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# POLYACETYLENES FROM *CHRYSANTHEMUM LEUCANTHEMUM*

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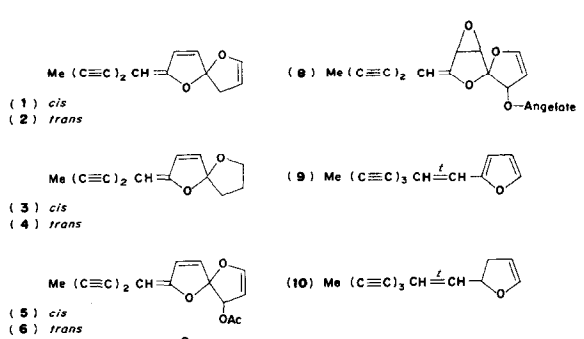
**Key Word Index**—*Chrysanthemum leucanthemum*; Compositae; acetylenes.

**Abstract**—Fifteen acetylenic compounds have been isolated and characterized from the flower heads of *Chrysanthemum leucanthemum* L. Five of these acetylenes have not previously been published as naturally occurring compounds. These are: 1,7(*c*)-hexadecadien-10,12,14-triyne; 1,8(*c*)-heptadecadien-11,13,15-triyne; 4(*c*)-tridecen-7,9,11-triyn-1-ol; 1,8(*t*)-hexadecadien-10,12,14-triyn-(6·7)-oxirane; and [3(*t*),5(*t*)-tridecadien-7,9,11-triyn-1-yl]-3-methyl-2-butenoate. The first three compounds are interesting since they are important intermediates in previously postulated biogenetic pathways. Ten acetylenes have been identified from the roots of the same plant, all of these having different structures from those of the flower heads. One of the acetylenes, [3(*t*), 5(*t*)-tridecadien-7,9,11-triyn-1-yl]-3-methylbutyrate, from the roots has never been reported as a naturally occurring compound.

## INTRODUCTION

Previous examination of roots and leaves of *Chrysanthemum leucanthemum* L. (*Leucanthemum vulgare* Lam.) have shown the presence of compounds **1–11** [1–5], whereas no datum on the flower heads has so far been published. Therefore, the acetylenic compounds present in the flower heads and roots of wild flowering plants collected in a small area during the middle of June were examined in order to study biogenetic and chemotaxonomical aspects further.




## RESULTS AND DISCUSSION

### *Acetylenes in flowers*

The light petroleum and ether extracts from the flower heads of *C. leucanthemum* were subjected to repeated column and preparative TLC. The acetylenic compounds separated and characterized are shown in Table 1. The hydrocarbon fraction proved to be difficult to separate. It was divided into two fractions by repeated preparative TLC on silica gel. The less polar fraction showed the presence of an ene-diyne chromophore ($\lambda_{max}$ 282, 267 and 253 nm). NMR-data were in good agreement with those published for an acetylene corresponding to (**17**) isolated from *C. maximum* Ramond [6]. However, the $\tau$-value of 6·01 reported earlier for the doublet corresponding to $-C{\equiv}C-CH_2-CH{=}CH$ has been corrected to 6·96 $\tau$ in a more recent publication [7] which is in complete agreement with our data.

While the NMR spectrum clearly shows a *cis*-configuration for the conjugated double bond $Me-CH{=}CH-C{\equiv}C-$, 8·1 $\tau$ (*dd*, $J = 7 + 2$, 3H) and 3·98 $\tau$ (*qd*, $J = 7 + 11$, 1H), the configuration of the isolated double bond is less clear. The NMR pattern at 4·5–4·7 $\tau$ forms a complex multiplet con-

Table 1. New polyacetylenes from flower heads of *Chrysanthemum leucanthemum**

(12) $Me(C{\equiv}C)_3CH_2CH{=}^{c}CH(CH_2)_4CH{=}CH_2$

(13) $Me(C{\equiv}C)_3CH_2CH{=}^{c}CH(CH_2)_5CH{=}CH_2$

(14) $Me(C{\equiv}C)_3CH{=}^{t}CH{-}CH{-}^{t}CH{-}(CH_2)_3CH{=}CH_2$ (epoxide)

