

Best Method for the Extraction of Egg Carotenoid Pigments Golden Egg Snails (*Pomacea canaliculata* Lamarck)

Nurjanah Taufik Hidayat*, Batari Perbawani

Department Aquatic Product Technology, Faculty of Fisheries and Marine Sciences, Bogor Agricultural University, Bogor West Java, Indonesia

Abstract

Snails (*Pomacea canaliculata* Lamarck) is one type of freshwater snails which comes from the Americas. Snails have become a serious threat in agriculture due to high reproductive power and capability of rapid deployment area. This research result of chemical composition showed that golden snail egg contain to high moisture and fat. Golden snail eggs have a high total carotenoids is 313.48 ± 19.73 ppm. This research aims to optimize the extraction process of carotenoids with different types of solvents. Golden snail eggs, chopped and mashed in a blender then dried by the method of freeze dry for 58 hours. After getting the golden snail eggs, egg powder snails do multilevel extraction method using n-hexane, acetone and methanol in the ratio 1: 10 (w / v) and extracted with maceration for 24 hours. Furthermore, the single extraction with solvents methanol 1: 10 (w / v), ethanol 1: 10 (w/v), acetone: methanol with a ratio of 3: 7 and acetone: distilled water with a ratio of 3: 15 (w/v) method maceration for 48 hours. Results extraction then evaporated and the result filtrate diluted with methanol measured using a spectrophotometer to see the absorption wavelength. The optimum solvent extraction golden snail eggs is a polar solvent to extract the highest acetone: distilled water with a ratio of 3: 15 namely 1084.6 mg.

Keywords

Carotene, Egg Snails, Chemical Composition, Extraction

Received: June 18, 2016 / Accepted: June 28, 2016 / Published online: July 21, 2016

© 2016 The Authors. Published by American Institute of Science. This Open Access article is under the CC BY license.

<http://creativecommons.org/licenses/by/4.0/>

1. Introduction

Snails (*Pomacea canaliculata*) is easy to find in the rice fields or on the waters edge welled in Indonesia. Snails (*Pomacea canaliculata* Lamarck) is one type of freshwater snails which comes from the Americas. Snails can cause damage of up to 10-40% of the total rice areas in Indonesia, namely in Java, Sumatra, Kalimantan, West Nusa Tenggara and Bali (Budiyono 2006). Pests from the class of mollusks is potentially a major pest of rice crops due to rapidly proliferate and attack young plants. Snails during his lifetime is able to produce as many eggs 15-20 groups that each group numbered approximately 500 grains with hatching percentage of more than 85% (Budiyono 2006).

Efforts to prevent pest snails (*Pomacea canaliculata*) for this is still considered less effective, therefore it is necessary that another solution is the utilization of golden snail eggs. Golden snail eggs in Indonesia has been used as crackers and juice (a healthy drink) allegedly due for mineral content (calcium) is high and the fertilizer plant growth regulator (aphrodisiac grown) organic allegedly because carotenoids are high on golden snail eggs (Nurjanah et al. 2016). However, the utilization of its own golden snail eggs in Indonesia has not been fully utilized.

Golden snail eggs have a high potential especially in carotenoid pigments. Total carotenoids golden snail eggs are 313.48 ± 19.73 ppm. This is higher than the total carotenoids

* Corresponding author

E-mail address: besthd22@gmail.com (N. T. Hidayat)

carrots of various hybrid (60.21 ± 0.66 to 79.47 ± 0.42 ppm) and eggs chinook salmon (*Oncorhynchus tshawytscha*) (17.9 ppm) (Nurjanah et al. 2016). Flying fish eggs that have total carotenoid eggs 245.37 ppm and 37.92 ppm fibers. As well as levels of α -tocopherol flying fish eggs 1.06 ppm (Azka et al 2015).

