different properties from ordinary soft polymers.[8]

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RECEIVED DECEMBER 10, 1954

(8) S. H. Muthana and H. Mark, *J. Polymer Sci.*, **4**, 531 (1949).
(9) X-Ray structure determinations.
(10) Viscosity and density determinations.
(11) Infrared spectra determinations.
(12) Polymerization of α-olefins.

THE ISOLATION OF α-PELTATIN GLUCOSIDE FROM THE RHIZOMES OF *PODOPHYLLUM PELTATUM L.*

Sir:

The antimitotic substances obtained from the American species of Podophyllum (*P. peltatum L.*) are present in the form of sugar-free compounds isoluble in water as well as in the form of fairly readily soluble derivatives of these compounds with glucose. The aglucones, which are soluble in chloroform, can be isolated from the crude drug together with the resins: from this fraction, three different substances could be crystallized, *viz.*, podophyllotoxin, which is also found in *Indian P. emode* Wall., and in species of Juniperus,[1] and α- and β-peltatin,[2] which are characteristic of American Podophyllum.

The glucosides hitherto isolated from *P. peltatum* are derivatives of podophyllotoxin and β-peltatin.[3] Each of these two natural products is a white amorphous powder which, on treatment with β-glucosidase, easily splits into the crystalline aglucone and 1 mole D-glucose. We have now succeeded in isolating a further uniform glucoside from the more water-soluble fractions obtained by partition chromatography between different solvents during the isolation of the other two glucosides. In comparison with the previously known amorphous Podophyllum glucosides, the new glucoside readily crystallizes, a fact that facilitates its isolation.

The new glucoside crystallizes from acetone in colorless, long prisms which melt, with decomposition, at 168–171°. The specific rotation is $[\alpha]^{20}D$ −128.9° (*c* 0.5 in methanol). Analysis confirmed the formula $C_{27}H_{30}O_{13}$ (calcd. C, 57.65; H, 5.38; O, 36.98; OCH_3, 11.03. Found: 57.77; H, 5.50; O, 37.08; OCH_3, 10.93). The ultraviolet absorption spectrum exhibits a maximum at 280 mμ (log ε 3.51). The glucoside is readily soluble in alcohol and fairly readily soluble in water. The reaction with ferric chloride is positive: the color being rust-red in water and green in alcohol. These positive color reactions indicate the presence of a free phenolic hydroxyl group; thus, the new glucoside differs distinctly from the two hitherto known amorphous glucosides of *P. peltatum* which do not show any color reaction with ferric chloride.

On acetylation with acetic acid anhydride in pyridine, the penta-acetyl derivative, which crystallizes from methanol in bunches of thin, white prisms, can be obtained. It melts at 222-223° and has an optical rotation $[\alpha]^{20}D$ −96.0° (*c* 0.5 in chloroform). Analysis led to the formula $C_{37}H_{40}O_{18}$ (calcd. C, 57.51; H, 5.22; O, 37.27; OCH_3, 8.03; 5 $COCH_3$, 27.85. Found: C, 57.30; H, 5.46; O, 37.04; OCH_3, 7.98; 5 $COCH_3$, 27.60). This derivative is practically insoluble in water, and the ferric chloride reaction is negative.

The new glucoside is easily hydrolyzed by β-glucosidase at *p*H 5 into aglucone and sugar. The aglucone can be readily obtained in pure crystalline form because of the uniformity of the starting material, and it has been found to be identical with α-peltatin (Ib). The compound, which crystallizes from absolute alcohol in flat prisms, melts at 242–246°[4]; the optical rotation is $[\alpha]^{20}D$ −124.5° (*c* 0.5 in chloroform).[4] The mixed melting point with an authentic sample,[5] further purified by chromatography, showed no depression. The analysis confirmed the formula $C_{21}H_{20}O_8$ (calcd. C, 63.00; H, 5.04; O, 31.97; OCH_3, 15.50. Found: C, 62.98; H, 5.06; O, 31.81; OCH_3, 15.69). In the ultraviolet absorption spectrum there is a maximum at 274 mμ (log ε 3.40) which is in good agreement with the value given for α-peltatin in the literature.[4]

