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# LIPIDS OF THE SEEDS AND FLESH OF THE FRUIT OF *Mandragora turcomanica*

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*The lipid compositions of the seeds and the flesh of the fruit of* Mandragora turcomanica *have been investigated. The qualitative identity of the lipid compositions of the two parts of the fruit has been shown and it has been found that the degrees of saturation of the fatty acids of the triacylglycerides and phospholipids and of the free fatty acids of the fruit flesh are higher than those of the lipid classes of the seeds.*

We have previously investigated the composition of the lipids [1] and the structure of the triacylglycerides [2] of the tuberous roots of the mandrake *Mandragora turcomanica* Mizger. (fam. Solanaceae). Continuing the study of the lipids of this plant, we have made a comparative analysis of the lipids of the seeds (I) and of the flesh with peel (II) of the ripe fruit. The seeds made up 13% of the crude weight of the fruit.

The neutral lipids (NLs) of (I) and (II) were extracted repeatedly with hexane, after which the total polar lipids (PLs) were isolated with chloroform—methanol (2:1, v/v) according to Folch. The PLs, contaminated with a small amount of NLs, were separated with the aid of CC into NLs, glyco-GLs, and phospholipids (PhLs). The NLs eluted from the column were combined with the hexane extract. The components of the NLs and the GLs were obtained by the use of CC in combination with preparative TLC on silica gel, and were estimated gravimetrically. The PhLs were identified qualitatively. The assignment of the components of the three groups was made on the basis of their chromatographic and spectral characteristics in comparison with known substances and with the aid of qualitative reactions and chemical transformations. The composition of the fatty acids (FAs) of the main acyl-containing lipids and that of the free fatty acids were established by the GLC of their methyl esters. Alkaloids were present among the nonlipid components of the PLs of the fruit. The amounts and compositions of the NLs, GLs, and PhLs of (I) and (II) are shown in Table 1, and the fatty acid compositions of lipids (I) and (II) in Tables 2 and 3.

The yields of total lipids from the seeds and flesh of the fruit amounted to 221.02 and 19.95 mg/g a.d.w, respectively. In spite of the large difference in their amounts, the main components of both samples were NLs with a predominance of triacylglycerides. The second components of the NLs quantitatively were fatty acid esters with cyclic and aliphatic alcohols (FAEs) and, in the flesh, free fatty acids (FFAs). We must mention the presence of monoacylglycerols and diacylglycerols in the lipids of both samples, their proportion in the lipids of the flesh being higher than in the seeds and the tuberous roots [1].

The level of polar lipids in the seeds of *M. turcomanica* was the same as in the tuberous roots [1] but far smaller than in the fruit flesh. Of the two groups of polar lipids, the GLs predominated in the flesh, and the PhLs in the seeds. The qualitative compositions of the GLs in the samples studied and in the tuberous roots were the same, but substantial differences were observed in the quantitative levels of individual components. In the GLs of the tuberous roots, monogalactosyldiacylglycerols and digalactosyldiacylglycerols predominated, while in the seeds these galactolipids were present in only trace amounts. The main GLS of the seeds and the flesh were steryl glycosides, but in them the proportion of monogalactosyldiacylglycerols was 5 times greater than that of digalactosyldiacylglycerols. In addition to what has been said above, the fruit flesh contained a GL giving the qualitative reaction for a sugar with α-naphthol and having a chromatographic mobility close to that of cerebrosides [3]. However, because of the insignificant amount of this GL it could not be definitively identified. By TLC, as the main components of the PhLs we detected phosphatidylcholines, phosphatidylethanolamines, and phosphatidylinositols, while phosphatidylserines and phosphatidic acids were present in minor amounts.

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TABLE 1. Compositions of the Lipids of the Seeds and Fruit Flesh of *M. turcomanica*

| Class of lipids | Seeds | Flesh | Seeds | Flesh |
|---|---|---|---|---|
| | mg/g a.d.w. tissue | | % of the weight of the lipids | |
| Neutral lipids: | 210.90 | 18.30 | 95.4 | 91.8 |
| 1. Hydrocarbons | 0.38 | 0.2 9 | 0.2 | 1.4 |
| 2. Fatty acid esters with alkanols and sterols + methyl esters | 6.50 | 0.70 | 3.0 | 3.5 |
| 3. Triacylglycerols | 188.24 | 12.86 | 85.1 | 64.5 |
| 4. Higher alcohols | 4.36 | 0.37 | 2.0 | 1.9 |
| 5. Free fatty acids | 2.97 | 2.10 | 1.3 | 10.5 |
| 6. Diacylglycerols | 4.18 | 0.55 | 1.9 | 2.8 |
| 7. Sterols | 3.06 | 0.72 | 1.4 | 3.6 |
| 8. Monoacylglycerols | 1.11 | 0.71 | 0.5 | 3.6 |
| Glycolipids: | 3.25 | 0.98 | 1.5 | 4.9 |
| 9. Esters of steryl glycosides | 0.64 | 0.01 | 0.2 | 0.1 |
| 10. Monogalactosyldiacylglycerols | 0.01 | 0.28 | 0.1 | 1.5 |
| 11. Steryl glycosides | 2.59 | 0.52 | 1.1 | 2.5 |
| 12. Unidentified | - | 0.11 | - | 0.5 |
| 13. Digalactosyldiacylglycerols | 0.01 | 0.06 | 0.1 | 0.3 |
| Phospholipids: | 6.87 | 0.67 | 3.1 | 3.3 |
| 14—17. Phosphatidylethanolamines, phosphatidylcholines, phosphatidylinositols, phosphatidic acids | | | | |
| Σ lipids | 221.02 | 19.95 | 100 | 100 |

