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FOLIAR FLAVONOIDS OF
CAMELLIA CHRYSANtha (THEACEAE)

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ABSTRACT

Foliar flavonoids of Camellia chrysantha consist of two flavone C-glycosides which have chromatographic and spectroscopic properties very similar to corymboside and isocorymboside. The lack of detectable foliar flavonol glycosides contrasts with their presence in flowers of this species and may be anomalous within the genus Camellia.

Key words: Camellia chrysantha, Theaceae, flavonoids, flavone C-glycosides, tissue specificity.

INTRODUCTION

Recent studies of the petal flavonoids of Camellia chrysantha (Hu) Tuyama (Miyajima, Uemoto, Sakata, Arisumi, and Toki 1985; Scogin 1986) revealed the presence of the relatively uncommon flavonol, quercetin 7-O-glucoside. The present study seeks to determine whether this unusual flavonol glycoside is uniquely produced in the flowers or is present in all tissues of this species.

MATERIALS AND METHODS

Leaf material of Camellia chrysantha was provided by the staff of the Huntington Botanic Garden, San Marino, California, from plants in cultivation.

Leaf material (ca. 100 g) was ground and extracted in 80% methanol. The extract volume was reduced under vacuum, and aliquots were used as samples for two-dimensional paper chromatography (2D-PC) and column chromatography.

Solvents for initial 2D-PC separation on Whatmann 3MM paper were TBA and 15% HOAc (Mabry, Markham, and Thomas 1970). Preparative quantities of phenolic constituents were separated by column chromatography (3 x 10 cm) over Polyclar-AT. Elution began with water and progressed through increasing percentages of MeOH. Those eluted fractions which were found by one-dimensional paper chromatography (1D-PC) to contain phenolic compounds were combined, concentrated, and the compounds purified by 1D-PC in 15% HOAc. Purified compounds were characterized chromatographically and spectroscopically using standard methods (Mabry, Markham, and Thomas 1970).

RESULTS

Initial 2D-PC revealed three major phenolic constituents from the leaves of Camellia chrysantha, two of which appeared to be flavonoid in nature based upon fluorescence colors under UV light in the presence and absence of ammonia fumes.

Preparative amounts of these three compounds obtained from column chromatography were characterized chromatographically and spectroscopically. These
Table 1. Chromatographic properties of *Camellia chrysantha* flavonoids (Rf × 100).

|                  | Rf | 15% HOAc | TBA | Water | 2% HOAc | 3% NaCl |
|------------------|----|----------|-----|-------|---------|---------|
| *Camellia* C-glycosyl-flavone #1 | 19 | 45       | 15  | 17    | 38      | 16      |
| Isocorymboside (Chopin et al. 1977) | 18 | 44       | na  | na    | na      | na      |
| *Camellia* C-glycosyl-flavone #2 | 20 | 51       | 16  | 22    | 49      | 22      |
| Corymboside (Besson et al. 1979)   | 16 | 54       | na  | na    | na      | na      |

na: not available.

results for the two flavonoid compounds are summarized in Tables 1 and 2. The third compound yielded ellagic acid upon acid hydrolysis and was classified as a complex ellagitannin and was not further analyzed. The two flavonoid compounds were resistant to acid hydrolysis under standard conditions (Mabry, Markham, and Thomas 1970) and exhibited the spectral characteristics of flavones. These compounds were thus classified a flavone C-glycosides. The resistance of C-glycosides to routine hydrolysis precluded explicit identification of the sugar moiety, but spectroscopic and chromatographic properties of the flavonoids are consistent with those of isomeric forms of an apigenin 6,8-diglycoside. Among currently characterized members of this flavonoid class the chromatographic and spectroscopic characteristics of *C. chrysantha* C-glycosylflavones most nearly resemble corymboside (6-C-arabinosyl-8-C-galactosylapigenin) (Besson, Dombris, Raynaud, and Chopin 1979) and isocorymboside (6-C-galactosyl-8-C-arabinosylapigenin) (Chopin, Dellamonica, Besson, Skrzypczakowa, Budzianowski, and Mabry 1977). Identification of the *C. chrysantha* C-glycosylflavones must remain provisional at present until sufficient material is available for a more complete chemical characterization.