(15) $Me(C{\equiv}C)_3(CH{=}^{t}CH{-}^{t}CH)_2CH_2CH_2O{-}CO{-}CH{=}C$(Me)(Me)

(16) $Me(C{\equiv}C)_3CH_2CH{=}^{c}CH{-}CH_2CH_2CH_2OH$

(17) $MeCH{=}^{c}CH(C{\equiv}C)_2CH_2CH{=}CH(CH_2)_5CH{=}CH_2$

(18) $Me(C{\equiv}C)_3(CH{=}^{t}CH{-}^{t}CH)_2(CH_2)_3CH{=}CH_2$

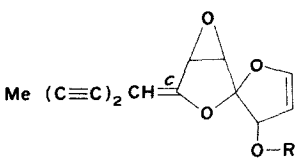
(19) $Me(C{\equiv}C)_3(CH{=}^{t}CH{-}^{t}CH)_2(CH_2)_4CH{=}CH_2$

(20) $Me(C{\equiv}C)_3CH{=}^{t}CH{-}COOCH_3$

(21) $Me(C{\equiv}C)_3CH{=}^{t}CH{-}CHO$

(22) $Me(C{\equiv}C)_3CH_2CH{=}^{t}CH{-}CH_2CH_2CH_2OAc$

(23) $CH_3(C{\equiv}C)_3(CH{=}CH)_2CH_2CH_2OH$



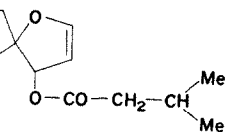
(24) R = —CO—$CH_2$—CH(Me)(Me)

(25) R = —CO—CH=C(Me)(Me)

* In addition (11) is also present.

Table 2. New polyacetylenes from roots of *Chrysanthemum leucanthemum**

(32) Me $(C{\equiv}C)_3(CH{=}^{t,t}CH)_2CH_2CH_2O{-}CO{-}CH_2CH$(Me)(Me)

(33) 

(34) 

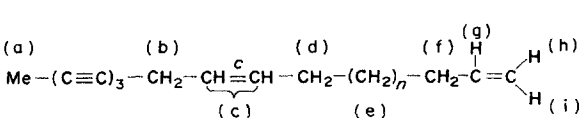
* In addition to (1), (2), (4), (5), (6), (9) and (10); previously reported to occur [1–5].

sisting of the signals from these double bond protons and one proton from the conjugated *cis*-double bond. The IR spectrum shows a band at 935 $cm^{-1}$ indicating a *trans*-configuration. Since similar acetylenic compounds with an isolated double bond usually exhibit a *cis*-configuration and as an artifact may have been formed, we cannot properly define the configuration on the basis of the present data. The MS in Table 3 show good agreement with data obtained earlier [8].

The second hydrocarbon fraction gave an UV spectrum corresponding to a diene-triyne chromophore ($\lambda_{max}$ 348, 325, 306, 290, 269, 259 and 254 nm). NMR data, however, indicated a mixture of several different compounds. Since preparative TLC gave no further separation, the fraction was subjected to column chromatography on Si gel mixed with 5% caffeine. The fractions of interest were then further separated by caffeine impregnated Si gel plates [9].

Light petroleum (bp $<50°$) was used for the development. Two fractions were obtained, one of which had a single, very intense band ($\lambda_{max}$ $<215$ nm) characteristic of a triyne chromophore. The presence of acetylenic bonds (2230 $cm^{-1}$) and a vinyl group (1640, 990 and 910 $cm^{-1}$) was evident from the IR spectrum.

