tute of Pharmacy, University of Oslo. *Previous work.* [1,2].

Plant part examined. Dried leaves and stems of *Cannabis sativa* were milled and extracted with light petrol. After drying, the residual material was extracted with H_2O at 40° under stirring for 2 hr followed by dialysis against H_2O. The non-dialysable material was purified by ion exchange chromatography on DEAE-cellulose and subsequent gel giltration on Sepharose 4B. The product consisted of polysaccharide with protein attached.

The component sugars were analysed by GLC after methanolysis and trimethylsilylation of the resulting methyl glycosides [3]. The trimethylsilyl ethers were separated on a column (200 × 0·3 cm) of SE 52 on Varaport 30, using a N_2 gas flow of 33 ml/min and a temperature program with start at 140° and an increase of 2°/min for 10 min, followed by an increase of 4°/min. Under these conditions the derivatized *N*-acetylgalactosamine and *N*-acetylglucosamine appeared at 197° and 200°, respectively. Quantitative GLC with mannitol as the internal standard resulted in a content of 6·7% *N*-acetylglucosamine and 3·1% *N*-acetylgalactosamine. The presence of the two amino sugars was confirmed by automatic amino acid analysis on a BioCal BC 200 Analyzer following acid hydrolysis with 6 N HCl at 110° for 20 hr. By this method a content of 5·3% *N*-acetylglucosamine and 1·9% N-acetylgalactosamine was found. The lower values obtained by the latter procedure are explained by partial degradation of hexosamine, known to occur under strong conditions of hydrolysis [4].

The purified biopolymer (0·2 g) was hydrolysed with 4 N HCl at 100° for 6 hr. After evaporation of HCl the hexosamines and amino acids were adsorbed to a column (2·2 × 6 cm) of AG 50 × 8 (H^+) resin. Neutral sugars were removed by washing with water followed by eluting the column with 2 N HCl (40 ml). The acid eluate was concentrated to dryness, and the residue subjected to TLC on Si gel in the solvent system (v/v) *n*-PrOH–NH_3 25% (13:7), and to PC in the solvent systems (v/v) EtOAc–pyridine–HOAc–H_2O (5:5:1:3), and *n*-BuOH–pyridine–1 N HCl (5:3:2) using as locating reagents: (a) Aniline oxalate or $AgNO_3$–NaOH, (b) ninhydrin, and (c) periodic acid–benzidine [5]. In all three solvent systems spots were obtained having the mobility and giving the same colour reactions upon spraying as the authentic hydrochlorides of glucosamine and galactosamine. Mannosamine hydrochloride, also run as a reference, was clearly separated from the two other hexosamines.

It is concluded that galactosamine is present as an integral part of a carbohydrate–protein polymer in *Cannabis sativa*. In the quantitative analyses the results are given as *N*-acetylhexosamine since these sugars almost always occur as *N*-acetyl derivatives in nature. Whereas glucosamine has been found to occur in tissues of a number of higher plants [6], the presence of galactosamine, although reported [7,8] has never been established conclusively.

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SUCROSE OCTAACETATE FROM *CLEMATIS*

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(*Revised received* 7 *July* 1975)

Key Word Index—*Clematis chinensis*; *C. apiifolia*; Ranunculaceae; bitter material; sucrose octa-acetate.

We wish to report the isolation of sucrose octa-acetate as a natural material from *Clematis* spp. This is the first occasion that a disaccharide peracetate has been found as a natural product.

From the fraction extracted with ether of dried Radix Clematidis (roots of *Clematis japonica*), which has been used as a diuretic or rheumatic, needles mp 84–86°, having a bitter taste were obtained in a yield of 0·15%. On the basis of IR, NMR and mass spectral evidence [1, 2] and the elementary analysis, a formula $C_{12}H_{22}O_{11}$-$(C_2H_2O)_8$ was given to this compound, that of an octaacetyl disaccharide. Finally, it was identified as sucrose octa-acetate by comparison of its TLC, IR, NMR and mass spectra with those of the authentic sample prepared by the known method [3] and by its failure to depress the m.p. of an admixture with the synthetic sample. The octa-acetate was also found in freshly collected roots of *Clematis chinensis* Osbeck and *C. apiifolia* DC. and is regarded as a new type of naturally occurring bitter principle.

