

GC-Mass Spectrometry of the Constituents of

Ligusticum wallichii Franch (I)

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INTRODUCTION

It is well known that *Ligusticum wallichii* Franch has been used as sedatives and anti-spasmodics in China for a long time. In the previous report,¹⁾ it showed that the papaverine-like action of the *Ligusticum* was six-fold to that of *Cnidium officinale* Makino, which is in the same family but in a different species, the atropine-like action of the former was twice to that of the latter and the antihistamine-like action of the former was non-significantly different from that of the latter. Furthermore, we confirmed that the primary antispasmodic active constituents were in the neutral oil portion. In the previous papers,^{2), 3)} some alkyl phthalides like n-butylidenephthalide, ligustilide, n-butylphthalide, cnidilide and neocnidilide were found in the neutral oil portion of *Cnidium officinale* Makino, however long-chain esters have not been reported yet. The neutral oil portion of *Ligusticum wallichii* Franch has not been studied in detail by chemical methods as well as GC-Mass spectrometry, either. The structures of long-chain esters containing in the *Ligusticum* are presented in this paper. The other series of alkylphthalides found in this plant will be reported in the near future.

It was found that there were seven long-chain esters, ethyl pentadecanoate, methyl isoheptadecanoate, ethyl hexadecanoate, ethyl isoheptadecanoate, ethyl heptadecanoate, ethyl isoctadecanoate and ethyl octadecanoate which could not be separated over a column of silicic acid and showed one spot (R_f 0.93) on thin-layer plate of silicic acid. However, they were successfully separated in the gas chromatograph and detected in the mass spectrometer. There were three isomeric pairs, shown as B & C in Fig. II, III, as D & E in Fig. III, and as F & G in Fig. IV. Their identifications were based on their total ion monitor chromatogram and (Fig. I.) mass spectra^{4), 5), 6), 7), 8)} shown as the following table:

Table I. The identified esters of fatty acid from *Ligusticum wallichii* Franch

Fraction	Component identified	M ⁺ ; m/e	Retention time (in mins.)
A	ethyl pentadecanoate	270	6.2
B	methyl isoheptadecanoate	284	7.5
C	ethyl hexadecanoate	284	9.5
D	ethyl isoheptadecanoate	298	11.1
E	ethyl heptadecanoate	298	12.1
F	ethyl iso-octadecanoate	312	14.0
G	ethyl octadecanoate	312	16.8

M⁺: a molecular ion,

EXPERIMENTAL

Extraction, Column and Thin-layer Chromatography of Long-Chain Esters

The dry rhizoma of *Ligusticum wallichii* Franch was collected in Taipei market*¹ in August of 1968. The rhizoma (1 Kg.) was cut into small pieces and refluxed with 95% ethyl alcohol for 48 hrs. The extract was filtrated and concentrated, then dissolved in chloroform. The chloroform soluble part was treated with 5% Na₂CO₃ and 2% NaOH aqueous solution in order to remove acidic and phenolic portion respectively. Then the neutral oil (38.4 g.) was obtained after being washed with distilled water and dried over anhydrous Na₂SO₄. After removal of the solvent, it was fractionally distilled under reduced pressure (4mm. Hg). The distillate (6g.) was chromatographed over a column of silicic acid*² and eluted with a mixture of n-hexane and dichloromethane (19:1, 9:1, 9:3). The R_f-value of each fraction (20ml.) was tested. Fractions (34~49) were established by thin-layer chromatography on silicic acid*³ with a mixed solvent of n-hexane and ethyl ether (8:2) and found one spot (R_f 0.93) with a blue color under UV light (ca. 350nm) or a brown color in iodine vapor, then combined together and yielded 312 mg.

Gas Chromatography-Mass Spectrometry of Long-Chain Esters

A small amount (2 μl., ether) of the combined portion was injected into a Hitachi 063-RMS-4 GC-Mass spectrometer. The flow rate of the carrier gas (helium) was kept at 1.7Kg./cm². Temperatures were: column (10% PEG, 3mm×2m): 200°C, injector: 250°C, helium separator: 270°C, ion source: 180°C. The ionizing current was 80 μA and the energy of the electron was 80eV. The total ion monitor was used as the detector of gas chromatography. The results are shown in figures I, II, III & IV.

