analyses were of analytical grade. Deionized water used for the preparation of all the solutions was purified by Sartorius Atrium[®] 611D1 (Sartorius Stedim Biotech, Germany).

Physical analysis. The κ -carrageenan samples were stored in a cool and dark place before the physical analysis. All the results were reported as the average of triplicate measurements (mean \pm standard deviation).

Colour. The colour of κ -carrageenan samples powder were measured using HunterLab UltraScan PRO colorimeter (Hunter Associate Laboratory Inc., Reston, USA) and values were expressed as L^* , a^* , and b^* .

Particle size. Particle size of κ -carrageenan samples were determined by using Malvern Mastersizer (Malvern Instruments Ltd, UK) and were reported in Sauter Diameter D[3,2].

pH. 1.5% w/v κ -carrageenan solutions were prepared by dispersing 1.5g of respective κ -carrageenan sample powders into a hot (80°C) deionized water. pH meter was then used to measure the pH values of the κ -carrageenan solutions.

Texture. κ -carrageenan solutions of 2.0-4.0% w/v were prepared for all the samples except SeaKem CM611. For SeaKem CM611, solutions of 4.0-6.0% w/v were prepared. The samples were allowed to set in the containers to form a gel of size 30mm x 30mm. Gel samples were stored overnight at room temperature (25°C) prior to measurement. The TA.XT2i texture analyser (Stable Micro Systems, New York, USA) was used to determine the rupture strength and brittleness of the gel samples. A cylindrical probe of 0.5 inches diameter (P/0.5) was used. The cylindrical gel samples underwent compression under a 5kg load cell at a deformation rate of 20 mm/min.

Syneresis. 4% κ -carrageenan solutions were prepared and the gels were set inside petri dish and were cut into a 30mm x 8mm gel. The cylindrical gel formed was stored overnight in a container at room temperature (25°C), 4°C, and -18°C for 2, 4, 6, 8, and 10 days. Initial weight of the gel sample (W_0) and weight of the empty container were recorded. At certain time intervals (days), the water condensed on the container walls was removed with tissue paper. After water removal, the weight of gel was measured (W_t). The syneresis of the gel was calculated as:

$$\text{syneresis (\%)} = [(W_0 - W_t) / W_0] \times 100. \quad (1)$$

where W_0 is the initial weight of gel; W_t is the weight of gel at day t .

Water holding capacity. Same as syneresis, 4% κ -carrageenan solutions were prepared. The gel was set inside a 15ml centrifuge tube to form a gel of height 100mm. The initial weight of gel was determined and was recorded as W_0 . The gel was stored overnight at room temperature (25°C), 4°C, and -18°C for 24 hours prior to centrifugation. Gel was centrifuged using a bench top centrifuge (Sigma 3-18K, Osterode am Harz, Germany) at 4000rpm for 30mins at 25°C. After centrifugation, the supernatant was drained and the gel was weighed immediately after the centrifugation runs. The weight ratio (W_t/W_0) was used to characterize the water holding capacity (WHC) of the gel.

$$\text{WHC (\%)} = W_t/W_0 \times 100 \quad (2)$$

where W_0 is the initial weight of gel; W_t is the weight of gel at day t .

Results and Discussions

Physical properties. Physical properties of five different κ -carrageenan samples: TA150, SeaKem CM611, Gelcarin GP812, Gelcarin GP911 NF and Grindsted[®] carrageenan CL220 are presented in Table 1, Table 2, Fig. 1, and Fig. 2. TA150 is the local κ -carrageenan produced from *Eucheumacottonii* cultivated in Tawau, Sabah while the others are the commercial κ -carrageenan obtained from the selected suppliers.

Colour, particle size, and pH. Hunterlab colorimeter results (Table 1) showed that κ -carrageenan samples were significantly ($p < 0.05$) different in term of lightness (L^*), redness (a^*), and yellowness (b^*). L^* represents the degree of lightness, whereby 100 indicates white and 0 indicates black. $+a^*$ represents redness while $-a^*$ indicates greenness. Yellowness is represented by $+b^*$ while blueness is shown by $-b^*$ [1,7]. In this study, the locally produced κ -carrageenan, TA150 was found to have the lowest L^* value (82.69) as compared to others commercial κ -carrageenan, indicating TA150 has the darkest colour among all the samples. Oppositely, SeaKem CM611 exhibited lightest colour with significant highest L^* value (88.87) at $p < 0.05$. Moreover, results showed that TA150 had the highest yellowness b^* (17.16) value but Seakem CM 611 had the lowest b^* (11.08). By combining L^* and b^* , colour of TA150 was darkest while colour of SeaKem CM611 was lightest. Results for particle size showed that the particle sizes of κ -carrageenan samples were ranged from 57.93 μm to 96.08 μm , in which all are fall below 100 μm . On the other hand, study showed that pH of all κ -carrageenan sample solutions are fall above 7, which varied from 8.04 to 10.09.

