

The Depolymerization of Dextran in Non-aqueous Solutions

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It is previously known¹ that dextran is successively depolymerized in aqueous solutions in the presence of acids, enzymes or alkalies. The depolymerization by acids and by enzymes has the character of a hydrolysis. Methods have also been described for the depolymerization of dextran by heat and by ultrasound.

We have found that dextran can also be successively depolymerized by alcoholysis in organic solvents with free hydroxyl groups. The monovalent aliphatic alcohols are poor solvents for dextran. However, at temperatures above 140° C dextran dissolves rapidly in polyvalent alcohols, *e. g.* ethylene glycol, glycerol, or sorbitol. In such a solution dextran slowly reacts with the solvent, but the reaction proceeds more rapidly, if the solution is heated to a higher temperature (up to about 200° C) or if a small quantity (about 0.1 to 1 %) of an acid catalyst (phosphoric acid, trichloroacetic acid or acid sodium sulphate) is added. In the same way dextran reacts with phenols, *e. g.* resorcinol.

This type of reaction was first carried out with starch by Zulkowsky². Berner found in his investigations on lichenin³, inulin⁴ and starch⁵ that the depolymerization of polysaccharides in glycerol solution is an alcoholysis.

The depolymerization reaction can easily be followed by measuring the viscosity of samples taken out at intervals, as the intrinsic viscosity of dextran is approximately proportional to its average molecular weight⁶.

By controlling temperature, catalyst and time in carrying out the alcoholysis, it is thus possible to obtain dextran glycosides with any desired average molecular weight.

The best way to isolate the reaction products from the solution is to add several volumes of water and then to precipitate them by adding a water-soluble organic liquid in which dextran is almost insoluble, *e. g.* ethyl alcohol or acetone. As the solubility of the dextran glycosides in this mixture decreases with increasing mole-

cular weight of the reaction product and increasing amount of precipitating agent, it is possible to make a fractionated precipitation and thereby obtain preparations with different average molecular weights.

If a partial depolymerization of dextran is carried out in glycerol solution and the reaction product is subsequently fractionated to give a preparation with an average molecular weight of about 70 000, a product is obtained which has proved to be excellent for the manufacture of blood plasma substitute.

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Dehydropodophyllotoxin, a New Compound Isolated from *Podophyllum peltatum* L.

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In his first report on the isolation of α -peltatin from podophyllin Hartwell¹ described the substance as exhibiting a strong blue fluorescence in solution, contrary to podophyllotoxin, which was reported to be but weakly fluorescent. The statement was amended in a subsequent paper², and none of the natural lignan-derivatives so far isolated from *Podophyllum peltatum* L. is in fact fluorescent when sufficiently purified.

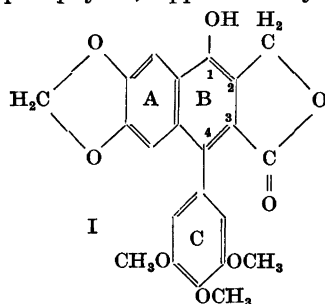
During a systematic reinvestigation of podophyllin by chromatography using sorbed formamide as a stationary phase and benzene as an eluent we noticed an intense blue fluorescence of certain fractions of the eluate between those containing podophyllotoxin and those containing α -peltatin.

Paper-chromatographic analysis of these and of other fractions by the method previously reported by us³ revealed five,

possibly six different spots detectable by their more or less pronounced blue fluorescence in ultra-violet radiation. Traces of these compounds may well have been responsible for the apparent fluorescence of Hartwell's first preparations of α -peltatin and podophyllotoxin.

The substance corresponding to the largest spot on the paper-chromatograms has been isolated as colourless needles (from ethanol). It melted at 272–274° C (corr.). (Found: C 63.6; H 4.8; 4.8; 3(OCH₃) 22.0. Calc. for C₂₂H₁₆O₈ (410.1): C 64.4; H 4.4; 3(OCH₃) 22.7).

Only minute quantities can be isolated from podophyllin, approximately 0.1 %.



The structure (I), 1-hydroxy-4-(3,4,5-trimethoxyphenyl)-6,7-methylenedioxy-2-hydroxymethyl-3-naphthoic acid lactone, is proposed for the substance, which appears not to have been described before. Formally it is podophyllotoxin or — equally well — picropodophyllin dehydrogenated at 1,2 and 3,4. Since podophyllotoxin is the better known, and since the dehydrogenation of this compound to (I) has been accomplished experimentally (*vide infra*) we propose the name dehydro-podophyllotoxin for the new substance.

The fluorescence and a striking similarity of the absorption spectrum in ultra-violet to that of dehydro-anhydro-podophyllin (I less the hydroxy-group), prepared according to Späth *et al.*⁴ is strong evidence for the fully aromatized naphthalene nucleus. The possibility that ring B contains only two formal double bonds can be ruled out with fair certainty, although it must be admitted that this structure would fit better in with the elementary analysis. In that case the substance would be a 1-hydroxy-derivative of one of the isomeric apo-picropodophyllins. These are, however, non-fluorescent, and their UV-spectra⁵ are fundamentally different from that of our substance. A decisive proof of

the naphthalene nucleus would be provided by demonstrating the failure of further dehydrogenation or by degradation. This is being attempted.

Although the substance does not give coloured coupling products with diazobenzene-sulphonic acid and does not respond to ferric chloride, we nevertheless propose a phenolic structure. The analytical data leave little doubt that the substance contains one oxygen atom more than dehydro-anhydro-picropodophyllin (calc. for C₂₂H₁₆O₇: C 67.0; H 4.6; 3(OCH₃) 23.6). Further evidence for the phenolic hydroxy-group is the location of the substance on the paper chromatograms close to other phenolic podophyllin constituents (the peltatins). The relation of the R_F to the number of hydroxy-groups has been established in previous work³. Dehydro-anhydro-picropodophyllin has a much larger R_F -value. It almost follows the benzene front in accordance with its lack of a hydroxy-group.

The position of the hydroxy-group in ring B is consistent with the failure of the coupling reaction, since this ring is then fully substituted. In ring A, on the other hand, there would be a para-position free for coupling as in the peltatins, which form strongly coloured azo-compounds with diazobenzenesulphonic acid³. The absence of the colour reaction with ferric chloride is unimportant; several substituted phenols fail to react. The presence and the proposed position of the hydroxy-group is finally supported by the fact that podophyllotoxin upon dehydrogenation with lead tetra-acetate produces a spot on the paper-chromatograms which is identical with that of the natural compound (I).

Further details will appear in this journal at a later date. Attempts to isolate the remaining four or five fluorescent substances are in progress.

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