

## UNUSUALLY STABLE MONOACYLATED ANTHOCYANIN FROM PURPLE YAM *DIOSCOREA ALATA* 1)

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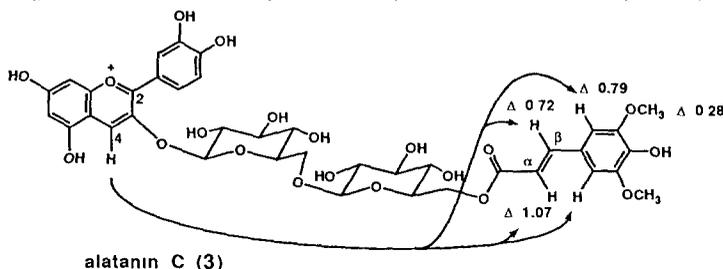
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**Abstract:** Alatanin C isolated from the tuber of purple yam *Dioscorea alata* is the first discovered unusually stable monoacylated anthocyanin in neutral aqueous solutions. The stability is ascribed to the intramolecular stacking of sinapic acid and to the chiral self-association of anthocyanidin nuclei.

Polyacylated anthocyanin pigments,<sup>2a)</sup> which contain two or more cinnamic acid derivatives, are very stable to decolorization in neutral or weakly acidic aqueous solutions. Intramolecular sandwich-type stacking of acids has been considered to prevent anthocyanidin nuclei from hydration at its 2-position and subsequent loss of color.<sup>2)</sup> Recently we have isolated alatanin C from pigments of purple yam and found it to be unusually stable anthocyanin although it is only monoacylated.<sup>1)</sup> Here we propose a possible mechanism for its stabilization.

The alatanins are very resistant to hydration at pH 6.0 as shown in Fig. 1. Alatanin A (**1**)<sup>3)</sup> and B (**2**)<sup>4)</sup> having two sinapic acid are stabilized by sandwich-type stacking.<sup>2)</sup> Surprisingly, monoacylated alatanin C (**3**) is also quite stable, despite the fact that monoacylated anthocyanins like shisonin are generally decolorized quickly.



In the circular dichroism (CD) spectrum of **3** an exciton-type positive Cotton effect in the visible region is observed (Fig. 2) indicating that the anthocyanidin nuclei of **3** are aggregated clockwise. This chiral self-association was verified as follows; the exciton-type Cotton effect was shown to be concentration dependent<sup>5)</sup> and addition of the co-pigment, flavocommelin, caused disappearance of the Cotton effect in the CD.<sup>2a,6)</sup> Simple anthocyanins *i. e.* pelargonin and delphin are stabilized by self-association at concentration of  $\geq 5 \times 10^{-3} \text{M}$  and chiral stacking of these anthocyanidin nuclei is suggested from their concentration dependent Cotton effects.<sup>5)</sup> At  $5 \times 10^{-5} \text{M}$  most anthocyanins exist in a monomeric state, and no exciton-type Cotton effect is observable in the visible region.

In alatanin C sinapic acid also plays an important stabilizing role. Deacylated **3** (**4**)<sup>7)</sup> does not show exciton-type Cotton effect in the visible region (Fig. 2) and is rapidly hydrated at position 2 (Fig. 1). Sinapic acid may allow **3** to undergo the self-association more easily and the intramolecular stacking of acid residues may also contribute to stabilization by sterically preventing hydration of the anthocyanidin nuclei. In acidic D<sub>2</sub>O, **3** showed long distance NOEs between the signals of the anthocyanidin nucleus and the sinapic acid residue, and the chemical shifts of the protons of the sinapic acid were observed at higher field than those of methyl sinapate probably due to an anisotropic effect of the cyanidin ring current (see Fig. 3). The same stacking of sinapic acid may occur in neutral solution. Thus, the unusual stability of **3** might be caused by the cooperation of unique self-association and intramolecular stacking (Fig. 3).