TABLE 2. Fatty Acid Compositions of the Acyl-Containing Lipids of *M. turcomanica* Seeds

| Acid | Triacylglycerols | Free fatty acids | Esters of steryl glycosides | Phospholipids |
|---|---|---|---|---|
| 14:0 | Tr. | Tr. | 0.7 | 0.8 |
| 16:0 | 6.8 | 6.9 | 50.2 | 31.4 |
| 16:1 | 0.5 | 0.3 | 3.7 | 0.6 |
| 18:0 | 1.8 | 0.8 | 8.0 | 3.3 |
| 18:1 | 26.4 | 17.6 | 22.7 | 25.8 |
| 18:2 | 64.5 | 74.4 | 14.7 | 38.1 |
| 18:3 | Tr. | - | Tr. | Tr. |
| 20:0 | Tr. | - | - | - |
| $\Sigma_{sat}$ | 8.6 | 7.7 | 58.9 | 35.5 |
| $\Sigma_{unsat}$ | 91.4 | 92.3 | 41.1 | 64.5 |

The qualitative fatty acid compositions of the acyl-containing classes of the seed lipids were practically identical (Table 2). Unsaturated acids predominated in the TAGs and FFAs. Both in the TAGs and FFAs and in the PhLs the proportion of the 18:2 acid exceeded that of the 18:1 acid; the esters of steryl glycosides were enriched with the 16:0 acid, and the amount of the 18:1 acid was 1.5 times greater than that of the 18:2 acid.

It can be seen from the fatty acid composition of the fruit flesh lipids (Table 3) that the TAGs, FFAs, and monogalactosyldiacylglycerols were the most unsaturated of the lipid classes, with a higher proportion of the 18:2 and 18:1 acids. As compared with the TAGs, the FFAs contained more of the 16:0 and 20:0 acids, which is characteristic for the components of surface lipids [4].

The esters of fatty acids with alkanols, triterpenols, and sterols were enriched with the 16:0, 18:0, and 20:0 saturated acids. Among the polar lipids, the monogalactosyldiacylglycerols and digalactosyldiacylglycerols had close fatty acid compositions, with identical levels of the 18:1 and 18:2 species, while, in the PhLs, the 18:1 acid predominated.

TABLE 3. Fatty Acid Compositions of the Acyl-Containing Lipids of *M. turcomanica* Fruit Flesh

| Acids | Triacylglycerols | Free acids | Esters of acids and alcohols | Monogalactosyl-diacylglycerols | Digalactosyl-diacylglycerols | Phospholipids |
|---|---|---|---|---|---|---|
| 12:0 | Tr. | Tr. | 1.5 | - | - | Tr. |
| 14:0 | Tr. | 0.6 | 4.0 | 0.5 | 1.7 | 2.0 |
| 15:0 | - | 0.4 | 1.8 | Tr. | Tr. | Tr. |
| 16:0 | 15.2 | 23.4 | 38.2 | 37.7 | 41.4 | 37.2 |
| 16:1 | 0.6 | 0.4 | 4.5 | 3.3 | 5.7 | 0.9 |
| 17:0 | - | 1.5 | 1.6 | Tr. | Tr. | Tr. |
| 18:0 | 4.7 | 5.0 | 18.5 | 6.0 | 9.9 | 6.9 |
| 18:1 | 35.7 | 27.1 | 11.1 | 21.2 | 18.3 | 44.3 |
| 18:2 | 42.5 | 32.1 | 7.1 | 22.4 | 19.2 | 7.3 |
| 18:3 | 1.3 | Tr. | Tr. | 8.9 | 3.8 | 1.4 |
| 20:0 | Tr. | 9.5 | 10.5 | - | - | - |
| 22:0 | - | - | 1.2 | - | - | - |
| $\Sigma_{sat}$ | 19.9 | 40.4 | 77.3 | 44.2 | 53.0 | 46.1 |
| $\Sigma_{unsat}$ | 80.1 | 59.6 | 22.7 | 55.8 | 47.0 | 53.9 |

It follows from the results obtained that the degree of unsaturation of the fatty acids of the TAGs and PhLs and that of the FFAs of the flesh were higher than those of the lipid classes of the seeds and of the tuberous roots of *M. turcomanica*, studied previously [1].

