Pakistan Journal of Pharmaceutical Sciences 33
3(2) 33-37 (1990)

FATTY ACIDS *OF INULA GRANTIOIDES*

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ABSTRACT

The flowers *of Inula grantioides* collected from the vicinity of University of Sindh, Jamshoro Campus, Pakistan were analysed for their fatty acid composition. The fatty acids were converted into methyl esters and identified by gas liquid chromatography-mass spectrometry as myristic, pentadecanoic, palmitic, margaric stearic, arachidie behenic, tricosoic, lignoceric, hexadecanoic and heptacosoic acid.

Introduction

The plants of the genus *Inula* (compositae) have been the subject of interest from phytochemical point of view. It is reported (Ahmed and Chughtai, 1961 and Kirtikar and Basu, 1933) that the oil obtained from the plant *Inula grantioides* has antibiotic properties. The plant has been used by locals in Lasbela as specific treatment for the patients suffering from asthma. The plant is locally known as "Naro" and "Kolmur". It is widely distributed on small hillocks in Sind province especially in Hyderabad and Karachi regions; Ziarat in Baluchistan province and Waziristan in North West Frontier Province.

Other plants of this genus are reported to possess bactericidal (Batlanovi 1949 and Ya-Rashba *et al.,* 1954), toxic (Go, 1938 and Goryaev, 1948), and physiological properties (Chopra *et al,* 1945 and Eadie, 1953). Previous workers have reported (Harborne, 1975, Sethi *et al,* 1979, Kaur and Kalsi, 1985 and Ulubelin *et al*., 1987), the presence of terpenoids, sterols, flavonoids, alkaloids, lipids, polyacetylenes etc. from different species of this genus. The literature survey reveals that no work so far has been c*arried* out on the fatty acid composition of *Inula grantioides.* Therefore an attempt was made to isolate and identify the fatty acids present in this species.

Materials and Methods

Experimental

*Plant mater*i*al: Inula grantioides* flowers collected from the vicinity of Jamshoro (Sindh) during the spring of 1986; dried under shade and milled.

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Extraction: The dried and coarsely milled flowers (1.5 kg) were exhaustively extracted by percolation at room temperature with acetone in aspirator. The procedure was repeated three times. Evaporation of solvent from collective extract yielded 67g of thick syrupy brownish material.

Chromatographic separation: Acetone extract was eluted with gradual increase of polarity with various ratios of n-hexane-ethyl acetate on silica gel column using silica gel 60, 0.63-0.200 mm (70-230 mesh ASTM) Fluka. After passing 4L of eluent in the ratio of 4:1, the subsequent elution of 1.5L with the same ratio afforded fraction D (2.001 g). It was rechromatographed on a small scale column of silica by eluting it in a same manner as mentioned above with various ratios of the mixture of n-hexane-ethyl acetate to obtain subtraction I (0.322 g).

Esterification: Subfraction I obtained by n-hexane-ethyl acetate (9:1) treated with freshly preparaed diazomethane, and chromatographed on a miniscule column of silica. Two consecutive fractions X (0.053 g) and Y (0.056 g) of fatty acids as their methyl esters were obtained by elution of column again with n-hexane-ethyl acetate (9:1).

Identification: The unknown fatty acids fractions (methyl esters) were analysed by GC-MS. The GC-MS of the methylated fatty acid fractions was performed on a JEOL JMS H X 110 GC-mass spectrometer equipped with DEC 11/73 DATA SYSTEM and a 2m x 2 mm glass packed column coated with O V101. The column temperature was programmed between 70°C-240°C with the rate of increase 8°C per 2 minutes. The carrier gas (helium) flow rate was 30 ml/min.

Results

Spectral Data

i) Tetradecanoic methyl ester (Methyl myristate): GC-MS: m/z 242 (M^+, $C_{15}H_{30}O_2$), 211 (M^+-31), 199 (M^+-43), 185, 157, 143, 129, 101, 87, 74 (100%).

ii) Pentadecanoic methyl ester (Methyl pentadecylate): GC-MS : m/z 256 (M^+, $C_{16}H_{32}O_2$), 225 (M^+-31), 213 (M^+-43), 199, 185, 157, 143, 101, 87, 74 (100%).

iii) Hexadecanoic methyl ester (Methyl palmitate): GC-MS : m/z 270 (M^+ $C_{17}H_{34}O_2$), 239 (M^+-31), 227 (M^+-43), 199, 185, 171, 157, 143, 129, 101, 87 (100%), 74.

iv) Heptadecoic methyl ester (Methyl margarate): GC-MS : m/z 284 (M^+-$C_{18}H_{36}O_2$), 253 (M^+-31), 241 (M^+-43), 227, 185, 143, 129, 87, 74 (100%).

v) Octadecanoic methyl ester (Methyl stearate): GC-MS m/z 298 (M^+-$C_{19}H_{38}O_2$), 267 (M^+-31), 255 (M^+-43), 199, 185, 143, 129, 101i 87, 74 (100%).

vi) Eicosanoic methyl ester (Methyl arachidate): GC-MS : m/z 326 (M^+-$C_{21}H_{42}O_2$), 295 (M^+-31), 283 (M^+-43), 227, 199, 143, 129, 101, 87, 74 (100%).

