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BIFLAVONES AND THE AFFINITIES OF *CUPRESSUS FUNEBRIS*

PAUL A. GADEK and CHRISTOPHER J. QUINN

School of Botany, University of New South Wales, P.O. Box 1, Kensington, 2033, Australia

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Abstract—The biflavonyl profile of *Cupressus funebris* consists of major amounts of amentoflavone and cupressuflavone, minor concentrations of hinokiflavone, 7″-monomethylhinokiflavone, and a monomethylrobustaflavone, and traces of monomethylamentoflavone and cupressuflavone derivatives. This is consistent with the pattern reported for other *Cupressus* species, and does not support the transfer of this species to *Chamaecyparis* as has been proposed.

INTRODUCTION

Cupressus funebris Endl. is widely distributed in central China. Recent taxonomic work on this species has suggested that it shows a few morphological character-states (dimorphic leaves, small cone size and low seed number) which occur more commonly in the genus *Chamaecyparis*, and which have caused some authors to place it in that genus [1–4]. Although previous chemotaxonomic work on wood tropolones supported its retention in *Cupressus* [5], the specimens used in that study originated from northern India, an area now considered outside the geographic range of *Cupressus funebris* [2]. Similarly, a specimen of *C. funebris* previously investigated for biflavonyls [6] was described as being of 'Indian' origin. Since no voucher specimen was cited in either case, it is impossible to check whether the material did in fact fall within the limits of the species; in the former case it has been suggested by Silba [4] that it actually belonged to *Cupressus corneyana* Carr. (= *C. torolosa* D. Don ex Lamb.).

Here we report on the leaf biflavonyls of a small amount of leaf material obtained from a collection of *C. funebris* from the Chekiang Province of China, the morphology of which falls within the limits of the species as defined by Franco [1] and Silba [4,7, Hunt, personal communication].

RESULTS AND DISCUSSION

Permethylation of an ethanolic leaf extract yielded amentoflavone hexamethyl ether, cupressuflavone hexamethyl ether, hinokiflavone pentamethyl ether and robustaflavone hexamethyl ether. Amentoflavone and cupressuflavone were identified as the major biflavonyl components of the leaf. As well, minor concentrations of hinokiflavone and its 7″-monomethyl ether were detected. All bands were checked by permethylation, and the hinokiflavone band showed evidence of trace amounts of amentoflavone and cupressuflavone monomethyl ethers. A third minor constituent chromatographing between the amentoflavone and hinokiflavone bands yielded robustaflavone hexamethyl ether on permethylation. The R_f of this band indicates that it is a monomethyl ether of robustaflavone. An attempt was made to characterise the compound using UV spectral shifts as outlined in ref. [8]. Band I showed a large bathochromic shift on the addition of sodium methoxide (+ 54 nm) with a slight decrease in intensity, a result similar to that described for compounds with both 4′ and 7 positions free on the same flavone unit. Band II showed a small bathochromic shift with a large increase in intensity in both sodium methoxide and sodium acetate, suggesting both 7 positions are free. These results suggest that one of the 4′ positions is methylated. The small size of the available material precluded further analysis.

The identification of a minor constituent as a monomethylrobustaflavone is the first report of this biflavonyl series in *Cupressus*, although robustaflavone derivatives have previously been reported as minor or trace constituents in a number of other genera of the family [9, 10].

Recent work has shown that members of the genus *Cupressus* are characterized by major amounts of amentoflavone and cupressuflavone in their leaves, sometimes with the addition of a minor or trace amount of one or two monomethyl ethers; the more highly methylated biflavones are absent [9]. In *Chamaecyparis* species, however, di- and trimethyl ethers of amentoflavone are conspicuous, while the cupressuflavone series is typically absent [except for the aberrant member of the genus, *C. nootkatensis* (D. Don) Spach, where a minor amount of cupressuflavone has been reported] [9]. Clearly, the biflavonyl profile of *Cupressus funebris*, having major bands of amentoflavone and cupressuflavone, aligns with that of *Cupressus* rather than of *Chamaecyparis*. This suggests that the characters of dimorphic leaves, cone size and seed number used by Franco [1], seeds per bract scale used by Gaussen [3], or cone size and the liberation of seeds used by Silba [4] to delimit *Chamaecyparis* from *Cupressus* do not define the actual discontinuity between the genera. Not withstanding the difficulty in defining these genera on morphological characters [see 1], there exists a clearly defined chemical discontinuity in leaf biflavonyls and probably also in wood tropolones. Whether this is best recognised at the generic, subgeneric or sectional level is a question that requires, for its resolution, a much broader data base than presently exists.

It is hoped that work underway in this laboratory will help reach a solution.

EXPERIMENTAL

Dried leaf material was obtained from an herbarium specimen lodged at Kew: Univ. Nanking Herb. No. 14809, C. Y. Chiao, China: Chekiang Prov: Yen Tang Shan. Extraction and identification of biflavonyls was carried out by the methods described previously [9]. UV spectral data of the monomethylrobustaflavone: λ_{max}^{MeOH} nm: 269, 283 (sh), 334; + NaOMe: 275, 307 (sh), 388; + NaOAc: 275, 310 (sh), 385.

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