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ELLAGITANNINS AS ULTRAVIOLET-ABSORBING FLORAL PIGMENTS IN *CROSSOSOMA CALIFORNICUM*

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INTRODUCTION

The reflection and absorption of ultraviolet (UV) wavelengths by floral structures impart colors and color patterns to flowers which are invisible to humans, but visible to pollinating insects. In all cases reported to date, the pigments shown to be responsible for floral UV absorption have been flavonoids (Thompson et al. 1972; Brehm and Krell 1975; Scogin and Zakar 1976). A recent report on the reproductive biology of Crossosomataceae noted two apparently inconsistent features: 1) low petal UV reflectance (less than 5%) and 2) the notable lack of detectable petal flavonoids (Scogin and Tatsuno 1982). These results raised the question of what pigment was accomplishing the UV absorption in *Crossosoma* petals, which is addressed in the present report.

MATERIALS AND METHODS

Floral materials of *Crossosoma californicum* Nutt. (Crossosomataceae) were collected from plants in cultivation at the Rancho Santa Ana Botanic Garden. Spectral reflectance measurements from freshly collected petal materials were performed using a Perkin-Elmer spectrophotometer equipped with an integrating sphere attachment. Vacuum infiltration of petal materials was accomplished by immersion of petals in distilled water at reduced pressures.

Phenolic extraction, chromatographic, and photographic techniques were as previously described (Tatsuno and Scogin 1978; Scogin and Tatsuno 1982).

RESULTS AND DISCUSSION

Extent of UV absorption.—The percentage of incident UV light which is reflected from *Crossosoma californicum* petals over the wavelength range 270–390 nm is 3 to 5%. Leaves of *C. californicum* reflect 5–7% of UV light over the same wavelength range. The similarity between floral and foliar reflectances over this wavelength range suggests that UV wavelengths contribute relatively little to the floral-foliar color contrast perceived by visiting insects.

Source of UV absorption.—Strickland (1974) noted that light reflection

from white flower petals of wavelengths visible to humans was due in part to physical (structural) phenomena involving reflection at air-water interfaces of the intercellular spaces in the mesophyll tissue of petals. Reflectance was substantially reduced across the human visibility spectrum by vacuum infiltrating these spaces with distilled water. Strickland attributes light reflectance (at wavelengths visible to humans) in white flowers solely to structural features.

Results consistent with Strickland's were obtained from the white petals of *Crossosoma,* in which petal reflectance decreased by 20% (from 50% to 30% reflectance) uniformly over the wavelength range 690–440 nm when petals were vacuum infiltrated with distilled water. Reduction in reflectance decreased from 18% at 430 nm to 1% at 390 nm. From 390 nm to 260 nm the reflectances before and after infiltration were indistinguishable. Whereas vacuum infiltration with water substantially reduced reflection in *Crossosoma* petals over the human visible spectrum, it has no effect on the reflectance of UV wavelengths. It is, therefore, concluded that UV reflectance in this case is dependent upon chemical properties (i.e., the presence of pigment molecules) and not upon physical/structural properties of the petal anatomy.

The nature of the UV absorbing pigments.—All efforts to detect flavonoid compounds in the petals of *Crossosoma californicum* have been unsuccessful; however, the presence of large quantities of ellagitannins has been demonstrated (Tatsuno and Scogin 1978). In the present investigation, when chromatograms of methanolic extracts of petals were photographed through an 18A, UV-transmitting filter, only a single discrete, weakly UV-absorbing spot was detectable. This spot exhibited a weak, purple fluorescence under UV illumination and corresponded to the compound numbered 4, which was purified from petals of *C. californcum* and *C. bigelovii* Wats. by Tatsuno (1976). Tatsuno (1976) showed that this compound was unaltered by glucosidase, but yielded both gallic and ellagic acid upon acid hydrolysis. This compound was determined to be a complex ellagitannin and is responsible for at least part of the UV absorption of *Crossosoma* petals.

In the present investigation, a pale-yellow precipitate formed in the methanolic, *Crossosoma* petal extract after storage for one week at −20 C. Precipitate formation under these conditions is characteristic of complex tannin substances. Both the supernatant and resuspended precipitate were again chromatographed and the chromatograms photographed through a UV-transmitting filter. The supernatant chromatogram revealed no UV-absorbing substances, but a strongly UV-absorbing material was detected on the chromatogram of the precipitate. This material has no mobility in the organic solvent (TBA) and little mobility in the aqueous solvent (HOAc). Such mobilities are characteristic of complex tannin structures.

From these results it is concluded that the pigments responsible for UV

absorption in petals of *Crossosoma californicum* are complex ellagitannins, rather than the more commonly encountered flavonoid compounds.

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