Carotene is an efficient scavenger of free radicals and can significantly reduce the risk of cancer (Henrikson 2009). Besides carotenoids are also widely used as additives in food is a food coloring (Mortensen 2006). Utilization of carotenoid pigments golden snail eggs have to be optimized in view of the domestic industry requirement of the carotenoid pigment is still filled with the pigment importing them from abroad (Desiana 2000).

The optimum method for the collection of carotenoid pigments of golden snail eggs so there is no need to be optimized to obtain optimal results carotenoid pigments of golden snail eggs. Carotenoids soluble in organic solvents such as hexane, toluene and ethanol (Brion et al 1995). Xantofil more soluble in alcohol and slightly soluble in petroleum ether than carotene (Desiana 2000). To search for optimum conditions required solvent according to the characteristic golden snail eggs. The purpose of this study was to get a carotenoid pigment golden snail eggs with finding the optimum solvent extraction maceration golden snail eggs.

2. Materials and Methods

2.1. Materials and Tools

Golden snail eggs used in this study came from a pool of Water Cultivation Faculty of Fisheries and Marine Sciences IPB, Bogor Dramaga. The solvents used are n-hexane, acetone and methanol with Pro quality analysis (P.A) is obtained from Raw Material Science Laboratory, Bogor Agricultural University and material for chemical compositions.

The tools (may be use equipments is better) used in the manufacture of powdered egg snails include spoons, Freezer dryer, dry blender and plastic. The tools (may be use equipments is better) used in the extraction of carotenoids golden snail eggs are tools maceration, filter paper is smooth, rotary evaporator, Erlenmeyer flask, analytical balance, spatula and aluminum foil. The tools (may be use equipments is better) used in the analysis of the extract carotene include a test tube, tube rack timber, aluminum foil, and a spectrophotometer cuvette.

2.2. Methods

The method used in this research is the manufacture of egg

powder snails with freeze drying method, the proximate analysis proximate analysis which includes the analysis of moisture, ash, fat, protein, and carbohydrate (AOAC 2005). Extraction maceration using multilevel extraction and a single as well as analysis of the value of maximum absorption with a spectrophotometer.

2.3. Making Powdered Eggs Snails

Samples golden snail eggs that have been collected in the washing with water after which the samples in the blender. Results of golden snail eggs that have been in the blender dried by freeze dry method with -50°C temperature for 58 hours. Once it got powdered eggs snails.

2.4. Extraction Caroten

Phase extraction is done with terraced extraction method using a non-polar solvent n-hexane, acetone and Semi polar Polar Pro Methanol quality analysis (P.A) with a ratio of 1: 10 with samples that are used as much as 10 grams. The first extraction was performed using n-hexane then in maceration for 24 hours until clear solvent, after which the sample is extracted again using acetone solvent using a maceration for 24 hours until a clear colorless solvent, followed by solvent methanol samples do ekstraksi (local words) maceration for 24 hours.

The second stage performed single extraction using polar solvents. Polar solvent used is methanol, ethanol at a ratio of 1: 1, acetone: methanol with a ratio of 3: 7 and acetone: distilled with a ratio of 3: 15 with quality solvent Pro. Analysis (P.A) for 2 x 24 hours.

2.5. Analysis Spectrophotometer

The results that have been extracted filtrate absorbance is measured using a spectrophotometer. Uptake value was measured at a wavelength of 479nm (astaxhantin) and optimum uptake seen in the sample.

2.6. Data Analysis

Result data used statistical descriptive with compare data and study literature. In addition result data interpretation with compare another data with study literature.

3. Result and Discussion

3.1. Proximate Golden Snail Eggs

Table 1 showed that the golden snail eggs have a high moisture content is 81.16% water content analysis principle is the measurement of the weight of the amount of water lost in the material of the original substance after heating (Yulianingsih and Tamzil 2007). The ash content in the egg

shell snails was 9.79%, this shows the levels of minerals in the golden snail eggs is quite high. Fat levels in golden snails is (Why used ie? Should use: it mean or mean, is...) 4.84% fat content in a biological material affected by species, food, habitat, size, and degree of maturity of the gonads will affect lipid levels (Belitz et al 2009).