Table 1 Physical properties of commercial κ -carrageenan and κ -carrageenan isolated from Sabah *Eucheumacottonii*

Sample	Colour			Particle size (μm)	pH
	L^*	a^*	b^*		
TA150	82.69 \pm 0.23 ^d	2.10 \pm 0.01 ^a	17.16 \pm 0.15 ^a	86.50 \pm 0.94 ^b	9.89 \pm 0.08 ^a
SeaKem CM611	88.87 \pm 0.13 ^a	1.60 \pm 0.05 ^b	11.08 \pm 0.13 ^c	74.60 \pm 3.36 ^c	10.09 \pm 0.28 ^c
Gelcarin GP812	83.84 \pm 0.68 ^c	2.13 \pm 0.02 ^a	13.13 \pm 0.27 ^b	57.93 \pm 2.36 ^e	8.04 \pm 0.68 ^c
Grindsted [®] carrageenan CL220	87.51 \pm 0.42 ^b	0.27 \pm 0.01 ^c	11.91 \pm 0.57 ^c	96.08 \pm 2.33 ^a	8.20 \pm 0.42 ^{bc}
Gelcarin GP911 NF	83.63 \pm 0.29 ^{cd}	2.18 \pm 0.09 ^a	12.37 \pm 0.51 ^{bc}	62.47 \pm 2.65 ^d	8.34 \pm 0.39 ^b

Data were expressed as mean \pm standard deviation ($n = 3$).

Mean values with different superscripts in the same column are significantly different at $p < 0.05$.

Textural properties. Addition of food additives such as emulsifier, thickener, and gelling agent may alter the texture of the final products. Therefore, study on the textural properties of κ -carrageenan becomes very important for a better control of final products texture. In this study, two textural properties of κ -carrageenan were examined, namely rupture strength and brittleness. According to Raina & Babbar (2011), gel strength is only the amount of force required to rupture the gel [14]. Rupture strength is an indication of the maximum force required to rupture the gel. The distance that the probe penetrates through the gel before the break occurs is expressed as gel brittleness [14]. Texture analysis is thus more descriptive of gel texture than simple gel strength measurements.

Textural measurement on κ -carrageenan samples were carried out at increasing gel concentration. Concentration of all κ -carrageenan samples varied from 2% w/v to 4% w/v except SeaKem CM611. When TA150 and other κ -carrageenan were dissolved in hot water and then followed by cooling, they form thermo-reversible gel at concentration as low as 2% w/v. SeaKem CM611 did not form gel at concentration below 4% w/v. Thus, a range of 4% w/v to 6% w/v was selected.

As tabulated in Table 2, rupture strength of κ -carrageenan increased significantly ($p < 0.05$) with its concentration. The higher the water content of the gels, the lower is their mechanical strength. In other words, greater force is required for gel rupture at increasing carrageenan content (decreasing water content) of gels. At the same concentration, Gelcarin GP812 showed the highest rupture strength followed by Gelcarin GP911 NF, TA150, Grindsted carrageenan CL220, and SeaKem CM611. Moreover, the increase in rupture strength of Gelcarin GP812 was also the highest, from 9.89N (at 2.0%) to 31.23N (at 4.0%), give a total increment of 215.77%. Brittleness of κ -carrageenan is presented in Table 2. A small brittleness value indicates a more brittle gel. Among all, Grindsted carrageenan CL220 was the most brittle gel as indicated by its smallest brittleness value.

