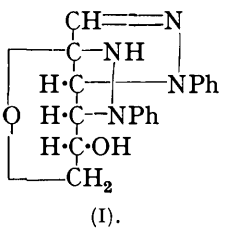
133. *Experiments on the Formation of Mixed Osazones and their Anhydrides.*

By E. E. Percival and E. G. V. Percival.

The preparation of *galactosephenylmethyl-phenylosazone* and of its *anhydride* are described and a structure is proposed for the latter. It is shown that the two isomeric glucosephenylmethyl-phenylosazones of Votoček and Vondráček give rise to the same *anhydride*, for which a structure is proposed embodying a 2 : 6-oxide ring and a 1 : 3-pyrazoline ring. Possible explanations of the structural differences between two isomeric fructosephenylmethylphenylosazones are considered.

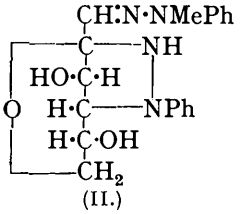
It was shown (J., 1936, 1770) that *d*-glucosazone tetra-acetate and *d*-galactosazone tetra-acetate yielded a dianhydrohexosazone on deacetylation and the proposed structure (I)

was shown later (J., 1938, 1384) to agree with the observation that *d*-gulosazone gave the same product, and that, since a primary alcohol residue was absent, the 2 : 6-oxide ring structure previously postulated for glucosazone (J., 1935, 1398) and later for galactosazone (J., 1940, 1479) was retained in this anhydride. The stereochemical structure of (I) was, however, undecided, although the isolation of the same anhydride from the three osazones (and of its enantiomorph from *l*-sorbosazone) made it clear that in appropriate circumstances inversion could take place at both C_3 and C_4 and probably also at C_2. By replacing each of the phenylhydrazine residues in turn by phenylmethylhydrazine, which cannot take part in anhydride formation, it was hoped to secure information from rotational data as to where inversion occurred, to determine also whether the 2 : 6-oxide ring would be retained when C_2 carried a phenylmethylhydrazine residue [since Wolfrom and Christman (*J. Amer. Chem. Soc.*, 1931, **53**, 3413) have demonstrated that galactosephenylmethylhydrazone is acyclic and their evidence is supported by the low specific rotation of this compound], and to throw light on the structures of the two isomeric fructosephenylmethylhydrazones (J., 1940, 1511).

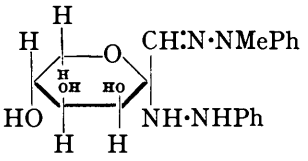


(I).

Galactosephenylmethylhydrazone and phenylhydrazine readily yielded *galactose-phenylmethyl-phenylosazone*, m. p. 178°, $[\alpha]_D^{17°}$ + 98°, the crystalline *tetra-acetate* of which, m. p. 183°, $[\alpha]_D^{13°}$ + 85°, yielded on deacetylation a *monoanhydrogalactosephenylmethyl-phenylosazone*, m. p. 172°, $[\alpha]_D^{13°}$ + 100°, which yielded a crystalline *diacetate*. The *di-p-toluenesulphonate* did not react with sodium iodide in acetone at 100° and it was therefore concluded that a primary alcohol residue was absent (Oldham and Rutherford, *J. Amer. Chem. Soc.*, 1932, **54**, 366). The anhydride is therefore considered to be (II). From the similarity in the specific rotations of galactosephenylmethyl-phenylosazone and its anhydro-derivative it would seem likely that the configuration on C_2 is not changed on anhydride formation, although if this is α (III) inversion is necessary on C_4 and the anhydride would be related to fructopyranose.



(II.)



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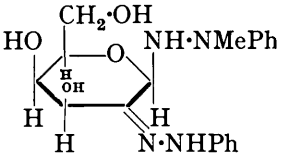


(IV.)

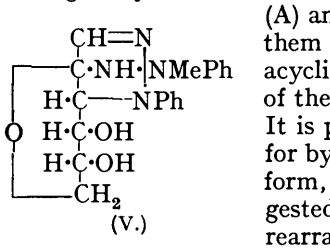
If the galactosephenylmethyl-phenylosazone possessed structure (IV) containing a 1 : 5-oxide ring, great strain would be imposed on anhydride formation owing to the rigidity conferred by the presence of the double bond, and this is a further argument in favour of (III).