The UV, IR and NMR-data strongly indicate the structures corresponding to (12) and (13) the assignments being shown in Fig. 1. Integration of the signal at 8·56 $\tau$ corresponded to six protons but this may be unreliable because of traces of solvent. The number of methylene groups was therefore determined from MS which showed two molecular peaks at *m/e* 210 and 224. GLC–MS gave two separated peaks whose MS (Table 3) were in good agreement with the proposed structures, (12) and



(12) $n=2$
(13) $n=3$

(a) 8·02 *s* (f) 7·94 *m*
(b) 6·98 *d* ($J=5·0$) (g) 4·25 *ddt* ($J=10+17·5+6·5$)
(c) 4·58 *m* (h) 5·09 *dm* ($J=10$)
(d) 7·94 *m* (i) 5·05 *dm* ($J=17·5$)
(e) 8·56 *m*

Fig. 1. NMR data for compounds 12 and 13.

Table 3. Mass spectral data for compounds (12)–(19), (22) and (32)

| (*m/e*) | Relative abundance (%)* | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | **(12)** | **(13)** | **(14)** | **(15)** | **(16)** | **(17)** | **(18)** | **(19)** | **(22)** | **(32)** |
| 41 | | | | 47 | 74 | 76 | | | 8 | |
| 43 | | | | 50 | 80 | | | | 100 | |
| 51 | | | | | 35 | | | | 8 | |
| 55 | | | 35 | 44 | 50 | 23 | | | | 20 |
| 57 | | | | 47 | | | | | | 89 |
| 63 | 30 | 15 | | | 35 | 22 | | | 9 | 13 |
| 65 | 17 | 10 | | | | | | | | |
| 67 | 28 | 22 | 69 | 12 | | 44 | | | | |
| 69 | | | | 34 | | | | | | 14 |
| 75 | 39 | 19 | | | 50 | | | | 13 | |
| 77 | 32 | 25 | | | | 32 | 21 | 47 | | |
| 79 | 31 | 20 | | | | 39 | | | | |
| 80 | | | | | | | | | 9 | |
| 81 | | | 61 | 16 | | | | | | |
| 83 | | | | 37 | | | | | | 6 |
| 85 | | | | 15 | | | | | | 20 |
| 87 | | | 41 | | | | | | | |
| 91 | 31 | 23 | | 8 | 51 | 58 | 11 | 36 | 9 | 9 |
| 95 | | | | 9 | | | | | | |
| 100 | | | 70 | | | | | | | |
| 101 | 17 | 12 | | | 30 | | 9 | 26 | 8 | |
| 103 | | | | | | 17 | | | | |
| 105 | | | | | | 19 | | | | |
| 114 | | | 86 | | | | | | | |
| 115 | 100 | 82 | 80 | 10 | 100 | 68 | 35 | 43 | 26 | 9 |
| 117 | | | | | | 37 | | | | |
| 127 | | | 43 | 7 | 35 | | 20 | 30 | 10 | 6 |
| 128 | 51 | 38 | | | 45 | 100 | 20 | 26 | 11 | |
| 129 | | | | | | 45 | | | | |
| 139 | 35 | 25 | | 10 | 45 | | 43 | 68 | 24 | 9 |
| 140 | 96 | 100 | | | | | 33 | 32 | 23 | |
| 141 | | | 46 | 8 | 43 | 31 | | | | |
| 142 | | | 42 | | | | | | | |
| 143 | | | | | | 40 | | | | |
| 152 | | | | 13 | 44 | | 100 | 98 | 29 | 14 |
| 153 | 55 | 37 | 37 | | 30 | | 82 | 72 | 24 | |
| 155 | | | | | | 12 | | | | |
| 156 | | | 34 | | | | | | | |
| 157 | | | | | | 13 | | | | |
| 165 | 32 | 24 | | 100 | 13 | | 87 | 100 | 22 | 100 |
| 166 | | | | 62 | | | | | 12 | 46 |
| 167 | 50 | 28 | | 10 | 8 | | | | 13 | 7 |
| 168 | | | | | | | | | 14 | |
| 169 | | | 100 | | | 12 | | | | |
| 178 | | | | | | | 47 | 70 | | |
| 179 | 9 | 10 | | | | | 43 | 89 | | |
| 181 | 20 | 21 | 29 | | | | | | | |
| 183 | | | 14 | 10 | | 6 | | | 3 | 8,5 |
| 184 | | | | | 3 | | | | 1 | |
| 185 | | | | | 5 | | | | 1 | |
| 186 | | | | | **4** | | | | | |
| 193 | 3 | 4 | | | | | 23 | 55 | | |
| 195 | 12 | 8 | 21 | | | | 15 | 26 | | |
| 197 | | | | | | 3,5 | | | | |

* Parent ion shown in bold type.

