385. *Pectic Substances. Part IV. Citrus Araban.*

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Citrus pectin is shown to contain an araban which can be separated by extraction with 70% alcohol. The methyl derivative of this araban, obtained by treatment of the thallium derivative with methyl iodide, gives rise on hydrolysis to 2 : 3 : 5-trimethyl *l*-arabinose, 2 : 3-dimethyl *l*-arabinose, and 3-methyl *l*-arabinose in equimolecular proportions, and it appears that all three sugars are combined in the polysaccharide in the furanose form. The araban is shown to be identical in the main features of its structure with the araban isolated from the pectic materials of the peanut and of the apple.

Arabans associated with various pectic substances have been discussed in previous papers (Hirst and Jones, J., 1938, 496; this vol., pp. 453, 454), and we now deal with the araban present in citrus pectin. This variety of pectin finds wide industrial application and its constitution is therefore of special interest. The citrus pectin used in this work was a purified commercial sample, which was free from added sugar or acids It contained methyl pectate (*ca.* 78%), araban (*ca.* 7%), galactan, and smaller amounts of other materials, including the glycoside hesperidin. Preferential solution of the araban occurred when the pectin was boiled with 70% alcohol. The solution on concentration deposited some hesperidin, the identity of which was further substantiated by the preparation of its acetyl derivative (King and Robertson, J., **1931**, 1704). Addition of acetone to the concentrated alcoholic solution precipitated crude araban, $[\alpha]_D$ about $-34°$. This material still contained pectic acid and galactan as impurities. After purification by reprecipitation, material having $[\alpha]_D -118°$ was obtained (degree of purity, approximately 70%), but by this simple procedure it was not possible to isolate araban in the pure condition. The impurities can be removed by acetylation, the resulting diacetyl araban having constants identical with those previously recorded for the diacetyl araban isolated from the pectic material of the peanut (*loc. cit.*). Citrus araban underwent hydrolysis with hot 0·01N-sulphuric acid at a rate which indicated the presence of arabofuranose molecules in the polysaccharide. The rate of hydrolysis is the same as that of apple and peanut arabans under similar conditions.

The methylation of citrus araban was effected by heating the thallium derivative with methyl iodide and after purification of the crude product by fractional precipitation, the fully methylated derivative was obtained. The structural similarity between this material and the methylated arabans from the peanut and from the apple was shown by its behaviour on hydrolysis, an equimolecular mixture of 2 : 3 : 5-trimethyl *l*-arabofuranose, 2 : 3-dimethyl *l*-arabinose, and 3-methyl *l*-arabinose being obtained. Identification of the trimethyl and the dimethyl arabinose presented no difficulties, but, as was also the case with the corresponding sugar from methylated peanut and apple arabans, difficulties were encountered in providing a rigid proof of the constitution of the monomethyl arabinose owing to the fact that no crystalline reference compound could be isolated. There is, however, strong evidence pointing to the identification of the sugar as 3-methyl *l*-arabinose.

The structural problem is therefore the same as that discussed in connection with the apple and peanut arabans (Hirst and Jones, *loc. cit.*). It will be noted, however, that the rotation ($[\alpha]_D -128°$) of the methylated araban from citrus pectin is greater than that recorded for the other methylated arabans. Since, as was previously pointed out, the data available do not suffice for an unique solution of the constitutional problem, it might be held that this difference of rotation indicates some structural difference. Nevertheless, we incline to the view that all three arabans are identical in their essential structural features, and that the rotation difference is due to the difficulty of effecting complete separation of araban and pectic acid (see above). Owing to the extremely high positive rotation of methylated pectic acid, a small proportion of this admixed with the methylated araban would have a marked effect on the rotation. It seems probable that the methylated araban now described is the purest sample so far isolated, the conditions for methylation being specially favourable owing to the fact that the crude araban employed contained about 60% of pure pentosan.