The protein content in golden snails is 2.63%. Ovourubin, 300-kDa thermostable oligomer is a major protein in the fluid perivitellin (PV) surrounding the embryo golden snails. Ovourubin has an essential role to the development of the embryo, protease inhibition, photoprotection, and storage of food. Lipoprotein on golden snail eggs serve to provide energy and nutrients for the development of the embryo (Dreon et al., 2008). The result of the calculation of carbohydrate content using the method by difference in golden snail eggs, is 1.58% Polysaccharides in perivitellin golden snail eggs, consisting of galactose and fucose unit (Heras and Pollero 2002). According to Nurjanah et al 2016 research about mineral content and carotene golden egg snails egg, in the result not significant. The high content is fat. Different result research with Nurjanah et al 2016 because by species, age, habitat and size. According to Azka *et al* 2015 with fly egg the high content to chemical composition was protein. Results of the proximate analysis golden snail eggs can be seen in Table 1.

Table 1. Chemical composition golden snail eggs.

Proximate	Golden snails egg (%)
Moisture	81.16
Ash	9.79
Fat	4.84
Protein	2.63
carbohydrate	1.58

3.2. Yield Extract Carotene with Terraced

Extraction terraced used N-hexane (non polar), acetone (semi polar), and methanol (polar). Extraction storey terraced golden snail eggs using maceration method. Maceration method used because with maceration can be simple and can high yield (Nurjanah et al 2016). However, maceration have negative if can find to molecule compound with high temperature. Extraction carotene can be seen in Table 2.

Table 2. Result extraction carotene golden snails egg.

Solvent	Long extraction (hour)	Result (mg)
n-Hexane	24	-
Acetone	24	-
Methanol	24	10

Table 2 showed that the golden snail eggs are not extracted in the solvent n-hexane and acetone, golden snail eggs can be extracted in a polar solvent is methanol. This is due to the ingredients and compounds kirnia be soluble in the same solvent polarity with the material to be dissolved (Slamet

Sudarmadji et al., 1996). Carotene dissolved in a solvent of low polarity and xantofil soluble in the solvent polarity higher (Britton et al., 1995). This proves that the golden snail eggs soluble in the solvent which has a high polarity.

Solvent extraction using methanol extracts showed that 10 mg yield. One of the factors that affect carotenoid group solvent extraction is kind xantofil soluble in alcohol and slightly soluble in ether petroleum (Desiana 2000). Type polar solvent has a polarity the same level with the golden snail eggs.

After extraction of single storey extraction using a polar solvent types that have different levels of polarity. Results maceration extraction using polar solvents can be seen in Table 3.

Table 3. Results of single extraction carotene.

Solvent	Long extraction (Hour)	Result (mg)
Methanol	48	20.2
Ethanol	48	3.2
Acetone: Aqua	48	1084.6
Acetone: Methanol	48	10.6

Based on the results of single extraction showed the highest extract using acetone: distilled water is 1084.6 mg followed by 20.2 mg Methanol, Acetone: Methanol Ethanol 10.6 mg and 3.2 mg.

Selection of solvents for maceration process will provide high effectiveness by observing the solubility of compounds of natural materials such solvents (Dervish 2000). This indicates that the compound is polar golden snail eggs so soluble in polar compounds. Based on the results of the highest yield golden snail eggs have a high polarity. The level of polar solvent used is acetone based on the dielectric constant (21), ethanol (30), methanol (33), Air (80).

3.3. Carotene Pigments Measurements

After the extract in getting samples was measured using a spectrophotometer to see the absorption wavelength. Analysis golden snail eggs extract has an absorption maximum is 495 nm According to Jan olsina, et al., (2004) the maximum wavelength of the pigment astaxanthin range between 470-495 nm. The results showed that the eggs of snails contains carotenoid pigments with astaxanthin types. This is consistent with research Wu and Yang (2008) who found that the eggs snails and snails gonad astaxanthin high value.