Functional properties. κ -carrageenan gel retains a large amount of water when undergoing a solution/gel transition. Due to intrinsic instability, water may be lost from κ -carrageenan via syneresis after extensive storage time [11]. Water lost may result in gel shrinkage, texture change, and quality reduction. This has become a challenge in the manufacturing of food gels. Results for syneresis are summarized in Fig.1 and Fig.2. Syneresis of κ -carrageenan was significantly ($p < 0.05$) increased with storage time (Fig. 1). The highest syneresis corresponded to TA150. In the first 2 days, 2.79-5.02% of water was extruded from the κ -carrageenan gel. Syneresis of κ -carrageenan gel reached 7.80-21.50% after 10 days of storage time. Lee *et al.* (2008) observed the same phenomenon in which syneresis of κ -carrageenan increased upon storage [10]. The effect of storage time on syneresis of κ -carrageenan gel was also investigated. As depicted in Fig. 2, lowest and highest syneresis was observed when κ -carrageenan stored at 4°C and -18°C, respectively. As compared to 25°C, syneresis of κ -carrageenan was lower when stored at 4°C. κ -carrageenan gel structures hold water better when gel stored at lower temperature. However, syneresis of κ -carrageenan was found to increase significantly ($p < 0.05$) when storage temperature went down to -18°C. At very low temperature, crystallization may occur and destroy the gel structure. Once gel structure deformed, κ -carrageenan lost its ability to hold the water in place. The water is thus diffuse out from internal to the surface of the gel.

Water holding capacity (WHC) which is the capability of the gel to retain this water when subjected to external forces was also examined. WHC of κ -carrageenan was observed at three different storage temperature (25°C, 4°C, and -18°C). The results showed that WHC of κ -

carrageenan were reported to be excellent (>90%) regardless of the storage temperature (data not shown).

Table 2 Textural properties of commercial κ -carrageenan and κ -carrageenan isolated from Sabah *Eucheumacottonii*

Sample	Concentration (%)	Rupture strength (N)	Brittleness (mm)
TA150	2.0	4.28±0.47 ^e	14.32±0.56 ^a
	2.5	8.14±0.24 ^d	15.87±0.33 ^a
	3.0	11.06±0.36 ^c	15.09±1.14 ^a
	3.5	12.93±0.36 ^b	15.83 ±0.92 ^a
	4.0	14.68±0.14 ^a	16.29±0.82 ^a
SeaKem CM611	4.0	0.64±0.05 ^e	16.57±0.41 ^a
	4.5	1.46±0.11 ^d	18.13±0.64 ^a
	5.0	2.51±0.13 ^c	17.51±0.89 ^a
	5.5	3.21±0.01 ^b	19.16±1.19 ^a
	6.0	3.98±0.09 ^a	18.97±0.97 ^a
Gelcarin GP812	2.0	9.89±0.19 ^e	15.59±0.58 ^a
	2.5	15.31±0.64 ^d	16.16±0.96 ^a
	3.0	19.67±1.23 ^c	16.41±0.83 ^a
	3.5	23.45±0.25 ^b	16.20±2.12 ^a
	4.0	31.23±1.14 ^a	14.93±0.41 ^a
Grindsted carrageenan CL220	2.0	1.15±0.01 ^d	8.90±0.120 ^b
	2.5	5.15±0.63 ^c	9.64±0.39 ^{ab}
	3.0	6.29±0.08 ^c	10.56±0.58 ^{ab}
	3.5	10.79±0.41 ^b	10.63±0.93 ^{ab}
	4.0	14.94±0.09 ^a	11.21±0.23 ^a
Gelcarin GP911 NF	2.0	6.08±0.03 ^d	18.44±0.68 ^a
	2.5	8.13±0.09 ^d	17.49±0.08 ^a
	3.0	12.85±0.28 ^c	16.92±0.16 ^a
	3.5	17.50±0.94 ^b	16.87±0.90 ^a
	4.0	20.48±0.88 ^a	16.80±0.40 ^a

Data were expressed as mean ± standard deviation (n = 3).

Mean values with different superscripts in the same column are significantly different at $p < 0.05$.