The ease of hydrolysis and the nature of the hydrolysis products of the methylated araban indicate that all the arabinose residues in the polysaccharide are furanose in structure and probably of the α-configuration. It would appear, therefore, that the arabans present in peanut, apple and citrus pectins are identical in their fundamental chemical structure. Since it has already been shown that citrus pectic acid (Smith, *Chem. and Ind.*, 1939, **58**, 363), strawberry pectic acid (Beaven and Jones, *ibid.*, p. 363), and apple pectic acid (Beaven, Hirst, and Jones, forthcoming publication) are also identical with respect to the main features of their chemical structure, it seems probable that the materials designated pectins consist essentially of pectic acid, usually in the form of its methyl ester, mixed with araban, galactan, and varying quantities of adventitious material such as hesperidin. It is of particular importance to note that the experiments now described show that citrus araban cannot be derived directly from pectic acid by decarboxylation of the galacturonic acid residues.

EXPERIMENTAL.

Citrus Araban.—Isolation. Commercial citrus pectin (1 kg.) (uronic anhydride, 74·9%; pentosan, 6·9%; galactan, 15·4%; $[\alpha]_D + 183^\circ$; equiv. wt. 268) was stirred with hot dilute alcohol (2 l.) (70%) for 10 hours. The filtered extract, which had a negative rotation, was concentrated under reduced pressure at 40° to a small volume. Some hesperidin which had separated was filtered off and the crude araban was precipitated by the addition of acetone (10 vols.) and dried in a vacuum. Three successive extractions of the pectin gave hesperidin (10 g.), m. p. 250° (acetyl derivative, m. p. 143°, $[\alpha]_D - 34^\circ$), and crude araban (26 g.), $[\alpha]_D^{20^\circ} - 34^\circ$ in water.

The araban was purified by solution in water and precipitation from the clear solution (filtration and centrifuge) by the addition of a large volume of acetone. The precipitate contained both pectic acid and sodium sulphate as impurities. It was dissolved in water, and barium hydroxide added until no further precipitate formed. The solution was spun in the centrifuge, and alcohol (1 vol.) added. A slight gelatinous precipitate was separated (centrifuge) and discarded. Addition of a large excess of alcohol then gave crude citrus araban (18 g.) as a pale brown solid, $[\alpha]_D^{20^\circ} - 118^\circ$ in water [Found: furfural, 39·8 (from the yield of phloroglucide obtained on boiling with 12% hydrochloric acid under the standard conditions); ash, 6·4; uronic anhydride, 5% (from the yield of carbon dioxide on boiling with 12% hydrochloric acid)]. The crude araban on hydrolysis with N/100-sulphuric acid at 90° for 25 hours (final rotation + 75°) gave *l*-arabinose, recognised as the diphenylhydrazone, m. p. 195°, in 70% yield. From this figure, the rotation, and the yield of furfural, it appears that the crude material contained approximately 70% of pure araban.

The impurities were removed by acetylation when the crude araban (8·0 g.) on treatment with pyridine (80 c.c.) and acetic anhydride (40 c.c.) at 80° for 10 hours gave diacetyl araban (4·2 g.), $[\alpha]_D^{20^\circ} - 94^\circ$ in acetone (for rotation, compare Hirst and Jones, this vol., p. 453) (Found: $CH_3\cdot CO$, 40·5; furfural on distillation with 12% hydrochloric acid, 29·2. Calc. for $C_9H_{12}O_6$: $CH_3\cdot CO$, 39·9; furfural, 32·0%).

Methylation of Araban.—Another sample of crude citrus araban (10 g.), having $[\alpha]_D - 80^\circ$ in water, was dissolved in water (100 c.c.), and thallium hydroxide (2·5 equivs.) added. The solution was evaporated to dryness in a vacuum at 50° with the exclusion of carbon dioxide. The residual greyish-green solid was powdered (120 mesh), and acid-free, anhydrous methyl iodide added. After the initial vigorous reaction had subsided, the mixture was heated at 45° for a further 32 hours with the exclusion of light and moisture. The excess of methyl iodide was boiled off, and the solid residue extracted exhaustively with methyl alcohol. The extracts on concentration gave a viscid brown solid (11·2 g.), of which 6·5 g. were dissolved in benzene and the solution evaporated to dryness with the addition of thallous ethoxide (2·5 equivs.). The product was powdered (120 mesh) and boiled with methyl iodide as before and the methylated araban was isolated in the usual manner. Two further methylations with Purdie's reagents, followed by one further methylation with thallous ethoxide, gave a crude methylated araban (5·9 g.) (Found: OMe 36·5%).

