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Note

Chromatographic differentiation of tamarixetin and isorhamnetin by spraying

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In the field of chemosystematics, trace amounts of flavonoids are often encountered. The identification of such components is sometimes difficult to achieve, especially if isomers are involved. Isomeric flavonoids are known to occur in nature, and their chromatographic separation is in many instances very difficult to effect. Only a few isomeric components have been successfully separated from one another by using either paper or thin-layer chromatography[1]. Two isomers that we found impossible to separate chromatographically, although applying a large number of solvent systems and using both paper and thin-layer chromatography, are tamarixetin (3,5,7,3′-tetrahydroxy-4′-methoxyflavone) and isorhamnetin (3,5,7,4′-tetrahydroxy-3′-methoxyflavone).

Sodium ethoxide and methoxide have been used to induce shifts during UV analysis of flavonoids[2–4]. When applying sodium ethoxide (0.05 *M*, in absolute ethanol) as a spray reagent, it was observed that tamarixetin gave a bright yellow colour, which was stable for up to 24 h. In contrast, the yellow colour that developed with isorhamnetin was unstable and faded after about 2 h. A difference in colour was also noted under UV light, but the contrast was not so sharp.

Based on these differences, it was possible to differentiate between the two isomers, when present in trace amounts (as low as 1.5 μg) on paper. Similar, but weaker, distinctions were also noticeable in thin-layer chromatography when using silica gel and polyamide. The colours of the 3-glucosides of isorhamnetin and quercetin were also brighter and more stable than their corresponding aglycones.

Although the flavonoids tamarixetin and isorhamnetin have not been reported to occur together in nature, the development of a stable bright colour should not exclude the presence of isorhamnetin. Both flavonoids would be expected to be present as their glycosides, which would be easier to separate than the respective aglycones.

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