Attempts to prepare galactosephenyl-phenylmethylosazone failed. Treatment of galactosephenylhydrazone with phenylmethylhydrazine invariably led to the production of galactosephenylmethylhydrazone (70%) together with the same osazone (12%) as described above, which yielded the same anhydride.

It was also found impossible in the glucose series to prepare one of the desired mixed osazones. Votoček and Vondráček (*Ber.*, 1904, **37**, 3848) claimed the isolation of glucose-phenylmethyl-phenylosazone (B), m. p. 205°, from glucosephenylmethylhydrazone and phenylhydrazine and of glucosephenyl-phenylmethylosazone (A), m. p. 192°, together with (B) from glucosephenylhydrazone (and fructosephenylhydrazone) and phenylmethyl-hydrazine. These facts were verified, but we disagree with Votoček's structure for (B). Glucosephenylhydrazone and phenylmethylhydrazine gave a product (A), m. p. 194°, $[\alpha]_D^{17°}$ − 53° ⟶ − 6°, and (B), m. p. 203°, $[\alpha]_D^{17°}$ − 60° ⟶ − 15°. Glucosephenylmethyl-hydrazone and phenylhydrazine gave an osazone identical with (B) and Votoček's observation that more vigorous treatment gave glucosazone was confirmed. When fructose-

phenylmethylhydrazone prepared according to Ofner (*Monatsh.*, 1905, **26**, 1165) was treated with phenylhydrazine, a mixed osazone identical with (B) was obtained, whereas the fructosephenylmethylhydrazone, m. p. 170° (J., 1937, 320) gave rise exclusively to (A). From this result it would appear that both (A) and (B) carry the phenylmethylhydrazine residue on C_2, although the formation of (B) from glucosephenylmethylhydrazone appears to indicate that it is on C_1 in this case. In one case, therefore, a phenylmethylhydrazine residue has been displaced and transferred to another carbon atom. That displacement can occur is evident, since glucosazone can be isolated when glucosephenylmethylhydrazone is heated with phenylhydrazine and in the galactose series the same phenomenon was noted. The weight of evidence suggests that the transference occurs in the latter reaction, since the osazone took seven minutes to appear in the case of glucosephenylmethylhydrazone, whereas when either of the fructosephenylmethylhydrazones was employed only one minute was required under identical conditions. The conclusion reached, therefore, is that both (A) and (B) are glucosephenyl-phenylmethylosazones. That both have essentially the same structure is shown by the fact that they gave amorphous acetates of similar properties, $[\alpha]_D^{15°}$ — 43°, which on deacetylation yielded the same *monoanhydroglucosephenyl-phenylmethylosazone*, m. p. 176—178°, $[\alpha]_D^{13°}$ — 158°, which yielded a crystalline *diacetate*, m. p. 158°, $[\alpha]_D^{15°}$ — 151°, and a *ditosyl* derivative which suffered no reaction on heating with sodium iodide in acetone. The structure (V) is therefore assigned to the anhydride and this was confirmed by the isolation of a crystalline *monoacetone* derivative.

If (A) and (B) possess the β-configuration (VI), then inversion has taken place on C_3 during anhydride formation and (V) is an allose derivative. The specific rotations of (A) and (B) would seem to be too similar for the difference between them to be accounted for by a cyclic structure in one case and an acyclic one in the other, and the fact that both yield an anhydride of the above structure suggests that both are fructopyranosazones. It is possible that the differences between them may be accounted for by the fact that one is a *syn*- (VII) and the other an *anti*- (VIII) form, or that one is the isomeric azo-form, $-CH_2-N{:}NPh$, as suggested by Zerner and Waltuch (*Monatsh.*, 1914, **35**, 1025), although rearrangement would be necessary in this case to supply the hydrogen atom for anhydride formation. In the case of (VII) anhydride formation on C_3 is also impossible and it would be necessary to suppose that during the acetylation which precedes anhydride formation the *syn*-form is converted into the *anti*-form; this is supported by the observation that the acetates formed from (A) and (B) are indistinguish-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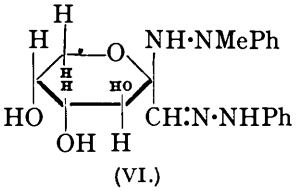
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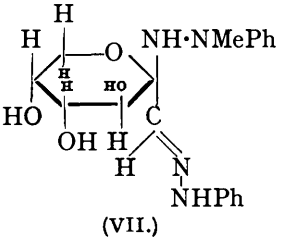
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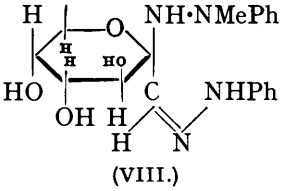
(VIII.)