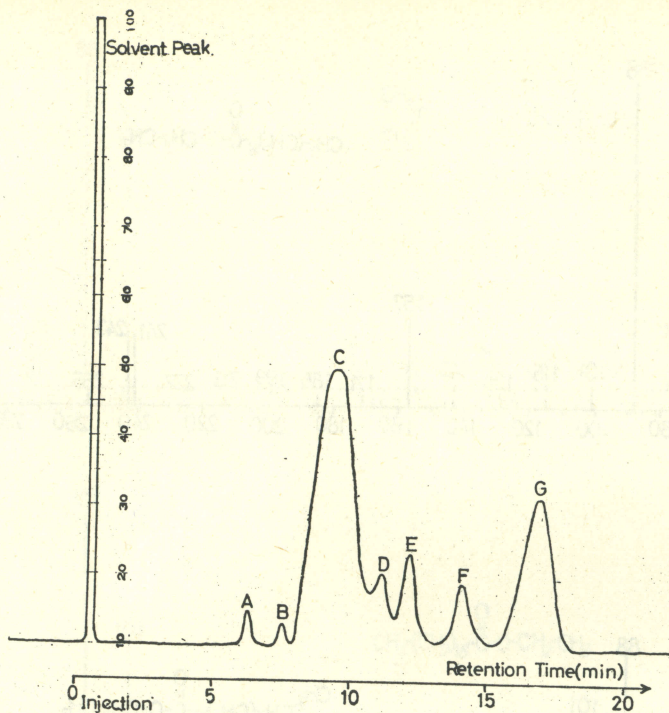


Fig. I. T. I. M. Chromatogram of Long-Chain Methyl and Ethyl Esters

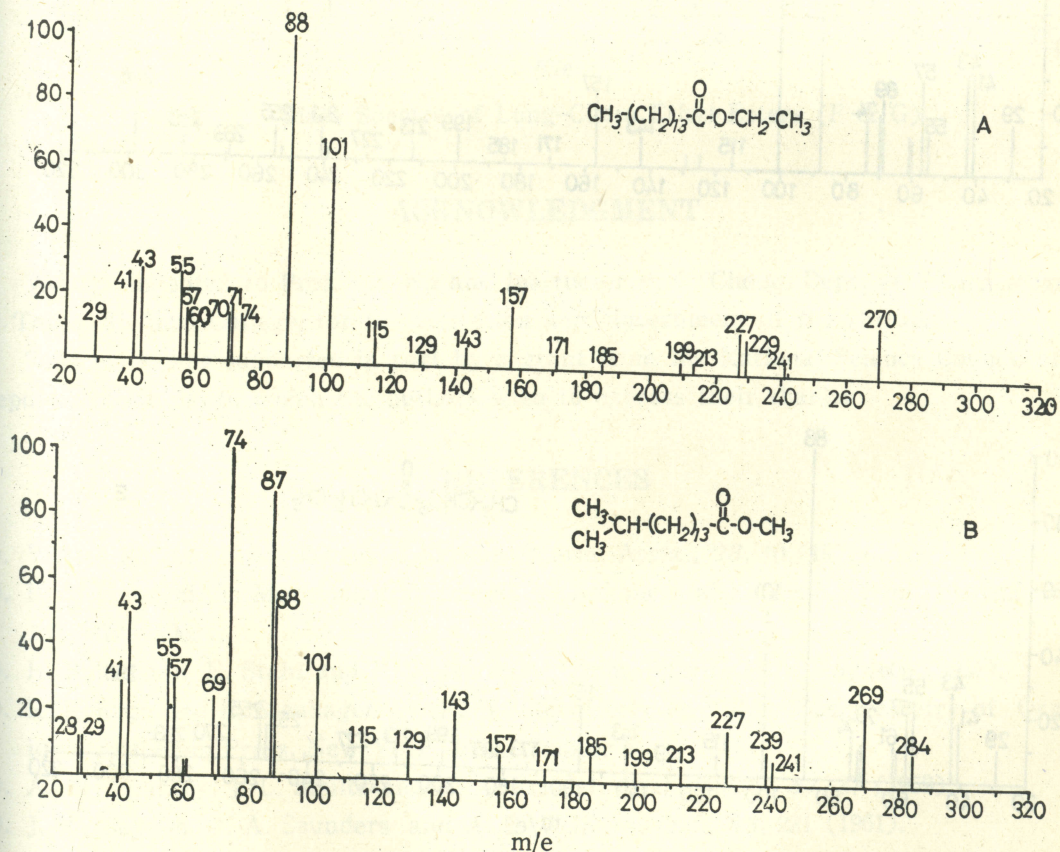


Fig. II. Mass Spectra of Long-Chain Methyl and Ethyl Esters (A & B)

