Phytochemistry, 1974, Vol. 13, pp. 957 to 959. Pergamon Press. Printed in England.

n-PARAFFIN COMPOSITION OF SOME LIVERWORTS*

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(*Received* 2 *September* 1973. *Accepted* 19 *November* 1973)

Key Word Index—Hepaticae; Liverworts; *n*-paraffins; chemotaxonomy.

ALIPHATIC hydrocarbons, although detected in almost all organisms, are known in plants to be important constituents of the epicuticular waxes which act as protective coatings. It has been suggested that many in plants, the composition of the hydrocarbon fraction is characteristic of the species, and thus can be used as a taxonomic aid. The distribution and metabolism in the higher plant paraffins have been the subject of several reviews.[1–5]

However, in the liverworts (Hepaticae), the hydrocarbon fraction has been hardly investigated.[6–8] In connection with our previous investigations on the liverworts, we have now examined the hydrocarbon fraction in 5 species, *Bazzania pompeana* (Lac.) Mitt., *Scapania parvitexta* Steph., *Isotachis japonica* Steph., *Pellia fabbroniana* Raddi and *Macrodiplophyllum plicatum* (Lindb.) Perss, all belonging to the Jungermanniales.

RESULTS AND DISCUSSION

Crude waxes were extracted and chromatographed over a silica gel column with hexane to separate the fastest-moving fractions as a colorless crystalline mixture, which showed a single spot (R_f 0·75) on TLC in the same system, and whose IR and NMR spectra revealed the substances to be *n*-paraffins. Furthermore, the mixture showed several components by GLC, which could be assigned as *n*-paraffin homologues by comparison with several authentic compounds (C_{18}, C_{22}, C_{24} and C_{28}) and by retention time. The

* Part XVI in the series "Chemical Constituents from Hepaticae". For Part XV see MATSUO, A., MAEDA, T., NAKAYAMA, M. and HAYASHI, S. (1973) *Tetrahedron Letters* 4131.

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paraffins were also submitted to GC–MS and showed MS with the pattern expected of *n*-paraffins.[9] [10]

TABLE 1. YIELDS OF CRUDE WAX AND CONTENTS OF *n*-PARAFFIN FRACTION IN LIVERWORTS

Species	Yield of crude wax from dry plant (%)	Content of *n*-paraffin fraction in wax (%)
Bazzania pompeana	0·79	0·9
Pellia fabbroniana	0·51	0·2
Isotachis japonica	0·18	0·4
Scapania parvitexta	0·14	1·0
Macrodiplophyllum plicatum	0·61	1·3

The yield of crude wax and the percentage of *n*-paraffins from the 5 liverworts are given in Table 1. Gas chromatograms of the 5 liverworts are represented in Fig. 1. *n*-Paraffins of the liverworts ranged from C_{15} to about C_{35}, and in general the *n*-paraffin homologues were distributed in two groups at C_{17}–C_{18} and C_{29}–C_{33} respectively although *M. plicatum* contained the *n*-C_{23}-paraffin, which is a main component in mosses,[11] as the major hydrocarbon. In addition, odd-carbon paraffins were usually predominant over even-carbon ones, as found for higher plants. However, this was not always found at the lower carbon numbers (see Fig. 1). The overall ratio of odd to even members were lower compared with that of higher plants, and this may be due to the liverworts being lower in the evolutionary scale.[6]

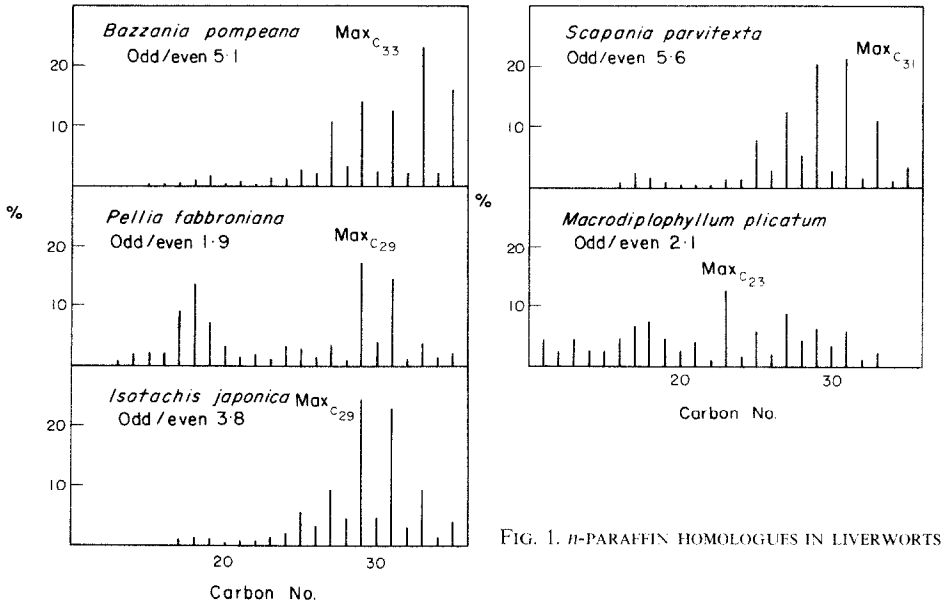

FIG. 1. *n*-PARAFFIN HOMOLOGUES IN LIVERWORTS

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EXPERIMENTAL

IR spectra were taken in CCl_4, and NMR spectra on a 60 MHz spectrometer in $CDCl_3$ using TMS as an internal standard.

Materials. B. pompeana and *P. fabbroniana* were collected at the suburbs of Hiroshima City, *I. japonica* and *S. parvitexta* at Yakushima of Kagoshima Prefecture, and *M. plicatum* in Hokkaido. The liverworts, after being dried in the shade for several days, were digested with hexane for a week at room temp., crude waxes being obtained as semi-solid substances (see Table 1).

Separation of n-*paraffin fractions.* 1·0 g of each of crude waxes was chromatographed over a column (1·9 × 50 cm) of silica gel (50 g) with hexane, and the fastest-moving fraction was collected as a colorless crystalline substance, which showed 1 spot (R_f 0·75) on TLC using silica gel and hexane, and whose IR and NMR spectra exhibited characteristic *n*-paraffin signals: ν_{max} 2920, 2860, 1430, 1380, 720 cm^{-1}; δ ppm 0·90 (*t*, *J* 5·0 Hz), 1·23 (*s*).

GLC analyses of n-*paraffin fractions.* GLC analyses of the paraffin fractions were carried out on a FID-type gas chromatograph by using two kinds of stainless-steel columns (2 mm × 2 m) packed with silicon SE 30 (10%) on Diasolid L (60–80 mesh) and silicon OV 1 (1%) on Uniport B (60–80 mesh) at the temp. programming of 3°/min from 150° to 300° with 15 ml/min of N_2.

GC–MS analyses on n-*paraffin fractions.* GC–MS analyses were performed on a combined Hitachi RMS-4 single-focus mass spectrometer and Hitachi K 53 gas chromatograph under the following conditions: silicon SE 30 (2%) on Diasolid L and silicon OV 1 (1%) on Uniport B, column temp. programming of 2°/min from 150° to 300°, 0·8 kg/cm^2 pressure of He carrier, 80 μA total emission, 1500 eV ion-accelerating voltage and 200° ionization chamber temp.