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# INVESTIGATION OF THE LIPID CONTENT OF THE ORANGE-STRIPED ANEMONE: *DIADUMENE LUCIAE* VERRIL (= *AIPTASIA IGENEA*)

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## ABSTRACT

The lipid content of *Diadumene luciae* (*Aiptasia igenea*) reached about 1.8% w/w of the fresh material. GLC analysis of the unsaponifiable matters revealed that squalene was the major saturated hydrocarbon followed by n-hexacosane, C-30, n-octacosane, n-docosane and n-tetracosane. Cholesterol was the major sterol followed by ergosterol and β-sitosterol. Analysis of the saponifiable matter revealed that oleic acid constituted the major fatty acid followed by linoleic, arachidonic, stearic, arachidic, palmitic, margaric and myristoleic.

## INTRODUCTION

The importance of the oceans and seas as a potential sources for food and new drugs has been demonestrated by several authors[1-7]. The biological and pharmacological activities of certain drugs obtained from the sea were extensively reviewed [1, 8].

*Diadumene luciae* verril=*Apitasia igenea*[9], is a sea animal which belonges to phylum Coelenterates and subphylum sea anemone. Sea anemones look like lovely flowers, resembling dahlias, chrysanthemums and the anemone after which they were named.

The tentacles around the mouth look-like long narrow petals, but they contract when exposed to dangerous environment. In the tropical seas, the ocean's bottom is almost entirely carpeted with brightly colored anemones[10]. Nothing could be traced in the available literature concerning the lipid analysis of the sea anemone: *Diadumene luciae* Verril.

This stimulated the authors to carry out an investigation of the lipid content of the sea anemone: *Diadumene luciae* Verril.

## EXPERIMENTAL

### Materials:

The sea anemone *Diadumene luciae* Verril was collected at about one feet off the rocks near the beach of Cairo University camp, at Marsa Matrogh, Egypt during August 1993. The anemone was identified by the authors and was confirmed by Prof. Dr. Helmi Bishai, Professor of Zoology, Faculty of Science, Cairo University, Egypt.

The anemone material was washed thoroughly from salt and sand with tap water. Foreign materials were removed from each anemone, packed in plastic bags and were frozen for further work. Voucher specimens are preserved in sea water containing 1% formalin in glass jars, at the Department of Pharmacognosy, Faculty of Pharmacy, Cairo University, Egypt.

## METHODS

### Lipid preparation:

One hundred grams of fresh anemone was ground in a blender and extracted with petroleum ether (60-80°C) till exhaustion. The solvent was distilled off under reduced pressure. The physical character of the residue obtained was listed in Table I.

### Saponification of lipids and preparation of the fatty acids methyl esters:

The lipoidal matter obtained was saponified adopting the method of AOAC[11]. The solvent was evaporated to dryness. The saponifiable fraction was methylated[12].

Analysis of each of the unsaponifiable and methylated saponifiable matters[13-15] was carried out on Sigma 3 B Perkin-Elmer gas chromatography equipped with dual flame ionization detector; packed column of OV-11 adopting the following conditions. Sample size was 1 µL of 10% solution in absolute ethanol. Nitrogen as a carrier gas at a flow rate of 30 ml/min was used. Hydrogen flow rate was 30 ml/min. Air flow rate was 300 ml/min; ramp rate 60°C/min. Injector temperature was 280°C. Detetor temperature was 350°C. Chart speed was 15 cm/hr. The oven was programmed from 100°C for one min to 325°C for 10 min at 10°C/min.

Identification of the hydroacrbons, sterols and fatty acids methyl esters was carried out by comparing their relative retention time with those of the pure available references, as well as, by referring to the published data on GLC analysis of lipids[13-15]. The quantitative estimation of the components of each sample of methylated fatty acids and unsaponifiable matter under investigation was based on peak area measurments

relative to other peaks in the chromatogram. Results of GLC analysis of each of the unsaponifiable and the methylated saponifiable matters are listed in Tables II and III.

## RESULTS AND DISCUSSION

GLC analysis of the methylated fatty acids of *Diadumene luciae* showed 20 components (Table II). Sixteen of them were identified as C8, C9, C10, C11, C12, C13, C14, C14:1, C15:1, C16, C17, C18, C18:1, C18:2, C20 and C20:4 fatty acids. They constituted about 95% of the total saponifiable matter. Oleic acid (20.56%) was the major fatty acid followed by linoleic (17.68%) and arachidonic (14.85%), stearic (12.15%), arachidic (9.66%), palmitic (8.88%) and margaric (6.65%) acids. It was rich in unsaturated fatty acids, where they reached abut (57%). Thus it is classified as a semi-drying oil.