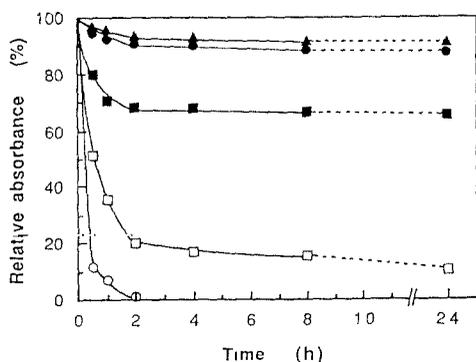


Fig. 1. Stability of alatanins  $5 \times 10^{-5} M$  in  $0.05 M$  phosphate buffer (pH 6.0) at  $20^\circ C$ . ●: **1**, ▲: **2**, ■: **3**, □: **4**, ○: shisonin **8**).

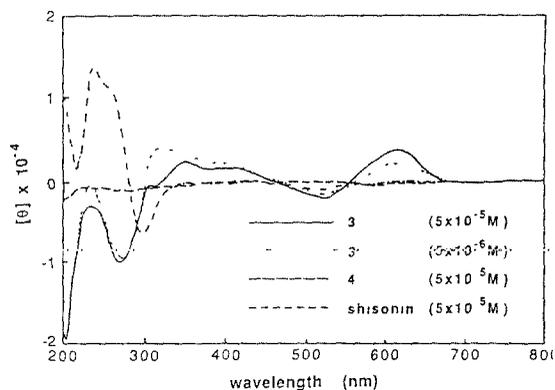


Fig. 2. CD of **3**, **4** and shisonin in  $0.05 M$  phosphate buffer (pH 6.0) at  $20^\circ C$ .

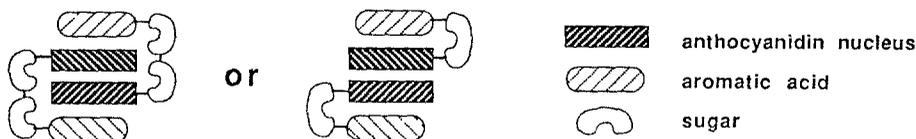


Fig. 3. Proposed stacking structure of alatanin C.

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## References and Notes

1. The isolation and structural determination of alatanin A, B and C is described in the preceding paper of Yoshida, K *et al*.
2. (a) Goto, T., Kondo, T. *Angew. Chem. Int. Ed. Engl.*, **1991**, *30*, 17; Goto, T. *Prog. Chem. Org. Nat. Prod.*, **1987**, *52*, 158; (b) Brouillard, R. *Phytochemistry*, **1983**, *22*, 1311.
3. 3-O-(6-O-(3-O-(β-D-glucopyranosyl)-6-O-(E)-sinapyl-β-D-glucopyranosyl)-β-D-glucopyranosyl)-7-O-(6-O-(E)-sinapyl-β-D-glucopyranosyl)-2'-O-(β-D-glucopyranosyl)cyandin.
4. 3-O-(6-O-(6-O-(E)-sinapyl-β-D-glucopyranosyl)-β-D-glucopyranosyl)-7-O-(6-O-(E)-sinapyl-β-D-glucopyranosyl)-2'-O-(β-D-glucopyranosyl)cyandin.
5. Hoshino, T., Matsumoto, T., Harada, N., Goto, T. *Tetrahedron Lett.*, **1981**, *22*, 3621; Goto, T., Tamura, H., Kondo, T. *Tetrahedron Lett.*, **1987**, *28*, 5907; Goto, T., Yoshida, K., Yoshikane, M., Kondo, T. *Tetrahedron Lett.*, **1990**, *31*, 713.
6. Addition of flavocoumestrol reduced the magnitude of the Cotton effect and with its 8 eq. the Cotton effect disappeared.
7. Deacylation of **3** was achieved with 1N NaOH-MeOH 1:1 at  $0^\circ C$  under nitrogen.
8. Kondo, T., Tamura, H., Yoshida, K., Goto, T. *Agric. Biol. Chem.*, **1989**, *53*, 797.