able. The experimental results, however, do not enable us to decide between any of these hypothetical structures.

As suggested previously (J., 1940, 1511), it is probable that the fructosephenylmethylhydrazones differ because one is cyclic ($[\alpha]_D^{17°}$ — 253°) and the other is acyclic. From the above results osazone (A) is derived from the cyclic and (B) from the acyclic form, although a pyranose ring then appears in (B). Why different osazones finally result is not clear, although it may be permitted to speculate that in the acyclic case the entering phenylhydrazine residue is forced into the *syn*-form by the repulsive effect of the $>C{:}N{\cdot}NMePh$, which is rigidly attached by the double bond in a plane perpendicular to that containing the carbon atoms. If the production of an azo-form is concerned, a similar explanation

could be advanced, for the transference of the double bond would also relieve congestion in the same way.

Experimental.

Galactosephenylmethyl-phenylosazone and its Tetra-acetate (With Miss M. C. Macraild).—Galactosephenylmethylhydrazone (5 g.), m. p. 182°, $[\alpha]_D^{18°}$ + 5° in pyridine–alcohol (3 : 2; *c*, 0·4), in alcohol (150 c.c.) was treated with phenylhydrazine (5 g.) and acetic acid (3 c.c.) at 100° for 20 hours. The *product* (6 g.) was isolated by the addition of water and cooling and had m. p. 178°, $[\alpha]_D^{17°}$ + 98° in pyridine–alcohol (1 : 1; *c*, 0·4), + 71° (21 hours); + 62° (45 hours); + 45° (100 hours, constant) (Found: C, 60·8; H, 6·6; N, 15·2. $C_{19}H_{24}O_4N_4$ requires C, 61·3; H, 6·5; N, 15·05%).

The osazone (5 g.) was dissolved in a mixture of acetic anhydride (10 c.c.) and pyridine (15 c.c.), and the *product* poured into water after 2 days and recrystallised from alcohol; it had m. p. 183°, $[\alpha]_D^{13°}$ + 85° in chloroform (*c*, 0·4) (Found: C, 60·1; H, 6·0; N, 10·5; CH_3·CO, 32·8. $C_{27}H_{32}O_6N_4$ requires C, 60·0; H, 6·0; N, 10·4; CH_3·CO, 31·8%).

*Anhydrogalactosephenylmethyl-phenylosazone, its Diacetate and Di-*p*-toluenesulphonate.*—The tetra-acetate (6·5 g.) in acetone (500 c.c.) was treated with 8% sodium hydroxide solution (80 c.c.) and water (300 c.c.). After 1 day the *product* (3 g.) was collected and recrystallised from acetone–light petroleum; it had m. p. 172°, $[\alpha]_D^{13°}$ + 100° in acetone (*c*, 0·4) (Found: C, 65·0; H, 6·2; N, 15·4. $C_{19}H_{22}O_3N_4$ requires C, 64·4; H, 6·3; N, 15·8%).

The *diacetate* was obtained on acetylation as above and recrystallised from alcohol, forming pale yellow needles, m. p. 170°, $[\alpha]_D^{14°}$ + 50° in chloroform (*c*, 0·4) (Found: C, 62·3; H, 5·8; N, 13·1; CH_3·CO, 19·6. $C_{23}H_{26}O_5N_4$ requires C, 63·0; H, 6·1; N, 12·8; CH_3·CO, 19·6%).

