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# CONJUGATED FATTY ACIDS FROM LATEX OF *EUPHORBIA LATHYRIS*

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**Key Word Index**—*Euphorbia lathyris*; Euphorbiaceae; caper spurge; conjugated fatty acids; diterpenes; triterpenes; esters; latex.

**Abstract**—The fatty acids esterified with triterpene and diterpene alcohols from the latex of *Euphorbia lathyris* were analyzed. 77% of the fatty acids esterified with triterpenes were conjugated, the main components of which were decadienoic and decatrienoic acids. Of the acids esterified with diterpenes, 81% were decatrienoic and dodecatrienoic acids.

## INTRODUCTION

The major constituents of the latex of many *Euphorbia* species are triterpenes and their esters [1–4]. Monoesters and diesters are also found in the latex of *Euphorbia* species but they are only minor components [5–6]. Although the terpenes of the latex of *Euphorbia* species have been extensively investigated, much less attention has been given to the saponifiable part of the terpene esters. Triterpene esters in the latex from *E. pulcherrima* appeared to be esterified mainly with conjugated decatrienoic acids [7]. Diterpenes isolated from the latices of *E. tirucalli* [8–10] and *E. lathyris* [11] were also esterified with conjugated fatty acids. These acids have 8–14 carbon atoms in the chain which has 2–5 double bonds conjugated with the carboxylic group. So far, the geometrical configuration of the double bonds of these acids has not been fully characterized.

Recently, the triterpenes from the latex of *E. lathyris* have been analyzed [12]. In the present study the fatty

Table 1. Fatty acid composition of the triterpene and diterpene esters from the latex of *E. lathyris*

| Fatty acid* | % of fatty acids | | ECL values Apiezon-L | |
|---|---|---|---|---|
| | Triterpene fraction | Diterpene fraction | 150° | 200° |
| 1. 10:3 (2, 4, ?) | 1.8 | 2.9 | 9.80 | |
| 2. 10:2 (3*t*, 5*c* or 3*c*, 5*t*) | 3.4 | — | 10.52 | |
| 3. 10:2 (3*t*, 5*t*) | 3.5 | — | 10.80 | |
| 4. 10:2 (2*t*, 4*c*) | 4.5 | 3.0 | 10.97 | |
| 5. 10:2 (2*t*, 4*t*) | 15.6 | — | 11.42 | |
| 6. 10:3 (3, 5, 7) | 2.5 | 7.5 | 11.62 | |
| 7. 10:3 (2*c*, 4*t*, 6*c*) | 11.2 | 18.9 | 11.84 | |
| 8. 10:3 (2*c*, 4*t*, 6*t*) | 5.0 | 1.9 | 12.12 | |
| 9. 10:3 (2*t*, 4*t*, 6*c*) | 15.4 | 18.9 | 12.21 | |
| 10. 12:0 | 0.1 | — | 12.00 | 12.00 |
| 11. 12:3 | 0.8 | 5.2 | | 13.65 |
| 12. 12:3 (2*c*, 4*t*, 6*t*) | 4.3 | 3.3 | | 14.18 |
| 13. 12:3 (2*t*, 4*t*, 6*c*) | 7.6 | 25.0 | | 14.36 |
| 14. 12:4 | 1.5 | — | | 15.21 |
| 15. 14:0 | 0.9 | — | | 14.00 |
| 16. 16:0 | 16.3 | 2.4 | | 16.00 |
| 17. 18:2 | 3.1 | 10.6 | | 17.53 |
| 18. 18:1 | 2.4 | — | | 17.66 |
| 19. 18:0 | 0.1 | 0.4 | | 18.00 |

* Number of carbon atoms followed by number of double bonds. In brackets the position of the double bonds numbered from the carboxyl carbon. Abbreviations: ECL = equivalent chain length, *c* = *cis*, *t* = *trans*.

acids esterified with the triterpenes and diterpenes from the latex of this plant are analyzed.

## RESULTS AND DISCUSSION

Table 1 shows the composition of the fatty acid fraction derived from the terpene esters of the latex of *E. lathyris*. Only fatty acids having relative percentages higher than 0.1% are recorded. Nineteen fatty acids esterified with triterpenes were identified. Six of them were the common acids: lauric, myristic, palmitic, stearic, oleic and linoleic acid. Palmitic acid was the main fatty acid. Lauric acid occurred in a trace amount only. Octanoic and decanoic acid, although present in relatively high concentrations in the latex of *E. pulcherrima* [7], were absent in the latex of *E. lathyris*. The remaining fatty acids (77.1%) were identified as fatty acids with conjugated unsaturation. Hydrogenation converted all conjugated fatty acids to the corresponding saturated analogues, namely decanoic (81.6%) and dodecanoic acid (18.4%). The content of fatty acids esterified with triterpenes, as determined by GC, was 5–6 mg fatty acid/g fresh latex.