Astaxanthin is a red carotenoid pigments that are naturally present in various living organisms that are classified as xanthophyll. Xanthophyll has a hydroxyl group at the end of the molecule that distinguishes carotenoids and astaxanthin have more hydroxyl groups are compared to other xanthophyll. Astaxanthin have stronger antioxidant activity

than all the carotenoids, including lutein and beta-carotene (Capelli and Chewsky 2007).

4. Conclusion

Type the optimum solvent extraction golden snail eggs are using a polar solvent. The yield is highest in acetone: distilled water to extract 1084.6 mg. Carotenoid pigments found in golden snail eggs that astaxhantin with maximum wave 495nm.

References

- [1] Azka A, Nurjanah, Jacob AM. 2015. Profil asam lemak, asam amino, total karotenoid, dan α - tokoferol telur ikan terbang. *Jurnal Pengolahan Hasil Perikanan Indonesia*. 18 (3): 250-261.
- [2] Belitz HD, Grosch W, Schieberle P. 2009. *Food Chemistry*. Ed rev ke-4. Verlag: Springer.
- [3] Briton G, S. L Jansen dan H. Pfander. Carotenoids. 1995. Vol IB. Spectroscopy. Academic Press Inc. Basel 386.
- [4] Budiyo S. 2006. Teknik mengendalikan keong mas pada tanaman padi. *Jurnal Ilmu-Ilmu Pertanian* 2(2): 128-133.
- [5] Cappeli B dan Cheswky G. 2007. Natural Astaxhantin. USA: Cyanotech Cooperation.
- [6] Darwis, D. 2000. Teknik Dasar Laboratorium dalam Penelitian Senyawa Bahan Alam Hayati. Workshop Pengembangan Sumber Daya Manusia dalam Bidang Kimia Organik Bahan Alam Hayati. FMIPA Universitas Andalas. Padang.
- [7] Desiana. 2000. Ekstraksi pigmen karotenoid dari limbah kulit udang windu (*Penaeus monodon Fabricus*) dengan bantuan enzim papain [skripsi]. Bogor: Fakultas Perikanan dan Ilmu Kelautan, Institut Pertanian Bogor.
- [8] Dreon MS, Ituarte S, Ceolin M, Heras H. 2008. Global shape and pH stability of ovorubin, an oligomeric protein from the eggs of *Pomacea canaliculata*. *FEBS Journal* 275 (18): 4522-4530.
- [9] Henrikson R. 2009. Earth Food Spirulina How this Remarkable Blue-Green Algae can Transform Your Health and Our Planet. USA: Ronore Enterprises. Inc.
- [10] Heras H, Pollero RJ. 2002. Lipoprotein from plasma and perivelline fluid of the apple snail *Pomacea canaliculata*. *Biocell* 26(1): 111-118.
- [11] Nurjanah N, Nurhayati T, Hidayat T, Agustina MA. 2016. Mineral content of Macro-Micro And Total Carotenoids Eggs Golden Apple Snail (*Pomacea canaliculata*) From Pond Culture FPIK IPB. *International Food Research Journal*. *Inpress*.
- [12] Mortensen A. 2006. Carotenoids and Other Pigments as Natural Colorants. *Pure Appl. Chem.*. 78 (8): 1477-1491.
- [13] Slamet Sudamadji, Barnbang Haryono, Suhardi. 1996. *Analisa Bahan Makanan dan Pertanian*. Yogyakarta: Liberty.
- [14] Wu YL, Yang CC. 2008. Method for obtaining natural astaxanthin from eggs and gonads of snails, US patent and trade mark office Patent US8030523.
- [15] Yulianingsih R, Tamzil. 2007. Analisis proksimat rumput laut produksi dari beberapa lokasi di Indonesia Timur. *Buletin Teknologi Litkayasa Akuakultur* 1(6): 51-55.