The anhydride (0·5 g.) was treated with *p*-toluenesulphonyl chloride (1 g.) in pyridine (4 c.c.) for 2 days. On pouring into water, a yellow powder was obtained, which separated from aqueous alcohol as a yellow microcrystalline powder, m. p. 65—70° (decomp.), $[\alpha]_D^{15°}$ + 37° in chloroform (*c*, 0·5) (Found: C, 60·5; H, 5·3; N, 8·55; S, 8·9. $C_{33}H_{34}O_7N_4S_2$ requires C, 59·8; H, 5·2; N, 8·5; S, 9·7%).

Ditosylanhydrogalactosephenylmethyl-phenylosazone (1 g.) was heated for 20 hours at 100° with sodium iodide (1·5 g.) in acetone (7·5 c.c.). Treatment with water yielded a brown solid (0·4 g.) devoid of iodine but containing nitrogen and sulphur. When this treatment was repeated for 7 hours on another specimen, the product was again devoid of iodine and appeared to be an impure monotosyl ester (Found: N, 11·4. $C_{26}H_{28}O_5N_4S$ requires N, 11·2%).

In attempts to condense the anhydride with acetone as described below in the glucose series by shaking with anhydrous copper sulphate the anhydride was recovered unchanged.

Galactosephenylhydrazone and Phenylmethylhydrazine.—Galactosephenylhydrazone (10 g.) was heated for 2·5 hours with phenylmethylhydrazine (10 g.) and acetic acid (5·5 c.c.) in alcohol (600 c.c.). Galactosephenylmethylhydrazone (7 g.) rapidly formed. In another experiment the heating was continued for 8 hours to yield galactosephenylmethylhydrazone (7·5 g.) and an osazone (1·2 g.), m. p. 175°, $[\alpha]_D^{15°}$ + 96° in pyridine–alcohol (1 : 1; *c*, 0·45), falling to + 65° in 47 hours. This proved to be the same osazone as described above, since it yielded an acetate, m. p. 180°, $[\alpha]_D^{15°}$ + 86° in chloroform (*c*, 0·5), which on deacetylation yielded an anhydride, m. p. 171—172°, unchanged on admixture with the anhydride described above, $[\alpha]_{D_l}^{15°}$ + 98° in acetone (*c*, 0·3).

Glucosephenylhydrazone and Phenylmethylhydrazine.—Glucosephenylhydrazone (12 g.), phenylmethylhydrazine (10 g.), acetic acid (5 c.c.), water (450 c.c.), and a little sodium bisulphite were heated at 95—100°. After 45 minutes a crop of crystals (1) (3·5 g.), m. p. 193—195°, was removed, and after a further hour crop (2) (1·2 g.), m. p. 180°, was isolated. Three further crops (2·9 g.) similar to (2) were obtained. On extraction of (1) with hot alcohol a solution was obtained from which an osazone (A) crystallised on cooling (2·2 g.), m. p. 184°, $[\alpha]_D^{15°}$ — 53° in pyridine–alcohol (1 : 1; *c*, 0·5); — 19° (19 hours); — 6° (68 hours, constant). The residue (B) (1·3 g.) had m. p. 202—203°, $[\alpha]_D^{15°}$ — 60° in pyridine–alcohol (1 : 1; *c*, 0·4), — 20° (24 hours); — 15° (40 hours, constant). Mixed m. p. of (A) and (B), 183° [Found: (A) C, 60·7; H, 6·5; N, 15·4. (B) C, 60·8; H, 6·5; N, 15·3. Calc. for $C_{19}H_{24}O_4N_4$: C, 61·3; H, 6·5; N, 15·05%]. Fraction (2) on recrystallisation gave an osazone, m. p. 192—194°, identical with (A), as did also the remaining fractions.