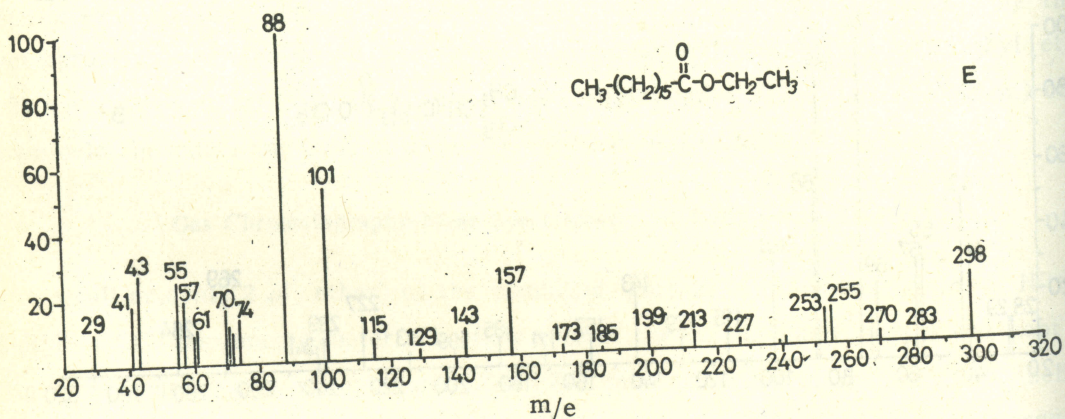
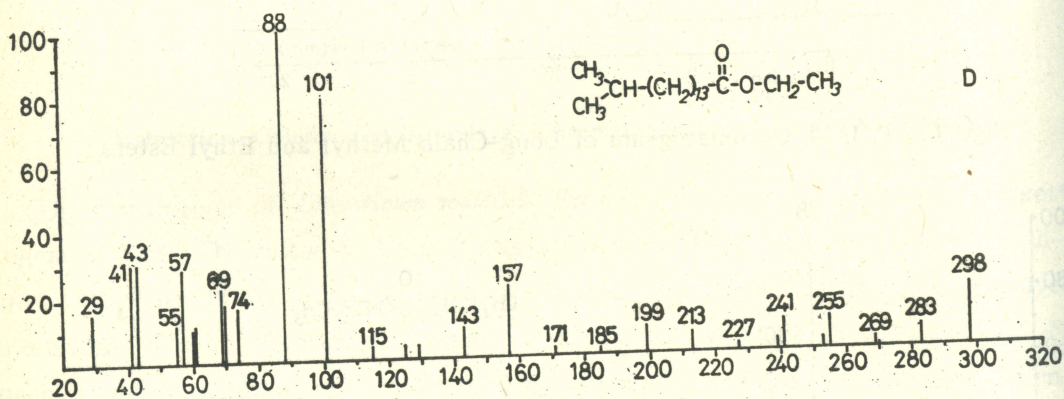
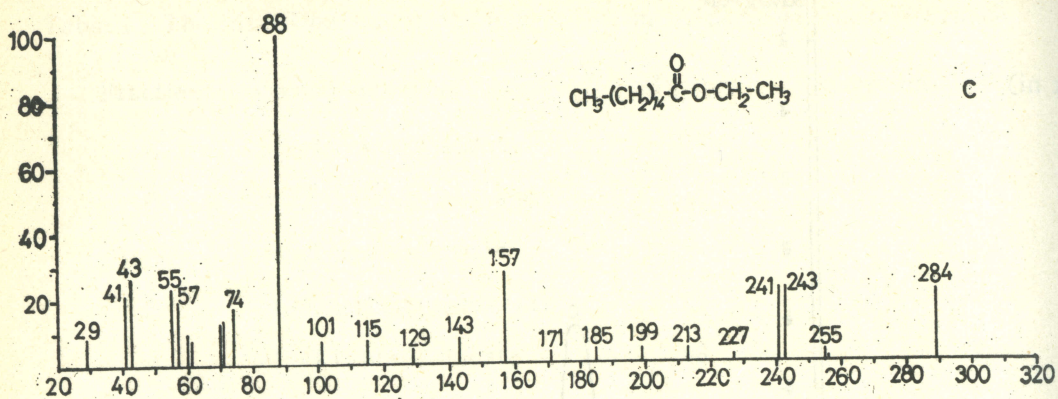


Fig. III. Mass Spectra of Long-Chain Ethyl Esters (C~E)