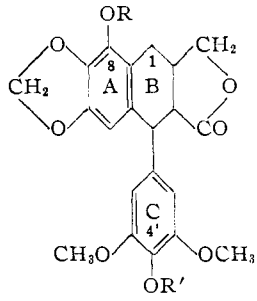
For further characterization, the aglucone was acetylated; this resulted in an acetyl derivative, which crystallizes in prisms from absolute alcohol. The melting point was 233–234°, the optical rotation was $[\alpha]^{20}D$ −113.2° (*c* 0.5 in chloroform). Analysis confirmed the presence of a diacetate and therefore the formula $C_{25}H_{24}O_{10}$ (calcd. C, 61.98; H, 4.99; O, 33.03; OCH_3, 12.81; $COCH_3$, 17.77. Found: C, 62.10; H, 5.12; O, 32.84; OCH_3, 12.86; $COCH_3$, 1758).

The sugar obtained on enzymatic cleavage could be identified as D-glucose in the form of α-methyl-D-glucoside<1.5>.

As α-peltatin contains two free phenolic hydroxyl groups, one in ring A and one in ring C, it was still uncertain as to which hydroxyl group the sugar residue was attached to. Methylation, by means of diazomethane of the hydroxyl group in the glucoside, produced an amorphous product which melted at 154–156° and showed an optical rotation $[\alpha]^{20}D$ −122.4° (*c* 0.5 in methanol) and $[\alpha]^{20}D$ −169.4° (*c* 0.5 in pyridine). The analysis of a sample dried in a desiccator confirmed the formula $C_{28}H_{32}O_{13}\cdot 1\ H_2O$ (calcd. C, 56.56; H, 5.76; OCH_3, 15.66. Found: C, 56.63; H, 5.90; OCH_3, 15.58). A maximum in the ultraviolet absorption spectrum occurs at 280 mμ (log ε 3.42). The product of methylation is, therefore, identical in all respects with 8-O(-β-D-glucopyranosyl)-β-peltatin (II),[3] previously isolated from *P. peltatum*.

(1) J. L. Hartwell, J. M. Johnson, D. B. Fitzgerald and M. Belkin, THIS JOURNAL, **75**, 235 (1953).
(2) J. L. Hartwell, *ibid.*, **69**, 2918 (1947); J. L. Hartwell and W. E. Detty, *ibid.*, **70**, 2833 (1948); **72**, 246 (1950).
(3) A. Stoll, A. von Wartburg, E. Angliker and J. Renz, *ibid.*, **76**, 5004, 6413 (1954).
(4) J. L. Hartwell and W. E. Detty, *ibid.*, **72**, 246 (1950), reported the melting point of α-peltatin, which was not quite free of β-peltatin, as 230.5–232.5° and the optical rotation as $[\alpha]^{20}D$ −120° in chloroform. The somewhat lower values seem to be due to the small admixture of β-peltatin; this was not the case with our preparations which were obtained from the uniform glucoside.
(5) Our thanks are due to Messrs. S. B. Penick Co., New York, for making available a sample of α-peltatin.

The results of our analysis of the degradation-reactions and of the methylation indicate that the new glucoside has the structure 8-O(-β-D-glucopyranosyl)-α-peltatin (Ia).



Ia, α-peltatin-β-D-glucoside (R = glucose residue, R′ = H)
Ib, α-peltatin (R = R′ = H)
II, β-peltatin-β-D-glucoside (R = glucose residue, R′ = CH_3)

The three lignane compounds occurring in the resin fraction of the American plant *Podophyllum peltatum L., viz.*, podophyllotoxin, β-peltatin and α-peltatin, are thus also present in the form of glucosides. The glucoside described here also exhibits antimitotic activity. Details of the isolation and properties will be published shortly in *Helvetica Chimica Acta*.

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RECEIVED JANUARY 24, 1955

STUDIES ON ADRENOCORTICOTROPIN. XI. A PRELIMINARY COMPARISON OF CORTICOTROPIN-A WITH β-CORTICOTROPIN

Sir:

The recent publication[1] of a tentative structure for β-corticotropin, prompts us to report the status of our work on corticotropin-A. Since the two materials are from the same source but isolated by different techniques, it will be of interest to determine the differences, if any, between them.