Glucosephenylmethylhydrazone and Phenylhydrazine.—Glucosephenylmethylhydrazone (18 g.), m. p. 132°, $[\alpha]_D^{19°}$ + 5° in water (*c*, 1·6), was heated with water (250 c.c.), phenylhydrazine (13·5 g.), acetic acid (7·5 c.c.), and sodium bisulphite. Five fractions of osazone (11 g.) were separated, the first after heating for 30 minutes. This had m. p. 200°, raised to 202° [unchanged

on admixture with (B)] on recrystallisation, $[\alpha]_D^{15°}$ — 62° in pyridine–alcohol (1 : 1; *c*, 0·5), — 30° (12 hours), — 14° (43 hours, constant). All the other fractions had similar properties.

In a second experiment half the above proportion of phenylhydrazine was used and six fractions were isolated. Fraction (1) had m. p. 201—202°, $[\alpha]_D^{15°}$ — 62° in pyridine–alcohol (1 : 1; *c*, 0·4), — 22° (21 hours), — 16° (45 hours, constant). The other five fractions were similar and all gave the same anhydride on acetylation and subsequent deacetylation.

In a third experiment glucosephenylmethylhydrazone (20 g.) in alcohol (500 c.c.) was heated for 8 hours with phenylhydrazine (25 g.) and acetic acid (13 c.c.) to yield an osazone (14 g.), m. p. 205°, $[\alpha]_D^{15°}$ — 70° in pyridine–alcohol (1 : 1; *c*, 0·5), — 29° (17 hours, constant). Acetylation and deacetylation gave the dianhydrohexosazone, m. p. 235°, previously described (*loc. cit.*), confirmed by the isolation of the monoacetate, m. p. 135°, and comparison with authentic specimens.

Glucosephenyl-phenylmethylosazone Anhydride.—Osazones (A) and (B) were acetylated in the usual way to yield amorphous acetates. *Acetate* (*A*) had $[\alpha]_D^{13°}$ — 44° in chloroform (*c*, 0·4) (Found : C, 59·8; H, 5·7; N, 10·7; CH_3·CO, 32·7. $C_{27}H_{32}O_8N_4$ requires C, 60·0; H, 6·0; N, 10·4; CH_3·CO, 31·8%), and *acetate* (*B*), $[\alpha]_D^{15°}$ — 43° in chloroform (*c*, 0·5) (Found : N, 10·6%). Acetate (A) (1·5 g.) was deacetylated as described for the galactose derivative to yield a crystalline *anhydride* (0·7 g.), obtained from acetone–light petroleum in lemon-yellow needles, m. p. 176—178°, $[\alpha]_D^{13°}$ — 158° in acetone (*c*, 0·4) (Found : C, 64·3; H, 6·3; N, 15·9. $C_{19}H_{22}O_3N_4$ requires C, 64·4; H, 6·3; N, 15·8%). Acetate (B) on similar treatment gave the same glucosephenyl-phenylmethylosazone anhydride, m. p. 176°, $[\alpha]_D^{15°}$ — 155° in acetone (*c*, 0·5); a mixed m. p. with the anhydride from (A) showed no depression.

Anhydroglucosephenyl-phenylmethylosazone Diacetate.—By acetylation as before a *product* was obtained in quantitative yield which, after recrystallisation from alcohol, had m. p. 158°, $[\alpha]_D^{15°}$ — 151° in chloroform (*c*, 0·5), and gave the original anhydride, m. p. 177°, on deacetylation (Found : C, 62·9; H, 6·0; N, 12·4; CH_3·CO, 19·7. $C_{23}H_{26}O_5N_4$ requires C, 63·0; H, 6·1; N, 12·8; CH_3·CO, 19·6%).

Acetone Anhydroglucosephenyl-phenylmethylosazone.—The anhydride (0·2 g.) was shaken with acetone (100 c.c.) and anhydrous copper sulphate (20 g.) for 3 days. After filtration and evaporation a *product* was obtained (0·25 g.) which on recrystallisation from acetone–light petroleum had m. p. 160°, $[\alpha]_D^{18°}$ — 33° in acetone (*c*, 0·5) (Found : C, 67·1; H, 6·7; N, 14·5. $C_{22}H_{26}O_3N_4$ requires C, 67·0; H, 6·6; N, 14·2%).