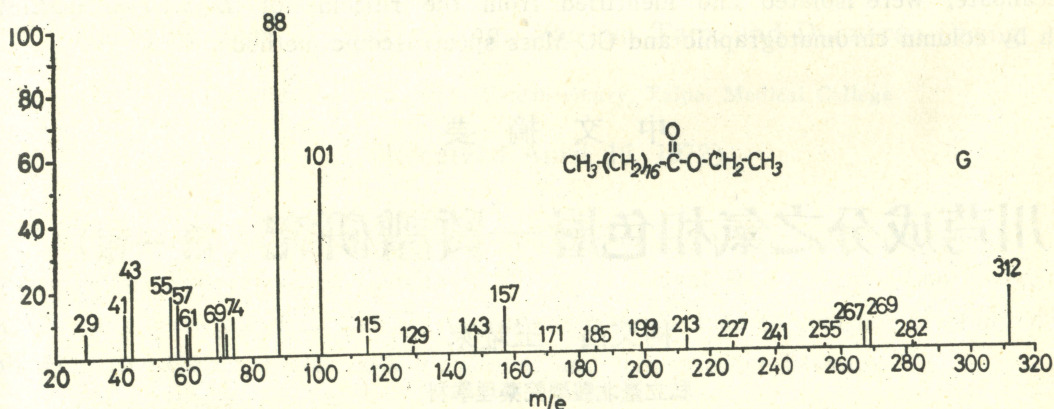
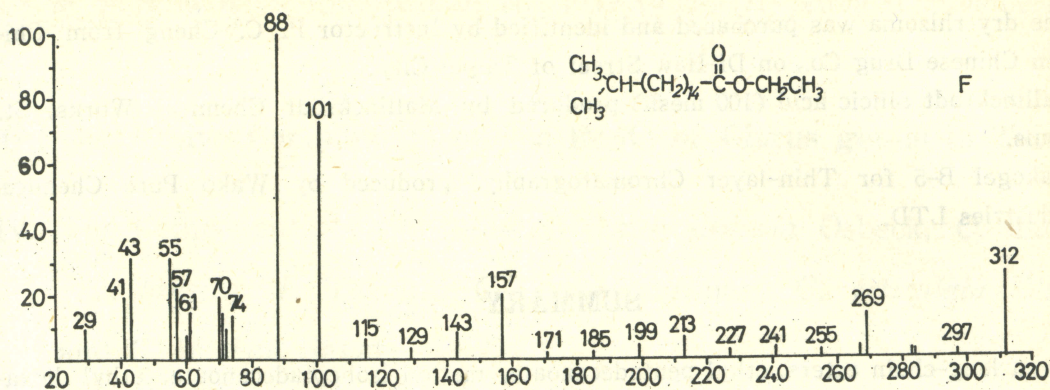


Fig. VI. Mass Spectra of Long-Chain Ethyl Esters (F & G)

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- 6). J. H., Beynon, R. A. Saunders and A. E. Williams, *Ibid.*, **33**, 221 (1961).
- 7). R. Ryhage and E. Stenhagen, *Arkiv. Kemi.*, **14**, 483 (1959).
- 8). R. Ryhage and E. Stenhagen, *Ibid.*, **13**, 523 (1959).

- *1. The dry rhizoma was purchased and identified by Instructor P. C. Cheng from Chi-Yen Chinese Drug Co., on Di-Haw Street of Taipei City.
- *2. Mallinckrodt Silicic acid (100 mesh) produced by Mallinckrodt Chemical Works, St. Louis.
- *3. Wakogel B-5 for Thin-layer Chromatography, produced by Wako Pure Chemical Industries LTD.

SUMMARY

Seven long-chain esters, ethyl pentadecanoate, methyl isoheptadecanoate, ethyl hexadecanoate, ethyl isoheptadecanoate, ethyl heptadecanoate, ethyl iso-octadecanoate and ethyl octadecanoate, were isolated and identified from the rhizoma of *Ligusticum wallichii* Franch by column chromatographic and GC-Mass spectroscopic methods.

中文摘要

川芎成分之氣相色層—質譜研究 (第一報)

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由川芎 (*Ligusticum wallichii* Franch) 之根莖，經柱層分析及氣相色層—質譜分析，確認其中性油含有七種長鏈酯 (long-chain esters) 為 ethyl pentadecanoate, methyl isoheptadecanoate, ethyl hexadecanoate, ethyl isoheptadecanoate, ethyl heptadecanoate, ethyl iso-octadecanoate 及 ethyl octadecanoate.