The most abundant fatty acid in the diterpene ester fraction was linoleic acid. Palmitic and stearic acid were present as minor components. 81.2% of the fatty acids in this fraction were conjugated. Hydrogenation of these acids yielded decanoic (61.3%) and dodecanoic acid (38.7%). The concentration of the fatty acids in the diterpene ester fraction was 0.2–0.3 mg/g fr. latex.

### $C_{10}$-*acids*

For compound **1** the mass spectrum revealed the presence of 10 carbon atoms and 3 double bonds in the chain. The UV spectrum was similar to that of a Me-2,4-decadienoate [13], indicating that the additional double bond was isolated. The amount of **1** isolated by GC was not sufficient for further identification. A decatrienoate having two double bonds conjugated with the carboxylic group and one isolated double bond has been reported in Bartlett pear essence by Creveling and Jennings [14]. On biochemical grounds they postulated the Δ7 position for the isolated double bond.

Compounds **2**–**5** were identified by GC–MS as Me decadienoates. UV spectra showed that the double bonds of **2** and **3** are not conjugated with the carboxylic group [15]. Oxidation with $KMnO_4$ yielded butanoic acid, indicating that the double bonds are located at the Δ3 and Δ5 positions. The IR spectrum of **2** was identical to that of a *cis–trans* or a *trans–cis* configuration not conjugated with a carboxylic group [15,16]. Compound **3** was identified by IR as the *trans–trans* isomer. An adduct was formed with maleic anhydride, providing additional evidence for a *trans–trans* diene linkage. Conjugated dienoic acids with the double bonds starting at the Δ3 position have not been isolated from plants before. The UV and IR spectra of compounds **4** and **5** were identical to those reported by Crombie [13] for Me deca-*trans*-2, *cis*-4-dienoate and Me deca-*trans*-2, *trans*-4-dienoate, respectively. From the latter a maleic anhydride adduct could be formed, which also indicates a *trans–trans* linkage. Compounds **6**–**9** were identical to the unsaturated $C_{10}$-acids isolated from *E. pulcherrima* [7]. The ECL values differed from those found before [7], because of ageing of the Apiezon-L column. Cartoni *et al.* [17] showed that *cis–trans* isomers were eluted before the corresponding *trans–cis* isomers from an Apiezon-L column. Therefore, it is very likely that **6** consists of deca-*trans*-3, *trans*-5, *cis*-7-trienoic acid.

### $C_{12}$-*acids*

The UV spectrum of compound **11** indicated that the double bonds were not conjugated with the carboxylic group and that the geometrical structure of the double bonds was *trans–trans–cis* or *cis–trans–trans* [18]. The spectral data of **12** and **13** were identical to those of **8** and **9**, respectively. Therefore, **12** must be Me dodeca-*cis*-2, *trans*-4, *trans*-6-trienoate, and **13** must be Me dodeca-*trans*-2, *trans*-4, *cis*-6-trienoate. The mass spectrum of compound **14** indicated the presence of 4 double bonds. UV [8] revealed that the probable structure of this compound is Me dodeca-2,4,6,8-tetraenoate.

The major conjugated fatty acids in the diterpene fraction were the decatrienoic and dodecatrienoic acids. Only one dienoic acid was identified in this fraction (Table 1). This is in contrast to the findings of Adolf and Hecker [11] who reported isolating a mixture of diterpene esters with $C_{10}$: 2Δ and 3Δ: and $C_{12}$: 2Δ and 3Δ (by MS) from the latex of *E. lathyris*. This latex, however, was collected from plants in their second year. Fürstenberger and Hecker [8,9] using NMR partly identified the geometrical structure of the conjugated diene and triene acids esterified with the diterpenes from the latex of *E. tirucalli*. All these acids had a *cis*-2, *trans*-4 structure. The configuration at the third double bond of the triene acids was not identified. These data indicate important differences in the conjugated acid composition of *Euphorbia* species and small differences within one species (*E. lathyris*). A comparison of the latex and the seed oil [11,18] of *E. lathyris* showed diterpene esters to be present in both parts, but diene and triene acids were isolated only from the latex. The seed oil contained a pentaene acid esterified with the diterpenes [11]. It would appear that triterpenes esterified with conjugated acids are restricted to the latex system. From the apolar neutral fraction of the seeds of *E. lathyris* only common acids were isolated [18], the chief one being oleic acid. These differences support the view that the latex system behaves as a metabolically autonomous system in the plant. Its role in plant metabolism as a whole is still obscure.

## EXPERIMENTAL

Latex was obtained by cutting the stems of 6-month-old seedlings of *E. lathyris* L. A voucher specimen has been deposited at the Institute for Systematic Botany of the University of Utrecht. Triterpene esters were extracted from the latex as described before [7] and diterpene esters were extracted according to ref. [5]. Saponification of the esters and derivatization procedures for the fatty acids were as described previously [7]. Equivalent chain length (ECL) values and mol ratios were determined by GC on a 10% Apiezon-L column at 150° or 200° isothermally. Free volatile fatty acids were determined on a 15% PPGS column at 120° isothermally. UV spectra were determined in cyclohexane. IR spectra were recorded in $CS_2$, and computerized GC–MS was carried out at 70 eV.