Ditosyl Anhydroglucosephenyl-phenylmethylosazone.—This *compound*, prepared as in the galactose series and isolated as a yellow powder, had m. p. 65—70° (decomp.), $[\alpha]_D^{15°}$ — 80° in chloroform (*c*, 0·4) (Found : C, 60·0; H, 5·2; N, 8·6. $C_{33}H_{34}O_7N_4S_2$ requires C, 59·8; H, 5·2; N, 8·5%). This product was treated with sodium iodide in acetone at 100° for 14 hours; the resultant brownish-yellow material contained nitrogen and sulphur but was devoid of iodine.

Fructosephenylmethylhydrazones and Phenylhydrazine.—(1) *Ofner's phenylmethylhydrazone, m. p.* 116°. This substance, $[\alpha]_D^{15°}$ — 7° in pyridine–alcohol (1 : 1; *c*, 0·9) (6·1 g.) in water (84 c.c.) was heated at 95—100° with phenylhydrazine (4·5 g.), acetic acid (2·4 c.c.), and sodium bisulphite; crystals appeared after 1 minute (in 0·6% solution no osazone appeared after 45 minutes until the solution was cooled). Five crops of osazone (5·6 g.) were obtained with m. p.'s varying between 202—203° and 197°, and on recrystallisation all yielded the same osazone, m. p. 203—204°, $[\alpha]_D^{18°}$ — 61° in pyridine–alcohol (1 : 1; *c*, 0·5), — 20° (18 hours), — 14° (43 hours, constant). Admixture with osazone (B) from glucose-phenyl- or -phenylmethyl-hydrazone did not depress the m. p. (Found : C, 60·9; H, 6·4; N, 14·9. Calc. for $C_{19}H_{24}O_4N_4$: C, 61·3; H, 6·5; N, 15·05%).

The first four fractions were acetylated to yield identical acetates, $[\alpha]_D^{18°}$ — 43° in chloroform (*c*, 0·5) (Found : C, 59·9; H, 6·0; N, 10·6; CH_3·CO, 32·9. Calc. for $C_{27}H_{32}O_8N_4$: C, 60·0; H, 6·0; N, 10·4; CH_3·CO, 31·8%), which were deacetylated as before. In every case a product was obtained in good yield which, on recrystallisation from acetone–light petroleum yielded the anhydride described above, m. p. 176°, $[\alpha]_D^{16°}$ — 155° in acetone (*c*, 0·5), confirmed by determinations of mixed m. p. This result was twice confirmed.

(2) *Fructosephenylmethylhydrazone, m. p.* 170°. Osazone formation in the usual way yielded pale yellow needles, m. p. 194—195°, not depressed by osazone (A) from glucosephenylhydrazone, $[\alpha]_D^{15°}$ — 54° in pyridine–alcohol (1 : 1; *c*, 0·5), — 23° (18 hours), — 7° (44 hours, constant). The osazone was formed in less than 1 minute from a 0·6% solution of the hydrazone (Found : C, 60·7; H, 6·5; N, 15·3. Calc. for $C_{19}H_{24}O_4N_4$: C, 61·3; H, 6·5; N, 15·05%). Acetylation yielded quantitatively a tetra-acetate, $[\alpha]_D^{13°}$ — 44° in chloroform (*c*, 0·4) (Found : C, 59·8; H, 5·9; N, 10·7; CH_3·CO, 33·0. Calc. for $C_{27}H_{32}O_8N_4$: C, 60·0; H, 6·0; N, 10·4;

CH_3·CO, 31·8%). Deacetylation of this acetate (2·2 g.) in acetone (150 c.c.) with 0·5N-sodium hydroxide (112 c.c.) yielded the anhydride previously described (1·1 g.), m. p. 176—177°, $[\alpha]_D^{15°}$ − 158° in acetone (*c*, 0·4) (Found : C, 64·3; H, 6·3; N, 15·5. Calc. for $C_{19}H_{22}O_3N_4$: C, 64·4; H, 6·3; N, 15·8%). Acetylation yielded the diacetate, m. p. 157—158°, $[\alpha]_D^{15°}$ − 151° in chloroform (*c*, 0·6) (Found : CH_3·CO, 19·6. Calc. for $C_{23}H_{26}O_5N_4$: CH_3·CO, 19·6%).

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