

PAPER CHROMATOGRAPHY OF THE OSAZONES OF SOME SUGARS

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In the study and identification of sugars and some of their derivatives, various methods of chromatography are widely used [1-4]. However, there is little information on the chromatography of the osazones [5, 6].

The present paper gives the results relating to the chromatographic behavior on paper in a number of solvent systems of the phenylosazones and p-nitrophenylosazones of the sugars most frequently found in nature.

We have established that the phenylosazones and p-nitrophenylosazones of the methylpentoses and pentoses can be chromatographed successfully in the chloroform-formamide system (Fig. 1a). The osazones of the hexoses are well separated on chromatography in the same system under elution conditions (6 h) or in the chloroform-tetrahydrofuran-formamide (50 : 50 : 6.5) system (Fig. 1b). The benzene-methyl ethyl ketone (1 : 1)-water (3.5%) system is particularly successful both for hexoses and for osazones of bioses. By increasing the proportion of benzene it is possible to chromatograph the osazones of the methylpentoses and pentoses (Fig. 1c, d). An increase in the concentration of methyl ethyl ketone improves the separation of the phenylosazones and p-nitrophenylosazones of the disaccharides robinobiose, rutinose, maltose, and lactose. The R_f values of the phenylosazones of the sugars are higher than those of the p-nitrophenylosazones. The phenylosazones show up in daylight in the form of light yellow spots and in UV

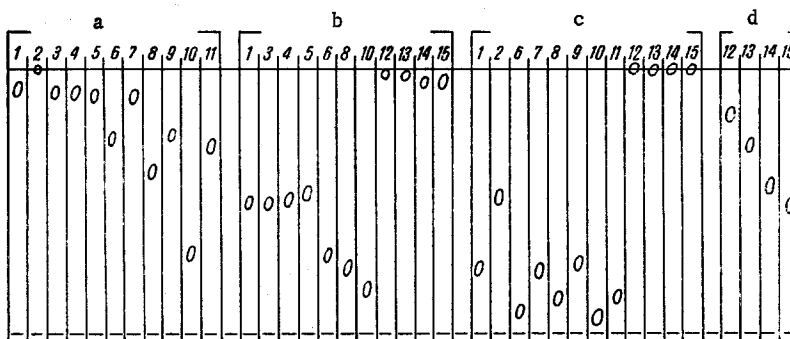


Fig. 1. Chromatograms of the osazones of some sugars in the solvent systems chloroform-formamide (a); chloroform-tetrahydrofuran-formamide (50 : 50 : 6.5) (b); methyl ethyl ketone-benzene (1 : 2)-water (3.5%) (c); and methyl ethyl ketone-benzene (1 : 1)-water (3.5%) (d). Phenylosazones of: 1) D-glucose; 3) D-fructose; 4) D-mannose; 5) D-galactose; 6) L-arabinose; 8) D-xylose; 10) L-rhamnose; 12) Lactose; 13) Maltose; 14) Rutinose; 15) Robinobiose. Nitrophenylosazones of: 2) D-glucose; 7) L-arabinose; 9) D-xylose; 11) L-rhamnose.

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light in the form of bluish yellow spots. When the chromatograms are sprayed with alkali their color becomes stronger. The p-nitrophenylosazones appear in daylight and UV light in the form of orange spots. On treatment with alkali the spots of the p-nitrophenylosazones become bright blue.

The phenylosazones and p-nitrophenylosazones of the sugars studied were obtained by published methods [7-9].

For chromatography, 0.1% solutions of the osazones in a mixture of ethanol and dimethylformamide were prepared and were then deposited (in amounts of 0.01-0.02 ml) on the prepared paper. For the solvent systems tetrahydrofuran-chloroform-formamide (50:50:6.5) and chloroform-formamide, the paper was impregnated with 25% formamide in acetone, and for the benzene-methyl ethyl ketone (various ratios) systems it was impregnated with a mixture of water and acetone (1:1). The Goznak paper prepared for chromatography must contain 35% of the aqueous phase. After the end of the chromatographic process, the chromatograms were dried at room temperature and inspected in daylight and filtered UV light and after being sprayed with a methanolic solution of alkali (6 g of KOH-25 ml of water-45 ml of methanol). Thus, we have performed a qualitative chromatographic analysis of the phenylosazones of the monosaccharides D-xylose, L-arabinose, L-rhamnose, D-glucose, D-mannose, D-fructose, and D-galactose and of the disaccharides maltose, lactose, rutinose, and robinobiose, and of the p-nitrophenylosazones of D-xylose, L-arabinose, L-rhamnose, and D-glucose.

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