Selective detection of various types of sugars present in cardiac heterosides

About 20 simple sugars are found in cardiac heterosides; these include 2,6-deoxyaldohexoses, 2-deoxyaldohexoses, 6-deoxyaldohexoses, aldohexose (glucose) and aldopentose (xylose[1,2]). Their discrimination in paper chromatography by comparison of R_F values is often unconvincing, as many sugars have similar R_F values even in various solvent systems. In such cases identification is furthered by using reagents which yield various colors with the different types of sugars.

In our work the best results were obtained by the following technique:

The dried chromatograms were sprayed with a saturated solution of 2-thiobarbituric acid in glacial acetic acid, dried immediately at 120–130° and then kept over boiling water for some minutes[3]. The spot of xylose became olive, that of glucose orange-yellow, and that of the 6-deoxyhexoses yellow. Pink spots due to 2-deoxyhexoses and 2,6-deoxyhexoses were faintly visible under these conditions. The chromatograms were then immersed in MACLENNAN *et al.*'s[4] reagent (equal parts of 1% vanillin in ethanol and of a 3% aqueous solution of 70% perchloric acid) and heated in a stream of hot air (from a hair-drier). The paper turned yellow, the spots of xylose and glucose darkened markedly, and those of 6-deoxyhexoses vanished almost completely whilst dark-blue spots due to the 2,6-deoxyhexoses and weaker dark-orange spots of 2-deoxyhexoses were made clearly visible.

A good result was also obtained on heating chromatograms sprayed with a saturated solution of 2-thiobarbituric acid in glacial acetic acid for 5–7 min at 150–160° and then keeping them for some minutes over boiling water. The spot of xylose was a dirty-olive color, that of glucose yellow-brown, that of the 6-deoxyhexoses dark-yellow, and that of all the 2-deoxyhexoses and 2,6-deoxyhexoses red.

These methods permit detection and discrimination of 10 γ or more of 2-deoxyhexoses and of 5 γ or more of other sugars. The spots are well defined, the paper background homogeneous.

The method described may be useful in other sugar separations *e.g.* in bacteriological research.

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