

SHORT COMMUNICATION

THE CONVERSION OF THE 8-C-GLYCOSYLFLAVONE  
VITEXIN TO THE 6-ISOMER, ISOVITEXIN, IN *LEMNA*  
*MINOR*

JAMES W. WALLACE

Department of Biology, Western Carolina University, Cullowhee, North Carolina 28723, U.S.A.

and

TOM J. MABRY

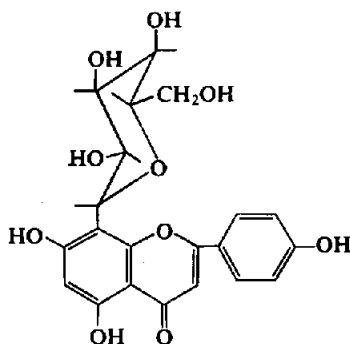
The Cell Research Institute and the Department of Botany, The University of Texas at Austin, Austin, Texas 78712, U.S.A.

(Received 21 February 1970)

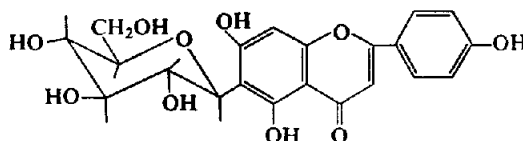
**Abstract**—<sup>14</sup>C-Labeled vitexin (I) and isovitexin (II), which were obtained from *Spirodela* species cultured on a medium containing phenylalanine-1-<sup>14</sup>C, were introduced in separate experiments into the growth media for *Lemna minor* plants, a species which normally produces both of these C-glycosylflavones. Under these conditions, vitexin was converted into isovitexin but the reverse transformation was not observed.

INTRODUCTION

MOST C-glycosylflavones are known to occur naturally in pairs as 6- and 8-C-glycosyl isomers,<sup>1</sup> and although it has been established<sup>2-6</sup> that for at least one of these pairs, vitexin (I) and isovitexin (II), an equilibrium mixture is readily reached during an acid reflux of either isomer, nothing is known with regard to their *in vivo* interconversions.



Vitexin (I)



Isovitexin (II)

<sup>1</sup> R. E. ALSTON, in *Recent Advances in Phytochemistry* (edited by T. J. MABRY), pp. 305-327, Appleton-Century-Crofts, New York (1968).

<sup>2</sup> M. K. SEIKEL and T. A. GEISSMAN, *Biophys.* **71**, 17 (1957).

<sup>3</sup> R. M. HOROWITZ and B. GENTILI, *Chem. & Ind.* 498 (1964).

<sup>4</sup> M. K. SEIKEL, J. H. S. CHOW and L. FELDMAN, *Phytochem.* **5**, 439 (1966).

<sup>5</sup> J. W. WALLACE, Ph.D. Dissertation, The University of Texas at Austin (1967).

<sup>6</sup> J. W. WALLACE, T. J. MABRY and R. E. ALSTON, *Phytochem.* **8**, 93 (1969).

As an extension of our previous investigation of the biogenesis of *C*-glycosylflavones<sup>6</sup> in the family Lemnaceae, we decided to determine whether or not the 6- and 8-*C*-glycosylflavones, vitexin and isovitexin, were interconverted in plants such as *Lemna minor*, a species which normally synthesizes both isomers.

Using the techniques previously described,<sup>6</sup> we obtained results which indicate that in *L. minor* vitexin is converted to isovitexin but, as previously observed,<sup>6</sup> the reverse transformation does not occur in this species.

## RESULTS AND DISCUSSION

The <sup>14</sup>C-labelled vitexin and isovitexin samples were obtained as previously reported.<sup>6</sup> When *Lemna minor* plants were cultured in the presence of <sup>14</sup>C-vitexin, radioactivity was subsequently detected autoradiographically in the two-dimensional chromatographic spots corresponding to both vitexin and isovitexin. In contrast, when <sup>14</sup>C-isovitexin was employed radioactivity was detected only in isovitexin.

The following additional evidence supports the conclusion that vitexin was converted to isovitexin in *L. minor*: (1) the compounds corresponding to both *C*-glycosylflavones were isolated from the chromatograms and spectrally identified by standard procedures;<sup>7</sup> (2) the chromatographically pure <sup>14</sup>C-labeled isovitexin (formed in *L. minor* from <sup>14</sup>C-labeled vitexin) was converted into an equilibrium mixture of <sup>14</sup>C-labeled vitexin and <sup>14</sup>C-labeled isovitexin by an acidic reflux; as expected, the radioactivity became distributed almost equally between vitexin and isovitexin (see Table 1); (3) finally, the formation of isovitexin did not

TABLE 1. SPECIFIC ACTIVITY DATA FOR ISOVITEXIN AND PRODUCTS THEREFROM

<div style="display: flex; align-items: center; justify-content: center;"> <div style="text-align: right; margin-right: 10px;">Isovitexin (1.7 <math>\mu\text{C}/\text{mM}</math>)*</div> <div style="text-align: center; margin-right: 10px;"> <math>\xrightarrow{\text{H}^+}</math> </div> <div style="display: flex; flex-direction: column; align-items: center;"> <div style="text-align: left; margin-right: 10px;">vitexin (1.8 <math>\mu\text{C}/\text{mM}</math>)†</div> <div style="text-align: left; margin-right: 10px;">isovitexin (0.9 <math>\mu\text{C}/\text{mM}</math>)†</div> </div> </div>		→	vitexin (1.9 $\mu\text{C}/\text{mM}$ )‡
		→	isovitexin (1.5 $\mu\text{C}/\text{mM}$ )‡

\* Specific activity for a sample of isovitexin isolated from *Lemna minor* plants cultured in the presence of <sup>14</sup>C-labeled vitexin; after isolation, the sample was diluted with non-labeled isovitexin.

† Specific activities for the compounds obtained by purifying the acidic equilibrium products by two-dimensional paper chromatography.

‡ Specific activities after the products were further purified by a second two-dimensional paper chromatographic run.

appear to be an artifact of the experiment since (a) the same results were obtained from different experiments one of which purposely employed room temperatures or below for all isolation procedures and (b) under the same experimental conditions, isovitexin was not converted to vitexin.

The results suggest that in *L. minor* one biosynthetic route to isovitexin is via vitexin.

<sup>7</sup> T. J. MABRY, K. R. MARKHAM and M. B. THOMAS, *The Systematic Identification of Flavonoids*, Springer-Verlag, New York (1970).

### EXPERIMENTAL

The *Lemna minor* plants (clone 5, McClure)<sup>8</sup> were maintained, cultured with <sup>14</sup>C precursors, harvested and extracted as previously described and the chromatographic and autoradiographic procedures were also those previously employed.<sup>6</sup> The acidic reflux conditions were 2 hr in 1.5 N HCl.

*Acknowledgements*—This work was supported by the National Science Foundation (Grant GB-5448X) and the National Institutes of Health (Grant GM-11111). The late Professor Ralph E. Alston participated in the early stages of this investigation.

J. W. McCLURE, Ph.D. Dissertation, The University of Texas at Austin (1964).