Table 3. Mass spectral data for compounds (12)–(19), (22) and (32)—*continued*.

| (*m/e*) | Relative abundance (%)* (12) | (13) | (14) | (15) | (16) | (17) | (18) | (19) | (22) | (32) |
|---|---|---|---|---|---|---|---|---|---|---|
| 207 | | | | | | | 10 | 66 | | |
| 208 | | | | | | | **26** | 20 | | |
| 209 | 5 | 5 | 6 | | | | | | | |
| 210 | **7** | 2 | | | | | | | | |
| 211 | | | | | | 3 | | | | |
| 222 | | | | | | | (2) | **45** | | |
| 223 | | | 9 | | | | | | | |
| 224 | | **5** | **8.5** | | | | | | | |
| 226 | | | | | | **3** | | | | |
| 228 | | | | | | | | | **1.5** | |
| 234 | | | | | | | | | | 3 |
| 238 | | | 2 | | | | | | | |
| 268 | | | | | | | | | | **0.1** |

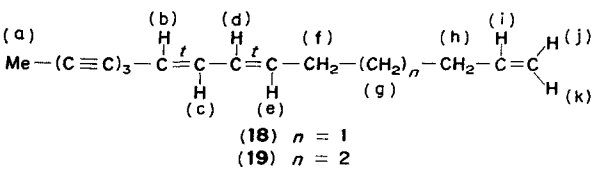
* Parent ion shown in bold type.

(**13**). Neither have previously been reported as naturally occurring, but their existence has been predicted on biogenetic grounds.

The final hydrocarbon fraction exhibited UV and IR data like those reported for synthetic centaur $X_3$ (**19**) [10]. Furthermore, the NMR data agree well with the structure shown in Fig. 2.

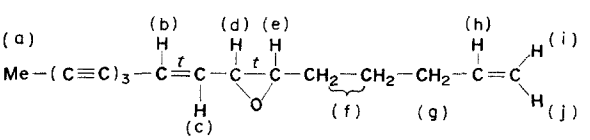
This fraction was again found to consist of two components with MWs 208 (**18**) (major) and 222 (**19**), respectively (see Table 3). The MS of (**19**) is in good agreement with centaur $X_3$ isolated from *Senecio jacobaea* [11]. While (**18**) is almost only present in those *Chrysanthemum* species which possess $C_{13}$-biogenesis, (**19**) is widely distributed in the Compositae [1].

Subsequent increase of polarity of the solvent gave more polar fractions: 2% $Et_2O$ in light petroleum gave two further acetylenes. The major one was identified as *trans*-dehydromatricaria ester (**20**) [1] by means of spectral data and chromatographic identity with an authentic sample. The second acetylene, (**14**), had an UV spectrum similar to that of an ene-triyne, but with a shift of 4–8 nm towards higher wavelengths, which is characteristic for an epoxide group $\alpha$ to the chromophore: [$\lambda_{max}$ 334 ($E_{rel.}$ 0·30), 312·5 (0·425), 293 (0·32), 276 (0·19), 250·5 (1·36) and 238 nm (1·11)]. Careful hydrolysis gave a diol with a characteristic ene-triyne chromophore ($\lambda_{max}$ 331, 310, 291, 274, 245·5, 235 nm). Treatment of this diol with $NaIO_4$ gave an aldehyde, which, by comparison of its UV and TLC data was identified as dehydromatricarianal (**21**). The infrared spectrum of (**14**) revealed –C≡C– (2210 $cm^{-1}$), epoxy (1225 and 880 $cm^{-1}$), –CH=$CH_2$ (1630, 990 and 910 $cm^{-1}$), *trans*–CH=CH– (940 $cm^{-1}$). The UV, IR supported by data obtained by NMR (Fig. 3) and MS (Table 3) suggest the structure of the naturally occurring compound as shown (**14**, Table 1). The




