(045) Kinetics of Astaxanthin with Encapsulation from *Haematococcus pluvialis* during Storage

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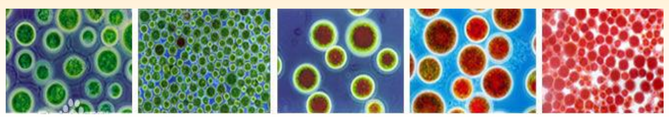
Abstract

ASX can easily be degraded by heat, light and oxygen during the processing and storage. Encapsulation of ASX by β-cyclodextrin has developed to increase the storage stability and solubility. However, the quantitative information of ASX encapsulation on storage stability is still very limited; therefore, the objectives of this study was to establish the kinetics model of ASX retentions by encapsulation and storage temperature. In this study, the stabilities of ASX and β-cyclodextrin encapsulated astaxanthin (CD-ASX) were analyzed under various storage conditions, namely, storage temperature (25, 40, 45, and 50ºC), and storage atmosphere (in a sealed tube and an uncovered tube) to evaluate kinetic parameters of astaxanthin degradation. The results showed that the kinetic model of ASX and CD-ASX retentions were the zeroth reaction order. In addition, the reaction rate constant k values were increased with higher storage temperature, and there were significant difference between uncover and sealed conditions. Therefore, temperature and air significantly affected the stabilities of ASX. The ASX retention rates were plotted with inverse of different absolute storage temperatures, and then the activation energy (Ea) could be obtained according to Arrhenius' equation. The results showed that the Ea of ASX after encapsulation in the seal condition increased from 56.82 kJ/mol to 113.08 kJ/mol. The Ea of ASX after encapsulation in the uncover condition increased from 51.72 kJ/mol to 106.02 kJ/mol; therefore, the encapsulation with low storage temperature could enhanced the storage stability of ASX from *Haematococcus pluvialis*.

Keywords: *Haematococcus pluvialis*, astaxanthin, encapsulation, kinetics

Introduction

Microalgae *Haematococcus pluvialis* is considered as a good candidate for producing nature astaxanthin (3,3'-dihydroxy-b,b'-carotene-4,4'-dione, ASX), and ASX is a high-value carotenoid with strong antioxidant properties, which has many applications in food, nutraceutical, cosmetic, feed industry. Astaxanthin extract from the *H. pluvialis* can was analyzed by HPLC and it was added 0.1N NaOH under 6℃ nitrogen saponification 24 hr at dark. HPLC condition was C18 reverse phase HPLC column chromatography with acetonitrile: methanol (15:85) as the mobile phase, 1 mL/min of flow rate, and detection at 476 nm wavelength. However HPLC analysis took much longer time than more than colorimetric analysis. Astaxanthin analyzed by HPLC was 76.38% of total carotenoid by colorimetric analysis.



Five different growth phases of *Haematococcus pluvialis*

Materials & Methods

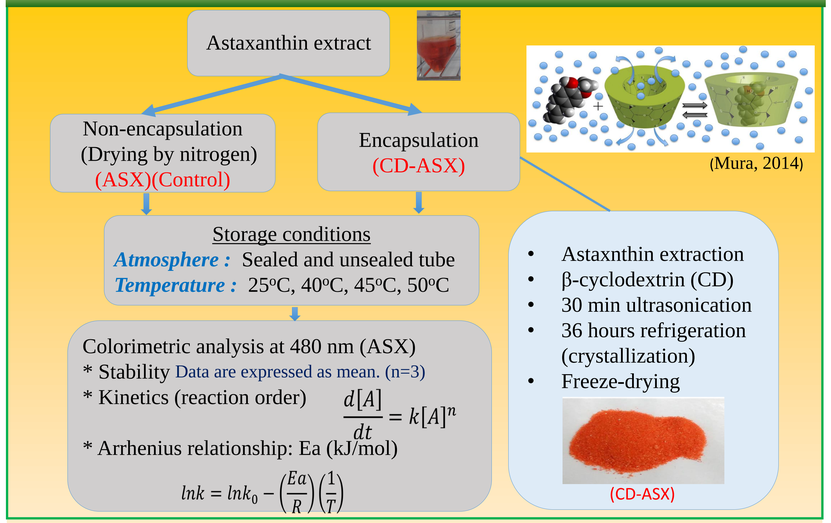
- ***H. pluvialis* powder**

It was provided by Industrial Technology Research Institute, Taiwan (R.O.C.)

- ***Astaxanthin Extraction***

H. pluvialis powder → Acid pretreatment: 4N HCl, 70 ℃, 10 min → Ultrasonic extraction: 95% ethanol, 10min → Astaxanthin extract

Experimental Design: Encapsulation/ Storage/ Kinetics



Results

The ASX recovery of *H. pluvialis* powder could reach 95.97 %. According to Table 1, the reaction order of ASX retention followed the zeroth order reaction. The rater constants and half life of ASX retention during four different temperature storage were shown in Table 2. Both temperature and CD encapsulation significantly affected rate constants (Fig. 1). According to Arrhenius' equation, the activation energies were shown in Table 3 and Fig. 2. CD encapsulation could avoid oxidation to improve ASX stability due to higher Ea.