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EXPERIMENTAL

Isolation of sucrose octa-acetate. Dried chips of Radix Clematidis (1·0 kg) were percolated with 3 l. 95% EtOH. The EtOH extract, after evaporation to dryness was exhaustively extracted in a Soxhlet with Et_2O. The residue from the ether crystallized from petrol. After 2 further recrystallization from ether, it was obtained as colorless needles, mp 84–86°, with $[\alpha]_D$ + 62° (*c* 1·0 in $CHCl_3$). The m.p. was unchanged by admixture of an authentic sample of sucrose octa-acetate. TLC (Si gel G); R_f 0·83 (CH_2Cl_2:C_6H_6 = 1:1). Elem. Anal., Found: C, 49·41; H, 5·82. Calc. for $C_{28}H_{38}O_{19}$: C, 49·56; H, 5·69%. ν_{max}^{KBr} 1740, 1250 (broad); 3480 cm^{-1} (overtone) [–OCOMe]. $\delta_{TMS}^{CDCl_3}$ 2·00 (3H), 2·04 (3H), 2·01 (15H), 2·18 (3H) [–OCOC<u>H</u>$_3$], 4·10–4·40 (6H), 4·72–5·76 (5H) [–C<u>H</u>–OCOMe]. Mass, *m/e*; 331, 229, 271, 211, 169, 109.

Identification of sucrose octa-acetate. Dried roots of (*C. chinensis* or *C. apiifolia*) were exhaustively extracted by Soxhlet with Et_2O. The ethereal solution was evaporated to dryness. The residue was chromatographed on a thin or thick-layer Si gel G plate in CH_2Cl_2–C_6H_6 (1:1). The component isolated from the thick-layer plate (R_f 0·75–0·85) was a crystalline solid, which was assigned as sucrose octa-acetate on the basis of the R_f value, mp, IR and NMR spectra.

Acknowledgements—We thank Prof. M. Tamura, Osaka University, for identifying the plant of *C. chinensis* and Prof. S. Kawano, Toyama University, for a sample of *C. apiifolia.*

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Phytochemistry, 1976, Vol. 15, p. 327. Pergamon Press. Printed in England.

THE IDENTIFICATION OF LENZITIN AS OOSPONOL*

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(*Received* 19 *June* 1975)

Key Word Index—*Gloeophyllum sepiarium*; *Oospora astringenes*; fungi; lenzitin; oosponol; antibiotic; antifungal activity.

Lenzitin, an antibiotic which from a basidiomycete *Gloeophyllum sepiarium* (Wulf. ex Fr.) Karst [1] but has never been examined chemically to date. We now present evidence which shows that it is identical with oosponol (4-ω-hydroxyacetyl-8-hydroxy isocoumarin), a metabolite from *Oospora astringenes* Yamamoto [2–5].

Lenzitin, thin yellow needles of mp 172°, analysed for $C_{11}H_8O_5$, showed IR absorptions at 3470 cm^{-1} (OH), 2500–3200 cm^{-1} (chelated OH), 1710 cm^{-1} (free C=O), 1690 cm^{-1} (chelated C=O of lactone), 1625 and 1568 cm^{-1} (isocoumarin). Its UV spectrum exhibited λ_{max} 215 nm (log ε 4·45), 256 nm (4·04), 333 nm (3·84) with an inflexion at 233 nm. The MS gave peaks of M^+ 220, *m/e* 202, 189, 161, 144, 105, 77, 63, 51. NMR in $CDCl_3$ (100 M Hz) showed signals at 1·56 δ (1H, *b s*, alcoholic OH), 4·63 δ (2H, *d*, *J* 5 Hz, CH_2 adjacent to C=O), 7·85 δ (1H, *s*, C-3), 7·08 δ (1H, *d*, *J* 8 Hz, C-7), 7·69 δ (1H, *t*, *J* 8 Hz, C-6), 8·04 δ (1H, *d*, *J* 8 Hz, C-5). Catalytic hydrogenation caused a shift of λ_{max} to 230, 245 and 311 nm indicating that the hydrogenated product has a dihydroisocoumarin nucleus. The mono-*p*-nitrophenylhydrazone, orange cubes mp 210·5°, showed CO absorption at 1690 cm^{-1} rather than 1710 cm^{-1} in the IR.

The above spectroscopic and chemical data suggested that lenzitin was oosponol. This was confirmed by direct comparison of IR and NMR spectra of the two compounds and a mp determination.

Biological significance. The antibiotic activity of lenzitin or oosponol against Gram-positive and Gram-negative bacteria is reportedly not very strong [1,2]. Now we found that it possesses strong antifungal activity. It inhibited the growth of *Candida albicans*, *Aspergillus fumigatus* and *Trichophyton asteroides* at the concentration respectively of 1·56, 1·56 and 3·12 μg/ml, as measured by liquid dilution method.

EXPERIMENTAL

The IR spectrum was measured in KBr discs, and the UV spectrum in EtOH.

Acknowledgement—We wish to thank Professor Y. Yamamoto for his kind donation of the sample of oosponol.

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* Part 3 in the series *Studies on Fungal Products.* For Part 2 see Kanazawa, T. and Nakajima, S. (1973) *Proc. Hoshi Pharm.* **15**, 44.