(a) 8·01 *s*
(b) 4·57 *d* (*J* = 15)
(c) 3·34 *dd* (*J* = 15 + 10)
(d) 3·92 *dd* (*J* = 10 + 14)
(e) 4·34 *dt* (*J* = 14 + 7·5)
(f) 7·9 *m*
(g) 8·5–8·6 *m*
(h) 7·9 *m*
(i) 4·3 *m*
(j) 5·09 *dm* (*J* = 10)
(k) 5·06 *dm* (*J* = 17)

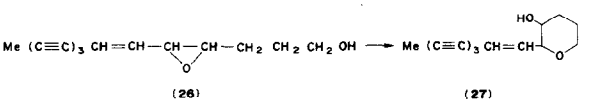
Fig. 2. NMR data for compounds **18** and **19**.




(a) 8·02 *s*
(b) 4·26 *d* (*J* = 15·75)
(c) 3·95 *dd* (*J* = 15·75 + 6·2)
(d) 7·01 *dd* (*J* = 6·2 + 2)
(e) 7·31 *dm* (*J* = 2)*
(f) 8·4–8·6 *m*
(g) 7·95 *m*
(h) 4·28 *ddt* (*J* = 17·5 + 10 + 6·5)
(i) 5·08 *dm* (*J* = 10)
(j) 5·02 *dm* (*J* = 17·5)

Fig. 3. NMR data for compound **14**. * The coupling constant of 2 cps indicates a *trans*-epoxide [12].

MS gives a molecular peak of 224 *m/e* indicating a compound with 16 carbon atoms although very weak peaks appear at 238 and 223 *m/e*, indicating the presence of minor amounts of the corresponding $C_{17}$-compound. This epoxide **14** has not previously been found in nature and seems to be biogenetically related to the hydrocarbon **28**. A related epoxide (**26**) is known from *C. serotinum* L. [13] where it is suggested to be an intermediate in the biogenesis of (**27**).

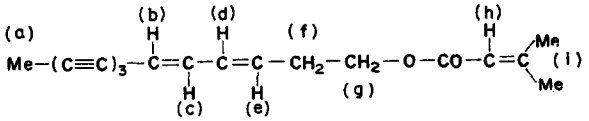


(26) (27)

With 20% $Et_2O$ in light petrol, four further acetylenes were eluted (**15**, **21**, **22** and **11**). The acetate (**11**) is the most abundant acetylene in the flower heads. It is characteristic of *Chrysanthemum* species with $C_{13}$-biogenesis and as mentioned previously was found in the roots of *C. leucanthemum* [2].

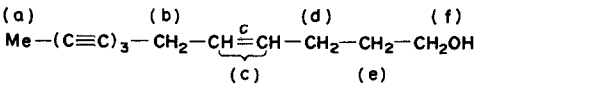
The triyne-acetate (**22**), a biogenetic precursor of (**11**), has recently been reported from *C. croaticum* Horwatie [1]. It was characterized by comparison of its spectral data with those of the synthetic product [14] (MS data see Table 3). Dehydromatricarianal (**21**) has been reported from *Lactuca plumieri* Gren. and Godr. (Cichorieae [15]) but is a rare compound and has not previously been found in Anthemideae.

Compound (**15**) had an UV spectrum ($\lambda_{max}$ 348, 325·5, 307, 269, 259 and 255 nm) similar to that of (**11**) but was less polar. Its IR spectrum indicated the presence of an $\alpha\beta$-unsaturated ester group (1720, 1230 and 1150 $cm^{-1}$), apart from –C≡C– (2230 $cm^{-1}$), *trans*–CH=CH– (1650, 975 $cm^{-1}$) and –CH=C< (860 $cm^{-1}$). These data considered together with MS (Table 3) and NMR (Fig. 4) suggest the structure (**15**). No peak corresponding to the molecular ion could be detected in the MS.