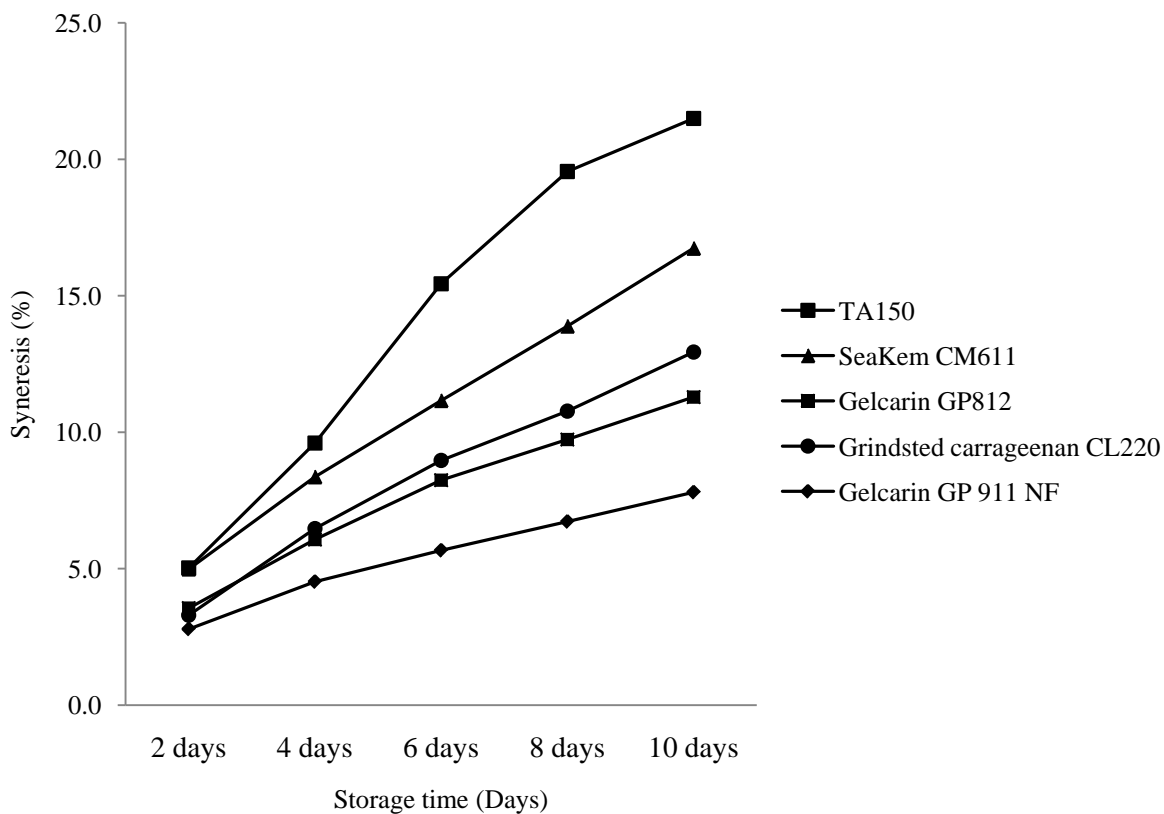


Fig. 1. Syneresis of commercial κ -carrageenan and κ -carrageenan isolated from Sabah *Eucheumacottonii*.