vii) Docosanoic methyl ester (Methyl behenate): GC-MS m/z 354 (M^+, $C_{23}H_{46}O_2$), 323 (M^+-31), 311 (M^+-43), 297, 255, 213, 199, 157, 143, 129, 101, 87, 74 (100%).

viii) Tricosoic methyl ester (Methyl tricosoate): GC-MS m/z 368 (M^+, $C_{24}H_{48}O_2$), 337 (M^+-31), 325 (M^+-43), 311, 297, 255, 213, 199, 157, 143, 129, 101, 87, 74 (100%).

ix) Tetracosoic methyl ester (Methyl lignocerate): GC-MS m/z 382 (100%) (M^+, $C_{25}H_{50}O_2$), 351 (M^+-31), 339 (M^+-43), 325, 283, 199, 143, 87, 74.

x) Hexacosoic methyl ester (Methyl cerotate): GC-MS : m/z 410 (100%) (M^+, $C_{27}H_{54}O_2$), 379 (M^+-31), 367 (M^+-43), 283, 199, 143, 87, 74.

xi) Heptacosoic methyl ester (Methyl heptacosoate): GC-MS : m/z 424 (M^+, $C_{28}H_{56}O_2$), 393 (M^+-31), 381 (M^+-43), 367 (M^+-57), 353, 339, 383, 241, 199, 143, 87, 74 (100%).

Discussion

The GC-MS data of the methyl esters of fatty acids have been described in the experimental section. It revealed the presence of methyl myristate ($C_{15}H_{30}O_2$), methyl pentadecylate ($C_{16}H_{32}O_2$), methyl palmitate ($C_{17}H_{34}O_2$), methyl margarate ($C_{18}H_{36}O_2$), methyl stearate ($C_{19}H_{38}O_2$), methyl arachidate ($C_{21}H_{42}O_2$), methyl behenate ($C_{23}H_{46}O_2$), methyl tricosoate ($C_{24}H_{48}O_2$), methyl lignocerate ($C_{25}H_{50}O_2$), methyl cerotate ($C_{27}H_{54}O_2$) and methyl heptacosoate ($C_{28}H_{56}O_2$).

The identity of fatty acids was confirmed by matching of the mass spectra of the unknown compounds with those of the NBS mass spectra library (Helles and Milne 1978). These are all common components of triglycerides. The confirmation of the identity of eleven saturated fatty acid methyl esters was carried out by direct comparison with the MS data and their fragmentation pattern of the authentic samples. The relative percentage of occurrence of these identified fatty acid methyl esters is indicated in Table-1. An appreciable amount of fatty acids has been reported from other species of Inula like *Inula grandis* (Harborne, 1975 and *Inula cappa*, Singha, 1983). However the fatty acids from *Inula grantioides is* reported herewith for the first time.

Table 1

Fatty acids of *Inula grantioides* analysed as methyl esters

	Systematic name	Common name	Mol. Formula	R.R.T*	Rel. % age
Fraction X					
1	Methyl hexadecanoate	Methyl palmitate	$C_{17}H_{34}O_2$	1.00	35.471
2.	Methyl octadecanoate	Methyl stearate	$C_{19}H_{38}O_2$	1.27	13.965
3.	Methyl eicosoate	Methyl arachidate	$C_{21}H_{42}O_2$	1.52	9.496
4.	Methyl decosanoate	Methyl behenate	$C_{23}H_{46}O_2$	1.80	10.334
5.	Methyl tricosoate		$C_{24}H_{48}O_2$	1.97	3.631
6.	Methyl tetracosoate	Methyl lignocerate	$C_{25}H_{50}O_2$	2.19	9.776
7.	Unidentified	–	–	2.34	3.910
8.	Unidentified	–	–	2.64	7.820
9.	Unidentified	–	–	2.68	3.072
10.	Methyl hexacosoate	Methyl cerotate	$C_{27}H_{54}O_2$	2.82	2.513
Fraction Y					
1.	Methyl tetradecanoate	Methyl myristate	$C_{15}H_{30}O_2$	1.08	4.611
2.	Methyl pentadecanoate	Methyl pentadecylate	$C_{16}H_{32}O_2$	1.20	5.673
3.	Methyl hexadecanoate	Methyl palmitate	$C_{17}H_{34}O_2$	1.30	45.034
4.	Methyl heptadecoate	Methyl margarate	$C_{18}H_{36}O_2$	1.38	5.673
5.	Methyl octadecanoate	Methyl stearate	$C_{19}H_{38}O_2$	1.47	18.439
6.	Methyl eicosoate	Methyl arachidate	$C_{21}H_{42}O_2$	1.62	8.155
7.	Methyl docosanoate	Methyl behenate	$C_{23}H_{46}O_2$	1.84	6.382
8.	Methyl tricosoate	–	$C_{24}H_{48}O_2$		
9.	Methyl heptacosoate	–	$C_{28}H_{56}O_2$	2.22	6.028

*RRT = Relative retention time with respect to methyl palmitate of fraction X.

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