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Identification of trans-Cinnamic Acid from Scrophularia Oldhami Oliver

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Identification of *trans*-cinnamic acid and *p*-hydroxycinnamic acid for estimation of the sequence of *p*-methoxycinnamic acid.

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禹麟根* 徐廷祥** 申國鉉* : 玄蔘成分 *trans*-Cinnamic Acid의 同定

玄蔘의 成分 *p*-methoxycinnamic acid의 生合成 機轉을 檢討하는 豫備試驗에서 trans-cinnamic acid 의 出現을 同定하였다.

Previous studies [1,2] on roots of *Scrophularia Oldhami Oliver* in this laboratory, have resulted in the isolation of *p*-methoxycinnamic acid, and its serum level has been discussed. In this paper, an attempt was made to identify the occuring of *trans*-cinnamic acid in the plant, since the component is implied as an intermediate in the formation of *p*-methoxycinnamic acid.

It has been suggested that methyl *p*-coumarate might be regarded as an intermediate in the metabolism of methyl *p*-methoxycinnamate by Lentinus lespideus and possibly in the biosynthesis of it [3]. It was shown that anethol was synthesized from phenylalanine through phenylpyruvic acid, cinnamic acid, and 4-hydroxycinnamic acid by *Foeniculum vulgare* [4-6]. Also, an acetone precipitate from an extract of spinach [7] and crude homogenates of pea seedlings or microsomes from the homogenates [8] catalyzed the hydroxylation of cinnamic acid to *p*-coumaric acid.

In view of those results, it might be supposed the presence of *trans*-cinnamic acid together with *p*-methoxycinnamic acid as an intermediate. Therefore, the distribution of *trans*-cinnamic acid, *p*-hydroxycinnamic acid and *p*-methoxycinnamic acid resolved into a study of the identification of them for further study using tracer.

EXPERIMENTAL

Isolation of *p*-methoxycinnamic acid.–It was conducted in the same procedure as described in a previous paper [1].

Isolation of trans-cinnamic acid-500 g. of fresh roots of *Scrophularia Oldhami Oliver* were

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extracted with 2 L. of 95% ethanol on 60° water bath for 3 hr. and the filtrate was concentrated under reduced pressure at 60° until a syrupy residue was left. It was then extracted with 300 ml. of ether and evaporated to a volume of 50 ml. and shaken with 10% of aqueous $NaHCO_3$ solution. The aqueous layer was acidified with *N* HCl and resulting precipitate was crystallized from 95% ethanol to give 0.34 g. of *p*-methoxycinnamic acid.

The filtrate freed from *p*-methoxycinnamic acid was extract with ether and the ethereal layer concentrated. From the residue, *p*-methoxycinnamic acid was removed by recrystallizing with water. The aqueous layer was shaken with ether and the ethereal solution was evaporated to dryness. This final residue was crystallized from 70% ethanol to give 0.12 g. of colorless crystals, m.p. 128°. Repeated recrystallization from 70% ethanol gave m.p. 133° undepressed upon admixture with an authentic sample, ν^{KBr}_{Max} 2550, 1690, 1615, 968 and 702 cm^{-1}.

Paper chromatography.–The filtrate freed from *p*-methoxycinnamic acid was shaken with ether and the organic layer was evaporated and then dissolved in 95% ethanol to be applied for chromatography.

An ascending paper chromatographic procedure was carried out on Whatman No. I paper at room temperature. The solvent system was n-BuOH: 3% NH_4OH (1 : 1) and detecting reagents were 0.4% brom phenol blue(A) and diazotized sulfanilic acid-10% $NaHCO_3$. (B)

The R*f* values of *p*-methoxycinnamic acid, *p*-hydroxycinnamic acid, and *trans*-cinnamic acid were identical with those of an authentic sample. (See Table I).

TABLE I. R*f* values of components

Component detected	R*f* Reference	Sample	Color by Reagent (A)	(B)
p-Hydroxycinnamic acid	0.18	0.18	Blue	Red
p-Methoxycinnamic acid	0.39	0.39	Blue	—
trans-Cinnamic acid	0.45	0.45	Blue	—
Unidentified	—	0.09	Blue	Orange

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