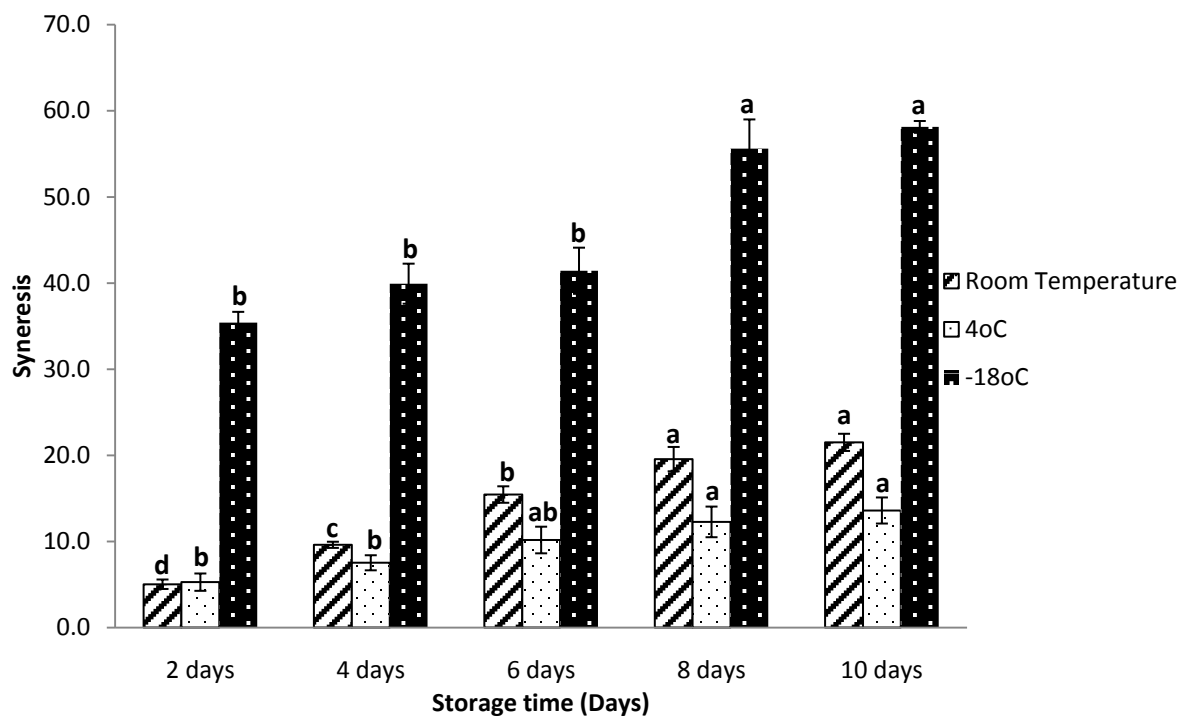


Fig. 2. Syneresis of κ -carrageenan isolated from Sabah *Eucheumacottonii* at different storage temperature (25°C, 4°C, -18°C). Values marked by different letters under the same temperature are significantly different ($p < 0.05$).

Acknowledgements. Financial support of this work by the Ministry of Science, Technology and Innovation of Malaysia through ABI Fund (08-05-ABI-PB033) is gratefully acknowledged. Special thanks are given to TacaraSdn. Bhd., Malaysia, FMC Biopolymer, USA, and Danisco, Denmark for providing the κ -carrageenan samples for this study.

References

- [1] A.A.N. Aziah and C.A. Komathi: *Journal of Food Science* Vol. 74 (2009), p. S328-S333.
- [2] M. Dyrby, R.V. Petersen, J. Larsen, B. Rudolf, L. Norgaard and S.B. Engelsen: *Carbohydrate Polymers* Vol. 57 (2004), p. 337-348.
- [3] A. Ellis and J.C. Jacquier: *Journal of Food Engineering* Vol. 94 (2009), p. 316-320.
- [4] Y. Freile Y. and D. Robledo: *Journal of Applied Phycology* Vol. 20 (2008), p. 537-541.
- [5] L. Hayashi, A. Q. Hurtado, F.E. Msuya, G. Bleicher-Lhonneur and A.T. Critchley, in: *A Review of Kappaphycus Farming: Prospects and Constraints*, edited by Seckbach and Joseph (Eds.), *Cellular Origin, Life in Extreme Habitats and Astrobiology*, Springer Science+Business Media (2010).
- [6] A.P. Imeson, in: *Carrageenan*, edited by G. O. Phillips and P.A. Williams, *Handbook of Hydrocolloids*, England: Woodhead Publishing Limited (2000).
- [7] B. Jamilah, C.E. Shu, M. Kharidah, M.A., Dzulkifly and A. Noranizan: *International Food Research Journal* Vol. 18 (2011), p. 279-286.
- [8] JECFA, *Combined compendium of food additive specifications*. Rome: Food and agriculture organization of the united nations. (2006)
- [9] S. Keppeler, A. Ellisa and J.C. Jacquier: *Carbohydrate Polymers* Vol. 78 (2009), p. 973-977.
- [10] J.S. Lee, Y.L. Lo, and F.Y. Chye: *Sains Malaysiana* Vol. 37 (2008), p. 71-77.
- [11] R. Mao, J. Tang, J. and B.G. Swanson: *Carbohydrate Polymers* Vol. 46 (2001), p. 365-371.
- [12] H.J. Mou, X.L. Jiang, Z.H. Liu and H.S. Guan: *Journal of Food Biochemistry* Vol. 28 (2004), p. 245-260.
- [13] L. Pereira, A.M. Amado, A.T. Critchley, F.V.D. Velde and P.J.A. Ribeiro-Claro: *Food Hydrocolloids* Vol. 23 (2009), p. 1903-1909.
- [14] R.J., Raina and S.B. Babbar: *Asian Journal of Biotechnology* Vol. 3 (2011), p. 153-164.
- [15] B. Rudolph, in: *Seaweed Products: Red Algae of Economic Significance*, edited by R.E. Martin, E.P. Carter, G.J. Flick and L.M. Davis, *Marine and Freshwater Products Handbook*, U.S.A.: Technomic Publishing Company, Inc (2000).
- [16] M.G. Sankalia, R.C. Mashru, J.M. Sankalia and V.B. Sutariya: *International Journal of Pharmaceutics* Vol. 312 (2006), p. 1-14.
- [17] P. Volery, R. Besson and C. Schaffer-Lequart: *Journal of Agricultural and Food Chemistry* Vol. 52 (2004), p. 7457-7463.