*Me deca*-2,4,?-*trienoate* (**1**). UV $\lambda_{max}$ nm: 256. GC–MS *m/e* (rel. int.): 180 ($M^+$) (18), 149 ($M^+ - 31$) (4), 121 (32), 91 (31), 79 (45), 77 (29), 59 ($CO_2CH_3^+$) (37), 43 [$C_3H_7^+$] (100).

*Me deca*-cis-3, trans-5-*dienoate* or *Me deca*-trans-3, cis-5-*dienoate* (**2**). IR $\nu_{max}$ $cm^{-1}$: 2960, 2932, 1744, 1166, 982, 951. UV $\lambda_{max}$ nm: 232. GC–MS *m/e* (rel. int.): 182 ($M^+$) (33), 150 (11), 108 (44), 84 (46), 79 (84), 67 (100), 59 (38), 41 (28).

*Me deca*-trans-3, trans-5-*dienoate* (**3**). IR $\nu_{max}$ $cm^{-1}$: 2960, 2934, 1745, 1260, 1200, 1170, 1140, 990. UV $\lambda_{max}$ nm: 230. GC–MS *m/e* (rel. int.): 182 ($M^+$) (34), 150 (15), 108 (44), 84 (50), 79 (78), 67 (100), 59 (34), 41 (21).

*Me deca*-trans-2, cis-4-*dienoate* (**4**) IR $\nu_{max}$ $cm^{-1}$: 2934, 1720, 1640, 1310, 1270, 1168, 1140, 1043, 1018, 992, 870. UV $\lambda_{max}$ nm: 261. GC–MS *m/e* (rel. int.): 182 ($M^+$) (25), 151 ($M^+ - 31$) (23), 111 (100), 81 (60), 67 (48), 59 (29), 55 (35), 41 (45).

*Me deca*-trans-2, trans-4-*dienoate* (**5**). IR $\nu_{max}$ $cm^{-1}$: 2958, 2925, 1722, 1645, 1615, 1262, 1245, 1140, 998. UV $\lambda_{max}$ nm: 257. GC–MS *m/e* (rel. int.): 182 ($M^+$) (26), 151 ($M^+ - 31$) (17), 111 (100), 81 (48), 67 (37), 59 (26), 55 (25), 41 (38).

*Me deca*-trans-3, trans-5, cis-7-*trienoate* (**6**). GC–MS *m/e* (rel. int.): 180 ($M^+$) (53), 149 ($M^+ - 31$) (18), 119 (57), 91 (89), 79 (100), 59 (40), 55 (54), 43 (82).

*Me deca*-cis-2, trans-4, cis-6-*trienoate* (**7**). GC–MS *m/e* (rel. int.): 180 ($M^+$) (15), 149 ($M^+ - 31$) (3), 121 (26), 91 (27), 79 (39), 77 (24), 59 (21), 43 (100).

*Me deca*-cis-2, trans-4, trans-6-*trienoate* (**8**). GC–MS *m/e* (rel. int.): 180 ($M^+$) (51), 149 ($M^+ - 31$) (25), 119 (35), 91 (78), 79 (100), 59 (44), 55 (31), 43 (79).

*Me deca*-trans-2, trans-4, cis-6-*trienoate* (**9**). GC–MS *m/e* (rel. int.): 180 ($M^+$) (55), 149 ($M^+ - 31$) (17), 119 (52), 91 (73), 79 (100), 59 (37), 55 (36), 43 (73).

*Me dodecatrienoate* (**11**). UV $\lambda_{max}$ nm (rel. int.): 261 (79), 270 (100), 280 (89). GC–MS *m/e* (rel. int.): 208 ($M^+$) (51), 177 ($M^+ - 31$) (12), 138 (37), 119 (51), 105 (65), 91 (85), 79 (100), 41 (33).

*Me dodeca*-cis-2, trans-4, trans-6-*trienoate* (**12**). UV $\lambda_{max}$ nm (rel. int.): 288 sh. (74), 298 (100), 308 (83). GC–MS *m/e* (rel. int.): 208 ($M^+$) (43), 177 ($M^+ - 31$) (14), 138 (36), 119 (41), 105 (36), 91 (63), 79 (100), 41 (31).

*Me dodeca*-trans-2, trans-4, cis-6-*trienoate* (**13**). UV $\lambda_{max}$ nm (rel. int.): 287 sh. (79), 295 (100), 306 (86). GC–MS *m/e* (rel. int.): 208 ($M^+$) (44), 177 ($M^+ - 31$) (13), 138 (36), 119 (43), 105 (37), 91 (67), 79 (100), 71 (42), 43 (46).

*Me dodecatetraenoate* (**14**). UV $\lambda_{max}$ nm: 345. GC–MS *m/e* (rel. int.): 206 ($M^+$) (53), 175 ($M^+ - 31$) (6), 147 (34), 131 (41), 117 (83), 105 (53), 91 (100), 41 (29).

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