

Journal of Current Chemical & Pharmaceutical Sciences

J. Curr. Chem. Pharm. Sc.: 3(1), 2013, 60-63 ISSN 2277-2871

ISOLATION OF ASTAXANTHIN FROM SHRIMP METAPENAEUS DOBSONI AND STUDY OF ITS PHARMACOLOGICAL ACTIVITY UMA NATH USHAKUMARI^{*} and RAVI RAMANUJAN^a

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(Received : 30.01.2013; Revised : 06.02.2013; Accepted : 07.02.2013)

ABSTRACT

The aim of the present study was to isolate and study about the antibacterial activity of astaxanthin from Shrimp of the species Metapenaeus dobsoni. Astaxanthin a carotenoid present in marine yeast and crustaceans posses a wide range of pharmacological activity. Shrimp was collected from Cochin, Kerala during the month of August 2012. The samples were collected and transported to the laboratory under iced conditions. The yield of dried shell was determined by weighing after dried at 50°C in oven for 24 h. Samples were stored at two temperatures, of 25°C and -20°C until use. The material was thawed in running water before use and homogenized in a laboratory mixer. The antibacterial activity was studied on several organisms like *Bacillus Subtilis, Salmonella typhi, Staphylococcus aureus* and *Pseudomonas aeroginosa*. The extract showed excellent antibacterial activity than the standard chloramphenicol. Among this pseudomonas aeroginasa showed maximum inhibition.

Key words: Astaxanthin, Metapenaeus dobsoni, Chemical extraction Antibacterial activity, Fermentation, Well diffusion assay

INTRODUCTION

Astaxanthin, unlike some carotenoids, does not convert to Vitamin-A (retinol) in the human body. Too much Vitamin A is toxic for a human, but astaxanthin is not. However, it is a powerful antioxidant. It is 10 times more capable than other carotenoids. While astaxanthin is a natural nutritional component, it can be found as a food supplement. The supplement is intended for human, animal, and aquaculture consumption. The commercial production of astaxanthin comes from both natural and synthetic sources¹.



Fig. 1: Astaxanthin

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The U. S. Food and Drug Administration (FDA) approved astaxanthin as a food colouring (or colour additive) for specific uses in animal and fish foods. The European Union (actually European Commission) considers it food dye within the E number system

Astaxanthin pronounced as (as-tuh-zan'-thin) is a carotenoid. It belongs to a larger class of phytochemicals known as terpenes. It is classified as a xanthophyll, which means "yellow leaves". Like many carotenoids, it is a colorful, fat/oil-soluble pigment. Astaxanthin can be found in microalgae, yeast, salmon, trout, krill, shrimp, crayfish, crustaceans, and the feathers of some birds. Professor Basil Weedon was the first to map the structures of astaxanthin. The study of the pharmacological activity of astaxanthin is vast and wide. The aim and scope of the present research work is to study the anti bacterial activity of astaxanthin^{2,3}. Commercial potential for Haematococcus microalgae as a natural source of astaxanthin⁴.

EXPERIMENTAL

Materials and methods

Sample collection

Shrimp was collected from coastal areas of Cochin, Kerala during the month of August 2012. The samples were collected and transported to the laboratory under iced conditions. The yield of dried shell was determined by weighing after dried at 50°C in oven for 24 h. Samples were stored at two temperatures, of 25°C and -20°C until use. The material was thawed in running water before use and homogenized in a laboratory mixer.

Chemical extraction of astaxanthin

Astaxanthin was extracted by mixing 5g shrimp waste powder homogenate, 50 mL of hexane and 5 mg of glass beads and vortexed for 30 seconds, place in the 50°C water bath for 10 minutes. Aqueous and organic layers were separated by 3000 rpm for 5 minutes. This step repeat until the hexane is colorless. At the final step 6 mL of di- methyl sulfoxide (DMS0) was added to the tube and vortex vigorously and place in the water bath for 10 minutes and vortex again. Concentrated carotenoid was subjected to Thin Layer Chromatography (TLC) using silica gel 60 F MERCK TLC paper⁵.

Study of antibacterial activity

Method: Well diffusion assay

Agar diffusion assay is used widely to determine the anti-bacterial activity of Leaf extract. The technique works well with defined inhibitors⁶. Nutrient agar prepared was poured in the Petri dish. 24 hrs growing culture (*Salmonella Typhi*; *Pseudomonas Aeruginosa*; *Bacillus subtilis* and *Staphylococcus Aureus*) were swabbed on it. The wells (10 mm diameter) were made by using cork borer. The different concentrations of the crude extract were loaded in the wells. The plates were then incubated at 37°C for 24 hours. The inhibition diameter was measured (Fig. 2).

Table 1 shows the diameter of zone of inhibition of extracts on various microorganism when compared to the standard. Fig. 2 shows the well diffusion assay zones produced by standards and various microorganisms.

S. No.	Microorganism	Extract	Standard
1	Salmonella typhi	20	24.5
2	Pseudomonas aeruginosa	22	21
3	Bacillus subtilis	18	24
4	Staphylococcus aureus	16	23

Table 1: Diameter of zone of inhibition



Salmonella typhi



Staphylococcus aureus



Pseudomonas aeruginosa



Bacillus subtilis

C – Control; 1- 25 $\mu g;$ 2- 50 $\mu g;$ 3- 75 $\mu g;$ 4- 100 μg

Fig. 2: Zone of inhibition

CONCLUSION

Astaxanthin is effective against gram positive and gram negative bacteria, when compared with standard chloramphenicol. However the experiments have to be tried by using different solvents. The antibacterial activity of the extract was studied by using well diffusion method salmonella typhi produced 20 mm diameter for zone of inhibition, pseudogonas aeroginasa 24 mm, bacillus subtilis 18 mm. staphylococcus aureus 16 mm. The extract showed excellent antibacterial activity than the standard chloramphenicol. Among this pseudomonas aeroginasa (Fig. 2, Table 2) showed maximum inhibition.

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