The solid product was dissolved in ether and the solution was spun in the centrifuge to remove traces of thallous iodide. Two fractions were obtained by the additions of light petroleum: (1) a pale brown, crisp solid (3·9 g.), $[\alpha]_D^{20^\circ} - 128^\circ$ in methyl alcohol (Found: OMe, 38·8. Calc. for dimethyl pentosan: OMe, 38·8%), and (2) a syrup (1·1 g.), which was not further examined.

Methylated araban (3·80 g.), $[\alpha]_D^{20^\circ} - 128^\circ$ in methyl alcohol, was boiled with 2% methyl-alcoholic hydrogen chloride for 22 hours. The solution was neutralised with silver carbonate, filtered, and concentrated at ordinary pressure to a syrup (4·30 g., $n_D^{20^\circ}$ 1·4580), which was fractionally distilled, giving (A) 1·34 g., b. p. 85°/0·001 mm. (bath temp.), $n_D^{20^\circ}$ 1·4354 (Found: OMe, 58·5%); (B) 1·15 g., b. p. 110—128°/0·001 m. (bath temp.), $n_D^{20^\circ}$ 1·4530 (Found: OMe, 48·0%); (C) 0·82 g., b. p. 170—200°/0·001 mm. (bath temp.), $n_D^{20^\circ}$ 1·4730 (Found: OMe, 35·2%); (D) 0·12 g., b. p. 200°/0·001 mm. (bath temp.) (Found: OMe, 28·0%); still residue, 0·36 g. Estimations based on n_D, OMe and $[\alpha]_D$ values of the above fractions indicated that their compositions were approximately as follows:

Fraction.	Trimethyl methyl-arabofuranoside, g.	Dimethyl methyl-arabinoside, g.	Monomethyl methyl-arabinoside, g.
A	1·17	0·17	—
B	0·13	1·02	—
C	—	0·09	0·73
D	—	—	0·08
	1·30	1·28	0·81

These yields correspond respectively to 84% of the calculated amount of trimethyl sugar, 90% of the calculated amount of dimethyl sugar, and 61% of the calculated amount of monomethyl sugar, estimated on the assumption that the methylated araban gives on hydrolysis equimolecular proportions of tri-, di-, and mono-methyl arabinose. Since there is always some loss of trimethyl and monomethyl arabinose during distillation (Hirst and Jones, *loc. cit.*), these figures are held to be in agreement with the view that this sample of methylated araban, like methylated araban from peanuts and the apple (Hirst and Jones, *loc. cit.*), does give these three methylated arabinoses in equimolecular proportions. The identity of the three products of hydrolysis was obtained in the following ways.

2 : 3 : 5-*Trimethyl Arabinose*.—Fraction (A) (1·32 g.) was hydrolysed by 0·5N-hydrochloric acid (50 c.c.) at 90—95°; $[\alpha]_D^{21^\circ}$ − 68° (initial value, *c*, 2·64) changing after 2 hours to − 27° (constant value). The mixed free sugars on isolation in the usual way (1·14 g.) had $n_D^{19\cdot5^\circ}$ 1·4512, $[\alpha]_D^{21^\circ}$ − 29° (*c*, 2·6 in water) (OMe, 46%). The mixed sugars (1·10 g.) were fractionally distilled, giving a distillate (0·96 g.), b. p. (bath temp.) 116°/0·001 mm., $n_D^{21^\circ}$ 1·4500, $[\alpha]_D^{20^\circ}$ − 40° (*c*, 1·6 in water) (Found : OMe, 48%. Calc. for $C_8H_{16}O_5$: OMe, 48·5%). On oxidation with bromine water the sugar (0·92 g.) gave crystalline 2 : 3 : 5-trimethyl γ-arabonolactone (0·84 g.), b. p. 105°/0·001 mm. (bath temp.), $n_D^{28^\circ}$ 1·4415, $n_D^{21^\circ}$ 1·4453 (superfused solid), m. p. 28° (not depressed on admixture with an authentic specimen), $[\alpha]_D^{20^\circ}$ − 47° in water (*c*, 1·0) (initial value). On treatment with liquid ammonia the lactone gave quantitatively 2 : 3 : 5-trimethyl *l*-arabonamide, m. p. 139° alone or on admixture with an authentic specimen.

