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SHORT REPORTS

RELATIONSHIP BETWEEN INHIBITORY ACTIVITY OF MYRMICACIN ANALOGUES ON *CAMELLIA JAPONICA* POLLEN GERMINATION AND THEIR LIPOPHILICITY

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Key Word Index—*Camellia japonica*; Theaceae; pollens; germination; inhibition; lipophilicity; HPLC; myrmicacin; fatty acid; 2-hydroxy fatty acid.

Abstract—Inhibitory activity of C_6 to C_{11} fatty acids and their corresponding 2-hydroxy derivatives to *Camellia japonica* pollen germination is related to their lipophilicity.

INTRODUCTION

Myrmicacin (3-hydroxydecanoic acid) is a herbicidal compound isolated from a South American leaf-cutting ant in 1971 [1], and studies on its biological activity and related compounds to various substrates such as pollens [2–6] bacteria [7], fertilized sea urchin eggs [8, 9] and human erythrocytes [10] have been reported. In 1979 Iwanami and Iwadare [3] demonstrated that the inhibitory activity of C_8 to C_{11} aliphatic monocarboxylic acids on pollen growth was comparable to that of myrmicacin and they suggested that the acids should be regarded as a new group of growth inhibitors named myrmic acids (MYA).

Our recent study [11] revealed that the degree of inhibition of myrmicacin analogues on pollen germination was related to the degree of unsubstitution of the molecules. We assumed that the degree of unsubstitution is related to the lipophilicity of the substances and therefore it seemed of interest to investigate the relationship between biological activity of the tested acids and their lipophilicity.

Recently Butte *et al.* [12] have suggested that chromatographically evaluated log k^0 § values should be used, instead of conventionally defined log P_{oct} values, to measure lipophilicity. As Butte's log k^0 value seemed suitable as an indication of lipophilicity for our study, it was employed in the present work.

RESULTS AND DISCUSSION

The acids submitted to this study were normal C_6 to C_{11} unhydroxylated and 2-hydroxy acids listed in Table 1. Evaluation of inhibitory activity of the tested acids was carried out using germination curves to *Camellia japonica* pollens at various concentrations of the acids appearing in our previous paper [11]. For the evaluation, the half-maximum germination potentials ($G_{0.5}$, mol) of the acids at which 50% of the sown pollens germinated were obtained as points of intersection of the curves and 50% germination line. The $PG_{0.5}(-\log G_{0.5})$ values were then calculated. Incidentally $G_{0.5}$ of 2-hydroxyhexanoic acid could not be evaluated because of its extremely weak activity [11].

Table 1. Tested acids and their $PG_{0.5}$ and log k^0 values

Acid	Number of carbon atoms	Unhydroxylated $PG_{0.5}$	Unhydroxylated log k^0	2-Hydroxy $PG_{0.5}$	2-Hydroxy log k^0
Hexanoic	6	3.08	1.24		0.63
Heptanoic	7	3.31	1.68	3.15	0.95
Octanoic	8	3.78	2.06	3.36	1.37
Nonanoic	9	3.88	2.50	3.78	1.72
Decanoic	10	3.99	2.84	3.90	2.03
Undecanoic	11	4.08	3.16	4.06	2.48

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§k (capacity ratio) $= (t_R - t_0)/t_0$ where t_R and t_0 are the retention times of a retained or an unretained peak respectively. The log k^0 value is calculated by extrapolation of a plot of log k for water-containing solvent mixtures to 100% water.

HPLC was carried out on an instrument equipped with a differential refractometer as detector. The stationary phase was Merck Hibar column (250 mm × 4 mm) LiChrosorb RP–18 (10 μm octadecyl-silylated silica gel) and the mobile phase (pressure = *ca* 56 kg/cm^2) was a mixture of tetrahydrofuran (THF) and water containing 0.01 mol phosphoric acid to suppress dissociation of the tested acids. The concentrations of THF in the mobile phase were 40%, 45%, 50% and 55%. The log k^0 values were carried out according to Butte's method [12] (Figs 1 and 2).

The obtained log k^0 and $PG_{0.5}$ values are given in Table 1 and plots of $PG_{0.5}$ against log k^0 value are shown in Fig. 3.

As seen in Fig. 3, the $PG_{0.5}$ of the tested acids is related to the log k^0 values. There is a considerable difference between the $PG_{0.5}$ of heptanoic acid (C_7) and that of octanoic acid (C_8) and the same difference is observed between the $PG_{0.5}$ of 2-hydroxyoctanoic acid (C_8) and that of 2-hydroxynonanoic acid (C_9). Previously we reported [3] a considerable difference between the inhibitory activity of C_7 and that of C_8. Furthermore, we have pointed out [11] that the inhibitory activities of 2-hydroxy acids are almost comparable to those of unhydroxylated acids having one less carbon atom. The result of the present work is in agreement with our previous reports.

The plot of $PG_{0.5}$ against log k^0 of unhydroxylated acids higher than C_8 is linear and the same is observed for 2-hydroxy acids higher than C_9. In a homologous series of the acids the factors other than the number of methylene groups, which is related to lipophilicity, are the same. Accordingly it seems reasonable to assume that the $PG_{0.5}$ is proportionally related to the log k^0 value for MYAs.

