

HIGH-PERFORMANCE THIN LAYER CHROMATOGRAPHY DETERMINATION OF ASTAXANTHIN IN *Euphausia superba* OIL

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The Antarctic krill (*Euphausia superba*) is a kind of marine zooplankton belonging among Crustacea [1]. It is the principal prey of many marine predators by reason of its gigantic population [2]. The study of Antarctic krill has become a hot research field in the past few decades. It is not only the key organism in the southern ocean but also the biggest fishery resource in the world. With its high content of astaxanthin as a valuable natural product [3, 4], it urgently awaits development and utilization. The ketocarotenoid astaxanthin (3,3'-dihydroxy- β,β' -carotene-4,4'-dione) and astaxanthin esters are the principal components of Antarctic krill pigment [4], and astaxanthin is one of most important carotenoids by virtue of its biologic functions as a vitamin A precursor and as a much more efficient antioxidant than β -carotene and vitamin E [5]. It is also a natural coloring (in fish muscles, bird feathers, yolks, etc.), supports the immune system, has an anti-tumor effect, and protects from ultraviolet radiation [6–8]. Overall, astaxanthin can play an important role in healthcare and cosmetic manufacture, with enormous commercial and industrial prospects.

As a general rule, high-performance liquid chromatography (HPLC) is preferred in astaxanthin assay due to its wide use all around the world [9], whereas the HPTLC technique with high sensitivity and resolution can also function excellently in the separation and purification of bioactive compounds from natural materials.

In short, we aim to establish and validate an HPTLC method that is adapted to astaxanthin determination in Antarctic krill oil and then can be applied to other materials that contain astaxanthin.

The astaxanthin in Antarctic krill is mostly in the form of astaxanthin esters for its molecular stabilization [10]. Some are diesters and others are monoesters, attributed to the two hydroxyls that can combine with different fatty acids at both ends of the astaxanthin molecule [11]. While the concentration of free astaxanthin was small, all of it and other impurities are fat soluble and can be dissolved in low-polarity reagents; thus, *n*-hexane-acetone (7:2, v/v) was chosen to be the mobile phase after repeated attempts, combined with the high-performance silica gel 60 F254, which can accomplish the HPTLC separation within 10 min. The R_f value of the free astaxanthin was 0.32.

It was necessary to apply the saponification procedure to release the astaxanthin combined with fatty acids because the concentration of free astaxanthin was from $5 \pm 1.0\%$ to $85 \pm 5.0\%$, so that the qualification and quantification of astaxanthin can be carried out more successfully, as well as the further purification, since there was no standard for astaxanthin esters. KOH was essential in saponification, but it can result in significant degradation of astaxanthin, especially when the reaction temperature was high. There was almost no degradation of astaxanthin at 4°C [12]; therefore it was selected as the saponification temperature, and the maximum concentration of free astaxanthin can reach 89.7466%. The linear regression equation of astaxanthin after saponification is $Y = 1309.2935 + 47.9693X$ (where Y is the response and X is the mass of astaxanthin). The correlation coefficient was found to be 0.9990, the RSD (relative standard deviation) was 2.56%, and the astaxanthin in the saponification sample was $0.7045 \text{ mg}\cdot\text{mL}^{-1}$; more data are shown in Table 1.

The three main components in the original sample were separated without interference from other impurities, and the spectral evaluation of the samples showed the same maximum absorption (λ_{max} 476 nm) as the standard; therefore, the specificity of this method is verified.

The RSD of intraday precision was 1.68% and the value of the interday precision was 5.07%. Both values were determined when the plates were stored in the dark.

The accuracy was clarified as percent recovery, and the average recovery of astaxanthin was 98.53%.

Key Laboratory of Animal Resistance, College of Life Science, Shandong Normal University, 88 East Wenhua Road, 250014, Jinan, P. R. China, fax : 86-0531-86185360, e-mail: hcdh@sdsnu.edu.cn. Published in *Khimiya Prirodnykh Soedinenii*, No. 1, January–February, 2013, pp. 128–129. Original article submitted September 23, 2011.

