

SHORT COMMUNICATION

ISOLATION AND IDENTIFICATION OF SESELIN  
IN *CITRUS* ROOTS

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**Abstract**—Seselin, a pyranocoumarin, has been isolated from roots of Shamouti orange, sour orange, sweet lime and grapefruit. It is specific to the roots and has not been detected in other plant organs. It was identified by m.p., TLC, u.v., i.r., NMR and mass spectral comparison with authentic material.

INTRODUCTION

MANY publications deal with the phenolic compounds of citrus fruits,<sup>1-5</sup> but only little is known about the phenols in other organs of this plant. During a study of phenolic distribution in citrus, an unknown compound—which is present only in roots—was detected. Its identification is described in this paper.

RESULTS

A paper chromatographic survey of flavonoids in fruits, leaves, shoots and roots of the Shamouti orange tree was carried out on dried and powdered plant material extracted with either 80% methanol or 0.2 N NaOH, using Whatman No. 1 paper and *n*-butanol–acetic acid–water (6:1:2, v/v). After u.v. examination chromatograms were developed with benzidine reagent. Using both extraction methods, a compound at  $R_f$  0.95 was found in root extracts only, which gave a red colour with benzidine. Root extracts of Marsh seedless grapefruits, sweet lime and sour orange roots gave the same compound. A detailed study of its distribution in young Shamouti orange seedlings, grown in both dark and light, confirmed that it was specific to the roots.

The unknown compound was isolated by homogenizing fresh sour orange roots in distilled water, followed by steam distillation, and chromatography on a silica gel-H column. The compound, m.p. 116–118, was identified as seselin by direct comparison with an authentic sample. The mixed m.p. was undepressed and the i.r. and u.v. spectra ( $\lambda_{\max}^{99\%}$  isopropanol 294,330, with a shoulder between 283–288 nm) were superimposable. The structure of this

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compound was further substantiated by analysis of its mass ( $m/e=228$ ) and NMR spectra, and by co-chromatography on silica gel-H with benzene-ethyl acetate (1:1 v/v) ( $R_f$  0.77).

### DISCUSSION

This is the first report of seselin in *Citrus*. This coumarin is mentioned in few publications only,<sup>6-11</sup> and has previously been obtained from Umbelliferae, e.g. from the essential oils of *Seseli indicum*.<sup>9</sup> It is also present in the fruits of *Ruta pinnata*,<sup>7</sup> which belongs to the same family (Rutaceae) as *Citrus*. *Citrus* roots appear to be a good source of this compound. Its specificity to one organ of the plant is not surprising since studies in apple and pears<sup>10</sup> and in other plants<sup>12</sup> show that the quantity and quality of phenolic compounds may vary considerably in different organs of the same plant.

### EXPERIMENTAL

#### *Isolation of Seselin*

Washed and frozen roots ( $-20^\circ$ ) were unfrozen, rewashed and homogenized (50 g fresh material in 250 ml water) at 24,000 rev/min. The homogenate was filtered through two layers of gauze and the filtrate centrifuged for 5 min at 2500 rev/min. The supernatant was steam distilled and the distillate extracted (3  $\times$  85 ml) with ether. The paste, obtained on evaporation of the ether, deposited crystals from cold 96% MeOH. Chromatography on a silica gel-H (Merck) column (2.0  $\times$  30.0 cm) eluted with benzene-methanol-acetic acid (45:2:0.125, v/v) gave pure seselin.

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