Phytochemistry, Vol. 28, No. 1, pp. 71–72, 1989.
Printed in Great Britain.

0031-9422/89 $3.00+0.00

CARRAGEENANS FROM TETRASPOROPHYTIC AND GAMETOPHYTIC STAGES OF *CYSTOCLONIUM PURPUREUM*

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(*Received* 26 *April* 1988)

Key Word Index—*Cystoclonium purpureum*; Rhodophyceae; Rhodophyllidaceae; iota-carrageenan; IR; ^{13}C NMR.

Abstract—IR and ^{13}C NMR spectroscopy demonstrated that iota-carrageenans are present in the gametophytic as well as in the tetrasporophytic stages of *Cystoclonium purpureum*.

INTRODUCTION

Carrageenans are sulphated polysaccharides present in the cell walls of Gigartinales. They are extensively used as gels and thickening agents in food and industrial preparations. Chemical differences are known to exist among carrageenans from the different phases of the life history of some carrageenophytes. The members of the Gigartinaceae family and some species in the Phyllophoraceae present a biochemical alternance in the structural composition of natural polyholosides [1–4]. On the other hand, Doty *et al.* [5] did not find any differences in carrageenan composition of the life history phases of six *Eucheuma* species, a genus of the family Solieriaceae. *Cystoclonium purpureum* , another Gigartinale, of the family Rhodophyllidaceae, was examined by Deslandes *et al.* [6] and found to contain a certain amount of carrageenans belonging to the iota family, but vegetative thalli only were examined.

We have examined the carrageenans isolated from two phases of the life history of *Cystoclonium purpureum*, the female gametophyte and the tetrasporophyte, and compared their chemical structure.

RESULTS AND DISCUSSION

In agreement with the conclusions of Deslandes [7] concerning plants from the Brittany coast, we found, after alkaline transformation, 20–25% carrageenans as an average for *C. purpureum* from Normandy. Carrageenan quantities changed with the seasons: maximum in July (25%) and minimum in September (6%). On the other hand, there was no difference between the quantity of product isolated from tetrasporophytes or from female gametophytes.

The IR spectral analysis of carrageenans from *C. purpureum* confirms a iota-carrageenan structure type with characteristic bands at 1240 (S=O of SO_4), 930 (3,6-anhydrogalactose), 845 (C–O–S of axial SO_4–4) and 805 cm^{-1} (SO_4-2 of 3,6-anhydrogalactose) in agreement with previous results [8]. In addition, IR spectra of tetrasporophytes and female gametophytes show that, in contrast to *Chondrus* [9], there is no difference between products extracted from tetrasporophytic and gametophytic stages. The ^{13}C NMR spectra complete and confirm these results by showing the presence of a iota-carrageenan structure, characterized by IG_1 and IA_1 signals which appear at δ 104.4 and 94.3 respectively.

The chemical shift values (δ) are given in Table 1. Our spectral data for τ-carrageenans are in accordance with the values reported by Usov [10]. The presence of some additional signals at δ 100 or 102 and δ 107 or 108 were attributed to a small proportion of ν-carrageenan [5], a precursor of τ-carrageenan. Our work shows that there is no fundamental difference between structures of carrageenans extracted from different generations of *C. purpureum*. All produce iota-carrageenans.

EXPERIMENTAL

Algal material. Cystoclonium purpureum (Hudson) Batters is a red alga (O. Gigartinales). The thallus is 15–50 cm long, erect and consists of a cylindrical axis repeatedly and irregularly branched. Branches are attenuated at the bases and apices. This species is abundant in spring and summer and grows on rocks and stones in the midlittoral and lower. The greatest abundance of *Cystoclonium* is reached in Sept. on the Normandy coasts.

Table 1. ^{13}C NMR spectra for materials A (vegetative *C. purpureum*) B (tetrasporophytic plants) and C (gametophytic plants)

A	B	C	*Assignment
107	107	108	Y
104,4	102,4	104	IG_1
94,3	94,1	93,6	IA_1
100,6	102	100,2	X
80,3; 80	80,7; 80,1	80; 79	IA_3; IA_4
79,3; 79,1	79; 79,5	77; 77,5	IA_5; IG_3
77,1; 77	77,5; 77	74; 73,5	IG_5; IA_2
74,3	72,3	71,9	IG_4
71;72	71,5; 70,9	70,5; 70	IA_6; IG_2
63,5	63,4	63,5	IG_6

*I: iota; G: galactose; A: anhydrogalactose; Y, X signals for ν precursor.

As with the majority of Gigartinales, the life history of *C. purpureum* varies between three generations. Gametangial plants are dioecious and there is no morphological difference between gametophytes and tetrasporophytes.

Generations may be distinguished only when plants are sexually mature: thus female gametophytes are easily recognized by numerous cystocarps forming prominent swellings on the ramifications and tetrasporophytes due to the presence of zonate tetrasporocysts, immersed in the external cortex of the upper part of frond. Maximal fertility appears in summer on the Normandy coasts.

Extraction and analysis. The material was divided into three lots after examination under a stereomicroscope. (A): Vegetative thalli; (B): tetrasporophytes; (C): female gametophytes bearing cystocarps. In each case, the thalli were washed with dist. H_2O and dried in a ventilated enclosure at 50–60°.

Two kinds of extraction were performed: extraction after alkaline transformation (industrial method) and total extraction. In the first case, 20 g of dry material were treated by a hot soln of aq. KOH (10% at 80°) for 20 min, crushed, filtered and boiled for 5 hr. Carrageenans poured into EtOH pptd.

In the second case, 20 g of dry material was treated by hot H_2O at 80° for 20 min, filtered and pptd as above. Carrageenans were analysed by IR and ^{13}C NMR spectroscoppy. ^{13}C NMR: 50.1 MHz, D_2O/TMS-DSS at 80°, 25 000 accumulations, pulse 14 μsec, angular pulse 60°, acquisition 3 sec.

Material A.: IR ν_{max}^{KBr} cm^{-1}: 1240, 930, 845, 805; ^{13}C NMR (50, 1 MHz, D_2O/TMS-DSS): table 1.

Material B: IR ν_{max}^{KBr} cm^{-1}: 1240, 930, 845, 805; ^{13}C NMR (50, 1 MHz, D_2O/TMS-DSS): table 1.

Material C: IR ν_{max}^{KBr} cm^{-1}: 1240, 930, 845, 805, ; ^{13}C NMR/50, 1 MHz, D_2O/TMS-DSS): table 1.

Acknowledgements—This research was supported by ANVAR of Basse-Normandie (France). We thank Dr Maume for ^{13}C NMR spectra.

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