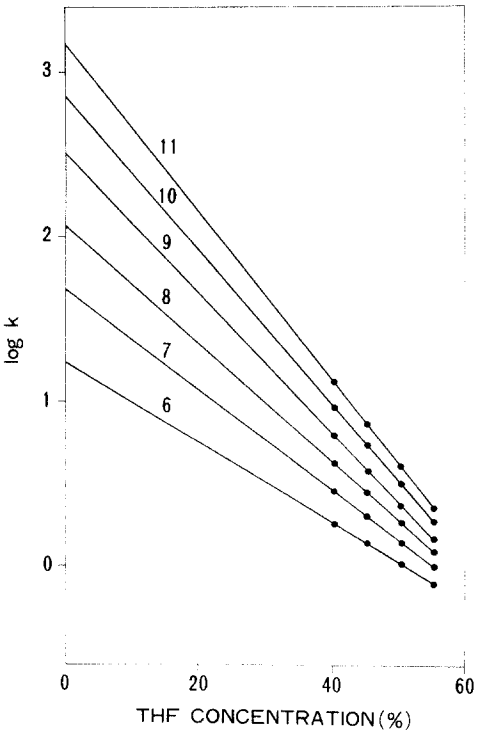

Fig. 1. Relationship between the log k^0 values of unhydroxylated acids and THF concentration in the mobile phase. The numbers along the lines indicate the number of carbon atoms of the acids.

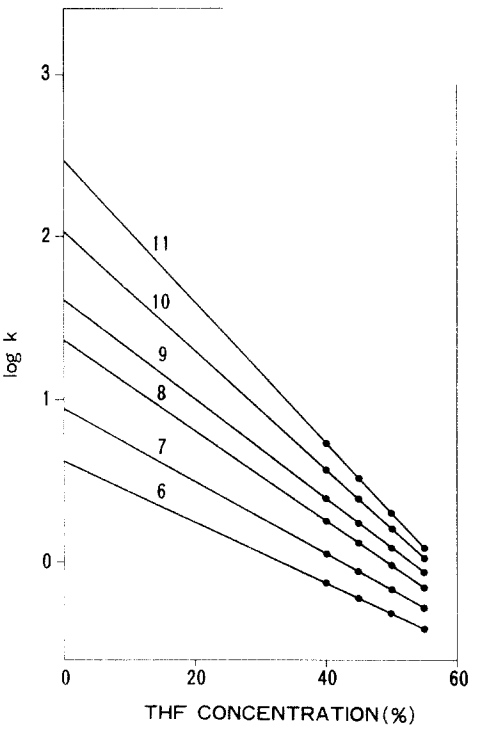

Fig. 2. Relationship between the log k^0 values of 2-hydroxy acids and THF concentration in the mobile phase. The numbers along the lines indicate the number of carbon atoms of the acids.

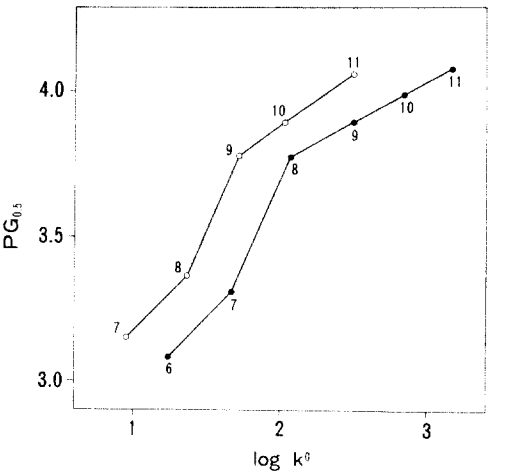

Fig. 3. Relationship between log k^0 and $PG_{0.5}$ of unhydroxylated and 2-hydroxy acids. ●, Unhydroxylated acids; ○, 2-hydroxy acids. The numbers by the symbols indicate the number of carbon atoms of the acids.

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CHANGES IN FREE AND BOUND FRACTIONS OF AROMATIC COMPONENTS IN VINE LEAVES DURING DEVELOPMENT OF MUSCAT GRAPES

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Key Word Index—*Vitis vinifera*; Vitaceae; Muscat of Alexandria; vine-leaf; terpenols; benzyl alcohol; 2-phenyl ethyl alcohol.

Abstract—Some aroma components of the grape *Vitis vinifera* cv Muscat of Alexandria (free and glycosidically bound terpenols, benzyl and 2-phenyl ethyl alcohols) were studied in vine leaves during the period of development and maturation of the fruit. These components seem to be synthesized in the leaf blade. The leaf stalk was characterized by high levels of terpenols, particularly free ones such as geraniol and citronellol.

INTRODUCTION

Muscat grape varieties are characterized by a special aroma, the most aromatic fraction of which is mainly made up of terpenols with, in addition, certain aromatic alcohols such as benzyl and 2-phenyl ethyl alcohols. A fraction of these compounds is also present in the form of glycosides which are not aromatic themselves but which constitute a large aroma potential [1, 2]. These compounds are highly concentrated in the berry where they increase during the development and the maturation of the fruit [3, 4]. Nevertheless, few data were available concerning their presence in the other parts of the vine plant. Essential oil from vine leaves has been reported containing some terpenols [5]. It was therefore particularly interesting to investigate whether the vine leaf, where sugars and flavonol glycosides are synthesized [6], was also the origin of free and glycosidically bound linalool, α-terpineol, citronellol, nerol, geraniol and benzyl and 2-phenyl ethyl alcohols previously studied in the berry [7] and to investigate their evolution at various physiological stages of the fruit.

RESULTS AND DISCUSSION

As shown in Fig. 1 and Table 1 the blade and stalk of the leaf contain terpenols and aromatic alcohols in their free and bound forms. These results are similar to those

*This paper is Part 3 of work on the aroma of grapes. For Part 2 see ref. [7].