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6,10,14-Trimethylpentadecan-2-one and 6,10,14-trimethyl-5-trans, 9-trans, 13-pentadecatrien-2-one from the androgenic glands of the male crab Carcinus maenas

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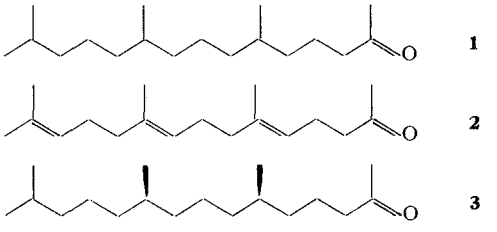
Summary. 2 C_{18} isoprenoid ketones, hexahydrofarnesylacetone (**1**) and farnesylacetone (**2**) have been identified for the first time in lipid extracts from the androgenic glands of the male crab Carcinus maenas, using coupled gas chromatography-mass spectrometry. The 2 compounds prepared by synthesis, are biologically active, inhibiting the incorporation of ^{3}H-leucine in Crustaceans ovaries subcultures.

The androgenic gland of Crustaceans is responsible for the induction and differentiation of male sexual characteristics in these animals[5]. It also inhibits the vitellogenesis in ovaries as previously shown by Charniaux-Cotton[5,6]. In vitro and in vivo bioassays have facilitated the following of the isolation of a lipidic fraction from the androgenic glands or from the hemolymph of the male crab Carcinus maenas[7,8]. From a Sephadex LH-20 column chromatography and GC, and approximate molecular weight of 250 was deduced for the active component, more active by a factor of 10^5 after the final purification than the total extract. This fraction possesses 2 of the known biological activities of the gland: inhibition of the incorporation of ^{3}H-leucine into the proteins of ovaries (Orchestia gammarella) and accumulation of astaxanthin in the secondary antennae of Talitrus saltator[9]. However, a third biological activity of the gland, the induction of spermatogenesis was never observed after injection of this fraction.

5000 glands of the male crab Carcinus maenas obtained by dissection have been submitted to extraction and isolation of the active lipid fraction as already reported[8]. After the LH-20 column chromatography, the product was analyzed by GC-MS (gas chromatograph Fractovap GI Carlo-Erba equipped with a 20 m glass capillary column, 0.32 mm i.d., coated with OV-1 and interfaced with a Varian MAT CH5 mass spectrometer). The sample injection in 1 µl of hexane solution was carried out according to the method of Grob[10]. The column temperature was programmed from 30 to 180 °C with a rate of 7 °C/min and then kept under isothermal conditions. A relatively abundant component has been identified as 6,10,14-trimethylpentadecan-2-one (**1**) (hexahydrofarnesylacetone). The molecular ion in the MS is not present. However, the following ions are found: m/e 250 $(M-18)^+$ 4%, 225 $(M-43)^+$ 1%, 210 $(M-58)^+$ 2%, 43 (79%), 58 ($C_3H_6O^+$ in agreement with a satured methylketone) 100% and 71 (59%). The MS is virtually identical with that of the synthetic product and also corresponds to the already published spectra[11]. The synthetic sample was prepared by H_2/Pt reduction of farnesylacetone (**2**) and purified by preparative column chromatography; its retention time is identical with that of the natural compound in a co-injection analysis.

6,10,14-trimethyl-5-trans, 9-trans, 13-pentadecatrien-2-one (trans, trans-farnesylacetone (**2**)) is present in the mixture as a minor component (ca. 10% of **1** as indicated by GC/MS and checked by co-injection analysis with an authentic sample). The 5-trans 9-cis or 5-cis 9-trans isomer is also present in trace amount.



1 **2** **3**

Most of the other peaks observed in GC were identified as pollutants (mainly phthalates) or fatty acid esters ranging from C_{14} to C_{18}.

In vitro bioassays with **1** have shown an inhibition of the incorporation of ^{3}H-leucine in ovaries subcultures with a concentration of 10 ng per ovary. No difference is observed, using (6R, 10R), 14-trimethylpentadecan-2-one (**3**) prepared from phytol by oxidation with chromic anhydride in acetic acid.

In vitro bioassays with **2** indicate a higher activity, an inhibition of the incorporation of ^{3}H-leucine being observed with as little as 250 pg per ovary. The C_{18}-isoprenoid ketone **1** has been found previously in recent marine sediments[11] and one of its possible source is phytol originally deriving from chlorophyll.

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5 H. Charniaux-Cotton, Ann. Sci. nat. Zool. Biol. anim. *19*, 411 (1957).
6 H. Charniaux-Cotton, in: Organogenesis. Ed. R. de Haan and H. Ursprung. Holt, Rinehart and Wilson, New York (1965).
7 J. Berreur-Bonnenfant and J. J. Meusy, C. r. Acad. Sci. Paris *275*, Ser. D, 1641 (1972).
8 J. Berreur-Bonnenfant, J. J. Meusy, J. P. Ferezou, M. Devys, A. Quesneau-Thierry and M. Barbier, C. r. Acad. Sci. Paris *277*, Ser. D, 971 (1973).
9 M. Barbier, H. Charniaux-Cotton and M. C. Fried-Montaufier, C. r. Acad. Sci. Paris *263*, Ser. D, 1508 (1966).
10 K. Grob, Chromatographia *5*, 3 (1972).
11 R. Ikan, M. J. Baedeker and I. R. Kaplan, Nature *244*, 154 (1973).