The identified components amounted about 94.00% of the total unsaponifiable matter (Table III). The identified components[18], included saturated hydrocarbons of C10-C30, squalene, cholesterol, ergosterol and β-sitosterol. Cholesterol (16.38%) represented the major constituent followed by squalene (15.00%), C26 (10.60%), C30 (10.50%), C28 (10.20%), ergosterol (9.38%), C22 (6.50%), C24 (6.30%) and β-sitosterol (5.60%).

The picture of the total lipids of the *Diadumene luciae* Verril provides good information which could be helpful in its chemotaxonomy. Also, it explains the importance of the seas and oceans as a good sources for foods and drugs.

**Table I.** Physical properties of the lipids of *Diadumene luciae* Verril.

| Characters | Percentage Lipid |
|---|---|
| Total lipids | 1.80% w/w, Fresh materials |
| Unsaponifiable matter | 0.21% W/W, Fresh materials |
| Saponifiable matter | 0.20% w/w, Fresh materials |
| Colour | Brownish green |
| Odour | Characteristic |
| Taste | Oily |
| **Solubility:** | |
| Petroleum ether | Freely soluble |
| Ether | Freely soluble |
| Chloroform | Freely soluble |
| Ethanol 90% | Soluble |
| Water | Insoluble |

**Table II:** Identified methyl esters of fatty acids of *Diadumene luciae* Verril, analysed by GLC.

| Peak No | Componenets | RRt*in min. | Percentage |
|---|---|---|---|
| 01 | Caprylic (C 8:0) | 0.03 | traces ** |
| 02 | Nonanoic (C 9:0) | 0.04 | 00.14 |
| 03 | Capric (C 10:0) | 0.06 | traces |
| 04 | Undecanoic (C 11:0) | 0.08 | traces |
| 05 | Laurate (C 12:0) | 0.09 | 00.88 |
| 06 | Tridecanoic (C 13:0) | 0.22 | 00.14 |
| 07 | Myristic (C 14:0) | 0.32 | 00.14 |
| 08 | Myristoleic (C 14:1) | 0.36 | 02.59 |
| 09 | C 15:1 | 0.49 | 00.96 |
| 10 | Palmitic (C 16:0) | 0.51 | 08.88 |
| 11 | Heptadecanoic (C 17:0) | 0.83 | 06.65 |
| 12 | Stearic (C 18:0) | 0.71 | 12.15 |
| 13 | Oleic (C 18:1) | 0.90 | 20.56 |
| 14 | Linoleic (C 18:2) | 1.00 | 17.68 |
| 15 | Arachidic (C20:O) | 1.14 | 09.66 |
| 16 | Arachidonic (C 20:4) | 1.31 | 14.85 |
| Total | | | 95.28 |

* RRt. = Relative retention time to methyl linoleate (Rt.=23.1 min.)

** Traces; less than 0.14%.

**Table III.** GLC analysis of the unsaponifiable matter of *Diadumene luciae* Verril

| Peak No | Componenets | RRt in min. | Percentage |
|---|---|---|---|
| 01 | n-Decane (C10) | 0.18 | 00.56 |
| 02 | Undecane (C11) | 0.28 | 00.56 |
| 03 | Dodecane (C12) | 0.29 | 00.12 |
| 04 | n-Tridecane (C13) | 0.41 | Traces |
| 05 | n-Tetradecane (C14) | 0.52 | 00.12 |
| 06 | n-Pentadecane (C15) | 0.65 | Traces |
| 07 | n-Hexadecane (C16) | 0.78 | 00.34 |
| 08 | n-Octadecane (C18) | 1.00 | 00.35 |
| 09 | n-Bicosane (C20) | 1.28 | 01.25 |
| 10 | n-Docosane (C22) | 1.41 | 06.50 |
| 11 | n-Tetracosane (C24) | 1.59 | 06.30 |
| 12 | n-Hexacosane (C26) | 1.76 | 10.60 |
| 13 | n-Octacosane (C28) | 1.90 | 10.20 |
| 14 | C-30 compound | 1.93 | 10.50 |
| 15 | Squalene | 1.98 | 15.00 |
| 16 | Cholesterol | 2.16 | 16.38 |
| 17 | Ergosterol | 2.35 | 09.38 |
| 18 | Sitosterol | 2.45 | 05.60 |
| Toral | | | 93.70 |

RRt, Relative retention time to n-octadecane (Rt.=19.34 min.)

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## دراسة المحتوى الدهنى لزهر البحر ديادومين لوسى فرل ( = ابتازيا ايجنيا)

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فى هذا البحث تم تحديد نسبة المحتوى الدهنى لزهر البحر ديادومين لوسى فرل كما تم تحليل المواد المتصبنة وغير المتصبنة بواسطة كروماتوجرافيا الغاز لاستنتاج التركيب الكيماوى لكل منها.