(a) 8·00 *s*
(b) 4·45 *d* (*J* = 16)
(c) 3·24 *dd* (*J* = 16 + 10)
(d) 3·80 *dd* (*J* = 10 + 15·5)
(e) 4·14 *dt* (*J* = 15·5 + 6·5)
(f) 7·52 *t* (*J* = 6·5 + 7)
(g) 5·84 *t* (*J* = 7)
(h) 4·32 *m*
(i) 7·84 *d* (*J* = 1), 8·10 *d* (*J* = 1)

Fig. 4. NMR data for compound **15**.



(a) 8·06 *s*
(b) 6·96 *d* (*J* = 5)
(c) 4·58 *m*
(d) 7·86 *t* (*J* = 7·5 + 6)
(e) 8·40 *tt* (*J* = 6 + 6)
(f) 6·42 *t*

Fig. 5. NMR data for compound **16**.

However, the fragmentation pattern is not inconsistent with structure (**15**). Fairly intense fragment peaks at 183 *m/e* and 166 *m/e* may correspond to loss of a senecionyl ion and senecionic acid, respectively. Furthermore, an intense peak at 83 *m/e* confirms the formation of the senecionyl ion. For confirmation (**15**) was synthesized from the alcohol (**23**) and senecionyl chloride. The synthetic product and the naturally occurring compound yielded identical spectral data (UV, IR, NMR and MS) and TLC. This ester has never been previously reported as a natural product.

With 40% $Et_2O$ content in light petroleum two spiro acetylenes appeared. Spectral data (UV, IR, NMR and MS) indicated that these were identical with (**24**) and (**25**) isolated from *C. monspeliense* L [16].

Finally, 80% $Et_2O$–light petrol eluented two acetylenic alcohols, (**16**) and (**23**). The latter alcohol corresponds to the ester (**11**) and has previously been isolated from various *Chrysanthemum* species [1]. The second alcohol had an UV spectrum indicating a triyne-chromophore ($\lambda_{max}$ <215 nm). IR-data show the presence of –C≡C– (2210 $cm^{-1}$), and a primary alcohol group (3350, 1050 $cm^{-1}$). The structure of (**16**) was finally confirmed on the basis of NMR (Fig. 5) and MS (Table 3) data. It has not previously been reported to occur naturally, but may be an important intermediate in the biogenetic pattern of $C_{13}$-acetylenes.

In addition to the compounds shown in Table 1 three ene-triyne and one ene-diyne acetylenes were detected by UV spectroscopy although they were present in amounts too small to permit further analysis. Four non-acetylenic hydrocarbons were also found. Three saturated hydrocarbons were isolated from the less polar fractions and identified by combined GLC–MS: *n*-dodecane (**28**), *n*-tetradecane (**29**) and *n*-hexadecane (**30**), but no trace of the odd numbered hydrocarbons, $C_{11}$, $C_{13}$, $C_{15}$ and $C_{17}$, was found. From another fraction the sesquiterpene $\beta$-farnesene [17] (**31**) was isolated and

was characterized by IR, NMR and MS spectral data.

*Acetylenes in roots*

The root extract was chromatographed as above and the isolated compounds were mainly identified by means of their spectral data. The new compounds found are given in Table 2. Seven acetylenes (**1**, **2**, **4**–**6**, **9** and **10**) have previously been isolated from the roots of *C. leucanthemum* by Bohlmann *et al.* [3, 4]. We did not, however, succeed in detecting compounds (**3**) [4], (**7**) [5], (**8**) [1], and (**11**) [2] in our root extracts.