Table 1 The average coefficient of determination (r^2) of reaction order of astaxanthin retention regression at different storage conditions

Sample treatment	n=0	n=0.5	n=1	n=1.5	n=2
ASX / seal	**0.9392**	0.9372	0.9328	0.9287	0.9222
ASX / Unsealed	**0.9272**	0.9258	0.9215	0.9137	0.9017
CD ASX / seal	**0.9457**	0.9389	0.9312	0.9227	0.9136
CD ASX / Unsealed	**0.9490**	0.9417	0.9327	0.9221	0.9101

Table 2 Rate constants of astaxanthin retention by different treatments at different temperature conditions

Temperature (ºC)	Sample treatment	y=A+Bx	R^2	Rate constant	$\tau_{1/2}$ (hr)
25℃	ASX/ seal	y = -0.0144x + 1.0585	0.9219	0.0144	39
	ASX / unsealed	y = -0.0215x + 1.0311	0.9507	0.0215	25
	CD ASX / seal	y = -0.0020x + 1.0045	0.9940	0.0020	252
	CD ASX / unsealed	y = -0.0023x + 0.9881	0.9369	0.0028	174
40℃	ASX/ seal	y = -0.0413x + 0.9753	0.9408	0.0413	12
	ASX / unsealed	y = -0.0641x + 0.9553	0.9157	0.0641	7
	CD ASX / seal	y = -0.2080x + 0.9990	0.9173	0.0208	24
	CD ASX / unsealed	y = -0.0309x + 0.9968	0.9374	0.0309	16
45℃	ASX/ seal	y = -0.0492x + 1.0183	0.9548	0.0492	11
	ASX / unsealed	y = -0.0766x + 1.0476	0.9152	0.0766	7
	CD ASX / seal	y = -0.0271x + 1.0132	0.9338	0.0271	19
	CD ASX / unsealed	y = -0.0304x + 1.0050	0.9432	0.0304	17
50℃	ASX/ seal	y = -0.0951x + 1.0200	0.9622	0.0951	5
	ASX / unsealed	y = -0.1103x + 0.9697	0.9519	0.1103	4
	CD ASX / seal	y = -0.0782x + 1.0235	0.9182	0.0782	7
	CD ASX / unsealed	y = -0.0899x + 0.9933	0.9576	0.0899	5

Table 3 Activation energy (Ea) and rate constants of astaxanthin retention at different treatments

Temperature (℃)	ASX/ seal	ASX / unsealed	CD ASX / seal	CD ASX / unsealed
25	0.0144	0.0215	0.0020	0.0028
40	0.0413	0.0641	0.0208	0.0309
45	0.0492	0.0766	0.0271	0.0304
50	0.0951	0.1103	0.0782	0.0899
R^2 (Arrhenius eq.)	0.9700	0.9936	0.9845	0.9626
y=A+Bx	y = -6833.7x+18.655	y = -6220.7x+17.052	y = -13601x+39.433	y = -12752x+36.962
Ea (kJ/mol)	56.82	51.72	113.08	106.02

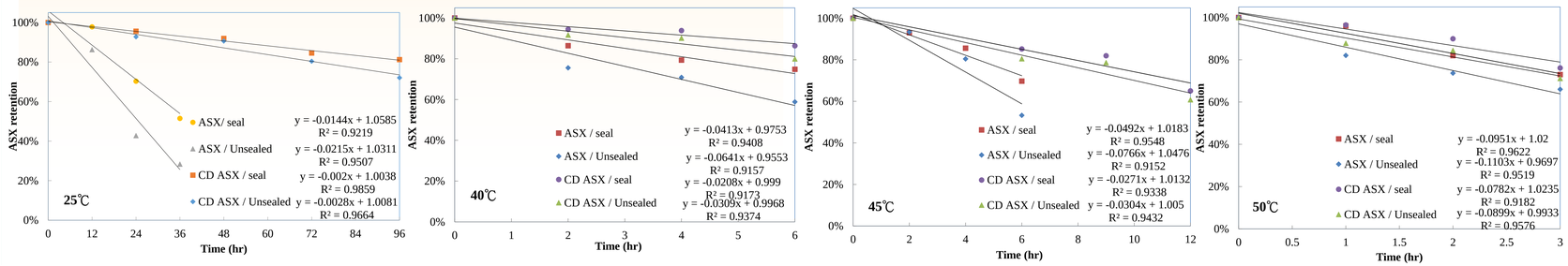


Fig. 1. Change of astaxanthin retention by different treatments during , 25, 40, 45 and 50ºC storage.

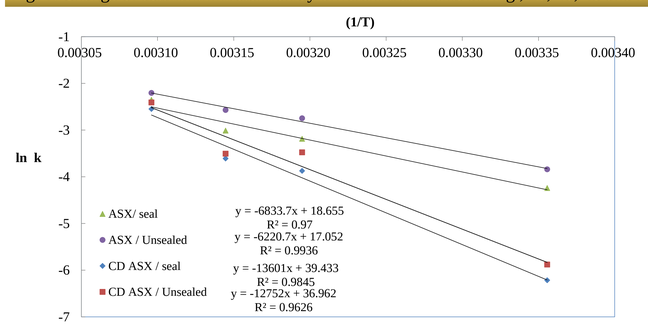


Fig. 2. Arrhenius equation of astaxanthin retention by different treatments.

Conclusions

The kinetics models was the zeroth reaction order and k values were increased with higher storage temperature. ASX in a seal tube had small k values than in an unseal tube due to oxidation. CD encapsulation could significantly improve ASX stability during storage.

References

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