In previous publications we have shown the first nine positions at the amino end,[2] and the last eleven positions at the carboxyl end[3] of corticotropin-A. These sequences are identical with those published by the Cyanamid group for β-ACTH. Additionally, in our publication on the carboxyl end, we proposed tentatively a further sequence of seven amino acids. This sequence, with the addition of a residue of tyrosine,[4] we now believe to be correct. The corrected sequence is shown in Table I, positions 21–28. Although the Cyanamid

(1) P. H. Bell, THIS JOURNAL, **76**, 5565 (1954).
(2) W. F. White and W. A. Landmann, *ibid.*, **77**, 771 (1955).
(3) W. F. White, *ibid.*, **76**, 4194 (1954).
(4) In our early work tyrosine (and methionine) was destroyed during acid hydrolysis of fractions isolated by paper chromatography. In later work this difficulty was eliminated by carefully washing the paper with dilute formic acid before use.

TABLE I

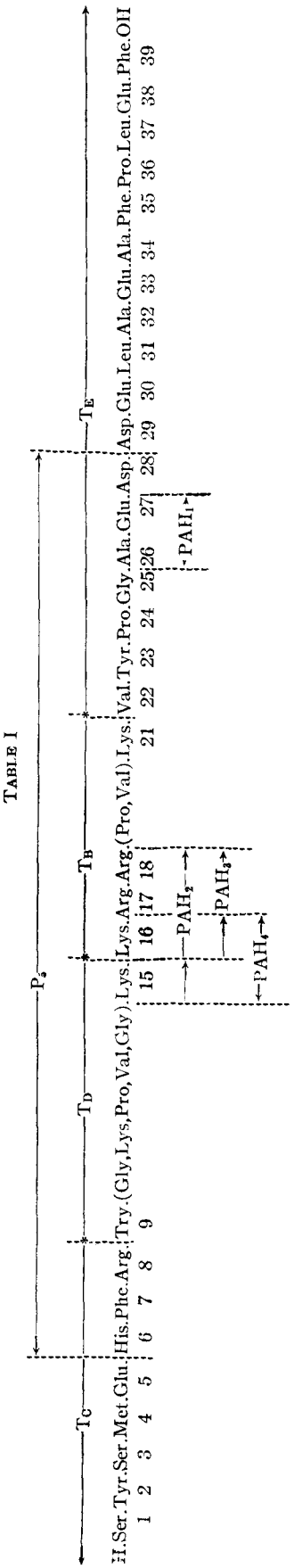

TABLE II

Peptide No.	Part.[a]	Rf 2-But.-NH_3[b]	W & W[c]	Iono-graphy dist., cm.[d]	Amino Acids on Complete Acid Hydrolysis[e]	N-Terminal Determination	C-Terminal Determination	Structure
PAH_1	0.34	Glu	...	−6.0	Ala,Glu	DNFB: Ala	...	Ala.Glu
T_D	.26	Tyr	...	...	Gly_2Lys_2Pro,Val	Aminopeptidase[f]: Try	Carboxypeptidase: Lys	Try(Gly₂Lys,Pro,Val)Lys
T_B	.05–.10	Zero	0.85xLys	...	Lys_2Arg_2Pro,Val	Aminopeptidase: Lys,Arg_2	Carboxypeptidase: Lys	(Lys,Arg,Arg)(Pro,Val)Lys
PAH_2	.05	Zero	.66xLys	...	Lys,Arg	DNFB: Both bis-DNP-Lys and ε-DNP-Lys	...	Lys(Lys,Arg,Arg)
PAH_3	.03	Glu-	.57xLys	...	Lys_2Arg	DNFB: Both bis-DNP-Lys and ε-DNP-Lys	...	Lys (Lys,Arg)
PAH_4	.05	Glu	.75xLys	...	Lys	DNFB: Both bis-DNP-Lys and ε-DNP-Lys	...	Lys.Lys

[a] *n*-Butanol–water–acetic acid (4:5:1). [b] 2-Butanol–ammonia (3:1). The R_f's are given in terms of the nearest amino acid. [c] *n*-Butanol–acetic acid–water–pyridine (30:6:24:20), S. G. Waley and J. Watson, *Biochem. J.*, **55**, 328 (1953). Since the system is used in an extended run, the R_f's are given with reference to the R_f of lysine. [d] 20 hours at 100 volts on Whatman 3 paper in 0.1 *N* ammonium acetate buffer (*p*H 6.6). As reference, glutamic acid moved −9.4 cm. [e] The subscripts indicate the number of residues of the amino acid as shown by quantitative measurement. [f] Aminopeptidase preparation made according to E. L. Smith, *J. Biol. Chem.*, **153**, 627 (1944).