The FAEs of the flesh were studied on the basis of their mass spectrum, starting from the fragmentation of acylated triterpenols, sterols [5], and wax esters [6], and also in the light of their fatty acid composition (Table 3) and their alcohol composition. In the alcohols obtained from the FAEs, GLC [1] showed the presence of (%): $C_{20}H_{41}OH$ (4.5), $C_{22}H_{45}OH$ (9.6), $C_{23}H_{47}OH$ (4.8), $C_{24}H_{49}OH$ (20.4), $C_{25}H_{51}OH$ (7.3), $C_{26}H_{53}OH$ (14.8), $C_{27}H_{55}OH$ (2.0), $C_{28}H_{57}OH$ (5.1), $C_{30}H_{61}OH$ (0.1), cholesterol together with an unidentified component (2.8), campesterol and lophenol (13.2), stigmasterol (0.1), β-sitosterol (10.8) and cycloartanol (4.5). Esters of the 16:0, 18:0, 18:1, and 18:2 acids with cycloartanol, lophenol, and campesterol, esters of saturated acids of the 14:0—20:0 even series and of the unsaturated acids 18:1 and 18:2 with β-sitosterol, and esters of the 16:0 and 18:0 acids with stigmasterol were identified. Wax esters were represented by saturated homologs of the even series with chain lengths from $C_{38}$ to $C_{48}$, esters of the 18:1 acid with the alkanols $C_{20\text{-}30}H_{41\text{-}61}OH$, and esters of the 18:2 acid with $C_{28}H_{57}OH$ and $C_{30}H_{61}OH$.

According to TLC on silica gel in system 1, the free sterols of both samples consisted of triterpenols ($R_f$ 0.40), 4-monomethylsterols ($R_f$ 0.33), and demethylsterols ($R_f$ 0.26). From the mass spectra of the native sterols and the $Ag^+$-TLC in system 2 of their acetates, the following were identified in both samples: cycloartanol ($R_f$ 1.33), cycloartenol ($R_f$ 0.56), lophenol ($R_f$ 1.18), β-sitosterol, stigmasterol, campesterol, and cholesterol ($R_f$ 1.0). These free triterpenols and sterols have been detected before in the seeds of plants of the Solanaceae family [7—9]. We have identified the above-mentioned sterols and triterpenols, including lophenol, in the free form and as esters in the tuberous roots of *M. turcomanica* previously [1].

## EXPERIMENTAL

For **general observations**, see [1].

Mass spectra were taken on a MKh-1321 instrument at an ionization energy of the electrons of 60/70 eV, the temperature of the ionization chamber being 150/170°C.

Analytical TLC was conducted on silica gel L 5/40 (Czech Republic) with the addition of 10% of $CaSO_4$, and $Ag^+$-TLC with 20% of $AgNO_3$. The following solvent systems were used: 1) $n$-$C_6H_{14}$—$(C_2H_5)_2O$—$CH_3COOH$ (70:30:1); and 2) $CH_2Cl_2$—$CCl_4$ (1:5, fourfold ascent).

*M. turcomanica* fruit was obtained from Yu. M. Murdakhaev of the laboratory of medical botany, F. N. Rusanov Botanical Garden, Academy of Sciences of the Republic of Uzbekistan.

The lipids were extracted from the air-dry seeds and flesh by Folch's method, while ballast substances were eliminated and the fatty acids were isolated from the acyl-containing lipids according to [11]. The fatty acids were esterified with diazomethane.

**Esters of Fatty Acids with Triterpenols, Sterols, and Alkanols in the Fruit Flesh.** Mass spectrum, *m/z*: $M^+$ 694, 692, 690, 666 and 411, 410, 288, 203 (cycloartanol esters); $M^+$ 708, 680, 678, 676, 652, 624 and 567, 539, 537, 535, 511, 483, 397, 396 (β-sitosterol esters); $M^+$ 664, 662, 638 and 287, 269, 245, 243, 227 (lophenol esters), 383, 382 (campesterol esters), 635, 607, 566, 538, 395, 394 (stigmasterol esters), 369, 368 (cholesterol esters) and others [5]; $M^+$ 704, 702, 700, 676, 674, 672, 648, 646, 620, 618, 592, 590, 564, 562 and 465, 437, 409, 381, 353, 325, 420, 392, 364, 336, 308, 280 (esters of the alkanols $C_{20\text{-}30}H_{41\text{-}61}OH$).

**Sterols of the Seeds.** Mass spectrum, *m/z* (*I*, %): $M^+$ 428 (6), 413, 410, 395, 367, 341, 288 (cycloartanol); $M^+$ 426 (3), 411, 408, 393, 365, 339, 286 (cycloartenol), 175, 69 (100) [8]; $M^+$ 414 (5), 396 (β-sitosterol); $M^+$ 412 (5), 394 (stigmasterol); $M^+$ 386 (7), 368 (cholesterol); $M^+$ 400 (5), 382 (campesterol), 385, 287, 269, 260, 245, 243, 227 (lophenol) and others [7, 10].

**Sterols of the Fruit Flesh.** Mass spectrum, *m/z* (I, %): $M^+$ 428 (6), 426 (3), 414 (33), 412 (25), 400 (33), 386 (3), 69 (100) and others [7-9].

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