The syrup (0·18 g.) remaining after removal of trimethyl *l*-arabinose was almost entirely dimethyl arabinose and was mixed with the syrup (1·06 g.), $n_D^{19^\circ}$ 1·4708, $[\alpha]_D^{20^\circ}$ + 95° (in water, *c*, 2·1) (Found : OMe, 36·0. Calc. for $C_7H_{14}O_5$: OMe, 34·8%), obtained by the hydrolysis of fraction (B) (1·13 g.) (see above) by 0·5N-hydrochloric acid (50 c.c.) at 90—95° for 4 hours [initial rotation of fraction (B) before hydrolysis, $[\alpha]_D^{20^\circ}$ + 90°]. A portion (0·72 g.) of the combined syrups (1·24 g.) was oxidised with bromine water at 60° for 6 hours. The product, isolated in the usual way, gave on distillation 2 : 3-dimethyl γ-arabonolactone (0·65 g.), b. p. (bath temp.) 145°/0·005 mm., $n_D^{25^\circ}$ 1·4595, $n_D^{28^\circ}$ 1·4580, $[\alpha]_D^{23^\circ}$ − 33° in water (*c*, 2·4, initial value); − 31° (90 hrs., mutarotation still not completed) (Found : OMe, 34·0%; equiv., 170. Calc. for $C_7H_{12}O_5$: OMe, 35·2%; equiv., 176). With liquid ammonia the lactone gave 2 : 3-dimethyl *l*-arabonamide in quantitative yield, m. p. 160° alone or on admixture with an authentic specimen (Hirst and Jones, *loc. cit.*).

Fractions C and D were combined (0·92 g.) and hydrolysed with 0·5N-hydrochloric acid (50 c.c.) at 95°; $[\alpha]_D^{20^\circ}$ + 48° (initial value), changing to + 90° in 2·5 hours (constant value). After neutralisation with silver carbonate, the solution was evaporated to dryness, giving a syrup (0·85 g.), $n_D^{20^\circ}$ 1·4850, $[\alpha]_D^{21^\circ}$ + 99° in water (*c*, 1·4) (Found : OMe, 19·0. Calc. for $C_6H_{12}O_5$: OMe, 18·9%). This syrup on standing deposited *l*-arabinose (50 mg.), m. p. and mixed m. p. 160° (Found : C, 40·2; H, 6·7. Calc. for $C_5H_{10}O_5$: C, 40·0; H, 6·7%). The syrup (0·75 g.) was oxidised with bromine water at 60° for 12 hours. Bromine was removed by aeration, and the liquid filtered (X), leaving a small crystalline deposit, m. p. 71° (recrystallised from light petroleum). This was insoluble in water, but soluble in alcohol, acetone, ether and benzene. It gave no colour with ferric chloride and was neutral to litmus. When heated with alcoholic silver nitrate, it gave a precipitate of silver bromide. The high bromine content (Br, 88%) indicates that this material is a decomposition product.

The filtrate (X) was neutralised with silver carbonate, filtered before and after precipitation of silver as sulphide, and evaporated to a syrup (0·60 g.); $[\alpha]_D^{20^\circ}$ − 35° (*c*, 0·82 in water; initial value); − 34° (26 hrs.); − 30° (90 hrs., mutarotation still incomplete) (Found : OMe, 20·0; *M*, by titration with N/100-sodium hydroxide, 168. Calc. for $C_6H_{10}O_5$: OMe, 19·1%; *M*, 162).

With liquid ammonia the lactone gave the corresponding amide as a glassy solid in quantitative yield, $[\alpha]_D^{20^\circ}$ + 27° in water (*c*, 0·77) (Found : OMe, 16·8. Calc. for $C_6H_{13}O_5N$: OMe, 17·3%). This amide, after treatment with sodium hypochlorite under Weerman's conditions (*loc. cit.*) and subsequent addition of semicarbazide to the solution, gave hydrazodicarbonamide, m. p. and mixed m. p. with an authentic specimen, 256°, in a yield of 40%.

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