Progress In Thin Layer Chromatograp Volume 1

Eli Grushka,Nelu Grinberg



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CRC Handbook of Chromatography Ram N. Gupta, 2018-04-17 These volumes provide a reference source of different gas chromatographic liquid chromatographic or thin layer chromatographic techniques for the qualitative determination of various therapeutic agents including antibiotics vitamins and hormones drugs of abuse in body fluids dosage forms or food stuffs Over 5000 publications were reviewed to prepare tables of chromatographic data for 800 compounds arranged alphabetically by generic drug name or by drug groups A detailed summary of the extraction procedure described in each publication included in the table of a particular drug is also provided This easy to read handbook is useful for selecting an appropriate chromatographic procedure for the determination of a given compound according to the available facilities

Thin-Layer Chromatography, Revised And Expanded Bernard Fried, Joseph Sherma, 1999-01-04 The fourth edition of this work emphasizes the general practices and instrumentation involving TLC and HPTLC as well as their applications based on compound types while providing an understanding of the underlying theory necessary for optimizing these techniques The book details up to date qualitative and quantitative densitometric experiments on organic dyes lipids antibiotics pharmaceuticals organic acids insecticides and more

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Ruane, I.D. Wilson, 2013-11-09 For many years TLC has suffered from the image of being a low sensitivity low resolution non quantitative technique suitable for chemists but not a tool for real chromatographers Whilst perhaps true in the past this attitude no longer reflects the capabilities of modern instrumentalized TLC in all its many forms This volume represents the proceedings of a meeting in Brighton in 1987 which formed part of a continuing series of one and two day events on TLC organized by the Chromatographic Society either alone or like this one in conjunction with other learned bodies These meetings are designed to keep chromatographers up to date with the latest developments and help promote a more positive image of TLC Ian Wilson November 1987 v CONTENTS The Chromatographic Society xi Contemporary Thin layer Chromatography An introduction I.D. Wilson INSTRUMENTATION Quantitative Methods in Thin layer Chromatography C F Poole S K Poole and T A Dean 11 Fluorescence Line narrowing Spectroscopy A New and Highly Selective Detection Technique for Thin layer and Liquid Chromatography J W Hofstraat C Gooijer U A Th Brinkman and N H Velthorst 29 Qualitative and Quantitative Image Analysis of Fluorescence from High Performance Thin layer Chromatography R M Belchamber S J Brinkworth H Read and J D M Roberts 37 Analytical Rotation Planar Chromatography Sz Nyiredy K Dallenbach Toelke and O Sticher 45 Analytical and Preparative Overpressured Layer Chromatography E Mincsovics and E Tyihak 57 Anticircular Planar Chromatography Analytical and Preparative Aspects H Traitler and A *Forced-Flow Layer Chromatography* Erno Tyihak, 2015-12-01 Forced Flow Layer Chromatography takes a close look at the specifics of forced flow layer chromatography techniques from their evolution to the nuances of using these techniques in a variety of applications where traditional thin layer chromatography TLC and high performance thin layer chromatography HPTLC are

not as effective This book presents a number of variations of TLC techniques with special emphasis on the overpressured layer chromatography OPLC technique and newer developments such as the BioArena System for biomedical analysis The versatility of these forced flow techniques opens up new avenues for the analysis of a large number of samples for high throughput screening and for the analysis of very complex matrices while the development of BioArena extends the use of these techniques to challenging new areas of bioanalysis Details a variety of forced flow techniques explaining how they markedly reduce developing time and result in less lateral diffusion and more compact spots Emphasizes the benefits of OPLC separation techniques a method pioneered by the authors nearly forty years ago Discusses new developments such as the BioArena system used to facilitate detection isolation and identification of new antimicrobials antineoplastics biopesticides and other biologically active substances Nuclear Science Abstracts ,1976 **Physical Methods in**

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