DISCUSSION

Polyphenolic compounds constitute as much as 30% of the dry weight of commercial tea and contribute significantly to the color, strength, and briskness of brewed tea. By virtue of their commercial interest to the tea industry, the polyphenolics of tea leaves have been extensively studied (Roberts 1962). The leaves of the commercial tea plant (*Camellia sinensis* Kuntze) as well as numerous related *Camellia* species, have been reported by Roberts, Wight, and Wood (1958) to be rich in flavonol glycosides. Sakamoto (1967) reported the occurrence of incompletely characterized apigenin C-glycosides as minor constituents of green tea infusion which also contained large amounts of flavonol glycosides. In subsequent studies Sakamoto (1968, 1970) identified the C-glycosylflavones as saponarin, isovitexin (saponaretin), and two isomeric forms of 6,8-diC-glycosylapigenin. These apigenin C-glucosides of green tea are spectroscopically and chromatographically distinct from the apigenin C-glycosides of *C. chrysantha*. Recently 6,8-diC-arabinosyl apigenin has been reported from *Thea sinensis* Sims (= *Camellia sinesis* Kuntze) (Chaboud, Raynaud, and Debouchet 1986). Our appreciation of the diversity of C-glycosylapigenins produced in the leaves of *Camellia* species is increasing and further expanded by the current report of corymboside and isocorymboside from leaves of *C. chrysantha*. 
Table 2. Spectroscopic properties of *Camellia* C-glycosyl flavones (absorption maxima in nm, shoulders in parentheses).

|                      | Camellia C-glycosyl flavone #1 | Isocorymboside (Chopin et al. 1977) | Camellia C-glycosyl flavone #2 | Corymboside (Besson et al. 1979) |
|----------------------|---------------------------------|-------------------------------------|---------------------------------|---------------------------------|
| MeOH                 | 274, 333                        | 271, 332                            | 274, 331                        | 274, 336                        |
| + NaOMe              | 285, 331, 396                   | 281, 331, 399                       | 283, 335, 395                   | 281, 399 (+NaOH)                |
| + AlCl               | 281, 307, 351, (386)            | 280, 306, 355, (386)                | 281, 306, 350, 388              | 283, 306, 352, 385              |
| + AlCl/HCl           | 281, 304, 344, (380)            | 279, 302, 346, (379)                | 281, 304, 343, 385              | 283, 306, 350, 385              |
| + NaOAc              | 284, 336, 391                   | 280, 332, 396                       | 283, (339), 393                 | 284, 399                        |

**Comparative Phytochemistry**

The foliar flavonoids of *C. chrysantha* contrast with the general pattern within this genus in that no flavonol glycosides were detected in foliar extracts. The limited foliar sample which was available (ca. 100 g) may have precluded detection of minor constituents. Other studies of tea leaf constituents often begin with samples as large as 20 kg (Sakamoto 1968). It can, however, be reliably stated that C-glycosylflavones, rather than flavonol glycosides, are the major flavonoid constituents in leaves of *C. chrysantha*. This situation is unique among *Camellia* species examined to date and could constitute a phytochemical feature characteristic of the section *Chrysantha* within this genus.

**Tissue Specificity of Camellia Flavonoids**

Only very limited data are available regarding tissue specificities of flavonoid biosynthesis and accumulation among *Camellia* species. In only a few cases have both floral and foliar flavonoids of the same taxon or cultivar been adequately examined and among these, tissue specificities with respect to flavonoid profile have been noted (C. F. Parks, pers. comm.). Quercetin 7-O-glucoside was recently reported as the major floral flavonoid of *Camellia chrysantha* (Miyajima et al. 1985), accompanied by lesser amounts of rutin and quercetin 3-O-glucoside (Sco- gin 1986). Quercetin 7-O-glucoside was undetectable in foliar material of *C. chrysantha*. A similar tissue specificity in flavonol 7-glucosylation activity has been reported from *Baptisia sphaerocarpa* Nutt. and *B. tinctoria* R.Br. (Alston, Rosler, Naifeh, and Mabry 1965). Clearly, the activity of the enzymes of flavonoid glucosylation are subject to strict genetic regulation and this must be constantly borne in mind when performing chemosystematic comparisons.

The most striking phytochemical feature of *C. chrysantha* is the absence of detectable flavonol glycosides in leaf materials, as contrasted with floral materials. The production of flavone C-glycosides rather than flavonol glycosides in *C. chrysantha* leaves again emphasizes the tissue-specific regulation of flavonoid biosynthesis, a topic rich in possibilities for subsequent investigation.

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