Only one polyacetylene with a diene-triyne chromophore was detected. It was found to be the ester (**32**) (Table 2), with a polarity similar to that of the senecionate (**15**), but clearly different from that of the acetate (**11**). The IR spectrum was not very informative although it indicated $-C\equiv C-$ ($2220\,cm^{-1}$) and $(Me)_2CH-CH_2COOR$ (1745, 1120, $1100\,cm^{-1}$). On basis of these data and those obtained by NMR (Fig. 6) and MS (Table 3) we suggest that the structure is as shown (**32**). The MS of **32** shows, as expected, the same overall pattern as that of **15**, except for the fragmentation of the ester group. It has not been previously reported as a naturally occurring compound.

Two spiro compounds, the isovaleric acid ester (**33**) and the corresponding alcohol, (**34**), have not previously been shown to be present in *C. leucanthemum*, although (**33**) has been reported to occur in *C. monspeliense* L. [16] and *Santolina rosmarinifolia* L. [18], while the alcohol has been found in several *Chrysanthemum* species [1]. Spectral data correspond to those previously published, and the hydrolyzed product of the acetate (**5**) proved to be identical with (**34**) by TLC. In addition to these compounds shown in Table 2 an unsaturated hydrocarbon, which proved to be identical with β-farnesene (**31**), was isolated from the roots along with two coumarins. Spectral data and melting points indicated these latter compounds were scopoletin and isofraxidin. Isofraxidin is present in various *Artemisia* species [19]. Finally, two ene-triyne-acetylenes were isolated in too small amounts for full characterization.

The difference between the acetylenes found in roots and in flower heads is remarkable (Tables 1 and 2). None of the acetylenes isolated from the flower heads was found in the roots. The acetylenes prevailing in the flower heads are all acyclic and can be divided in four main groups, i.e. $C_{17}$-, $C_{16}$-, $C_{13}$- and $C_{10}$-compounds. Apart from the $C_{16}$-epoxide, the only cyclic substances observed in the flower heads are the spiro-epoxides (**24**) and (**25**). These are clearly related to the acetylenes found in the roots and they are probably late intermediates in biogenesis. The small amounts of spiro-compounds isolated from the flower heads suggest that they may have been formed in the stems or roots and translocated to the flower heads, or that they were present in the flower stalks.

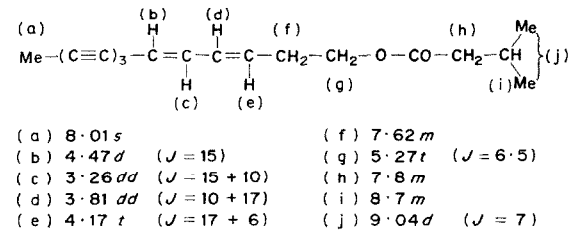



Fig. 6. NMR data for compound **32**.

The roots, however, apparently contain $C_{13}$-compounds only and, apart from a small amount of the diene-triyne ester (**32**), these compounds are all cyclic. In spite of structural differences, however, the acetylenes from the roots are closely related to the $C_{13}$-acetylenes from the flowers.

A possible outline of the biogenetic routes of acetylenes in *C. leucanthemum* is shown in Fig. 7. Various investigations emphasize that oleic acid may be the common metabolite from which all the acetylenes are derived [1]. By β-oxidation, β-hydroxyoleic acid is obtained as a possible precursor of the $C_{17}$-acetylenes [14, 20]. However, although (**17**), (**13**) and (**19**) are present in this plant, the $C_{17}$ route, unlike that in the majority of the Anthemideae, is less important than $C_{16}$-biogenesis. Here the main biogenetic route goes via the $C_{18}$-compound [(**35**), Fig. 7]. β-Oxidation, which is the most common reaction within the Anthemideae, leads to the $C_{14}$-series, but is not found in *C. leucanthemum*. Instead a successive α-oxidation (giving a $C_{17}$-compound) followed by β-oxidation gives compound (**36**).

The new acetylenic compound (**12**) is likely to be the first intermediate in $C_{16}$-biogenesis, presumably formed by decarboxylation and dehydration from (**36**), and oxidation, rearrangement and

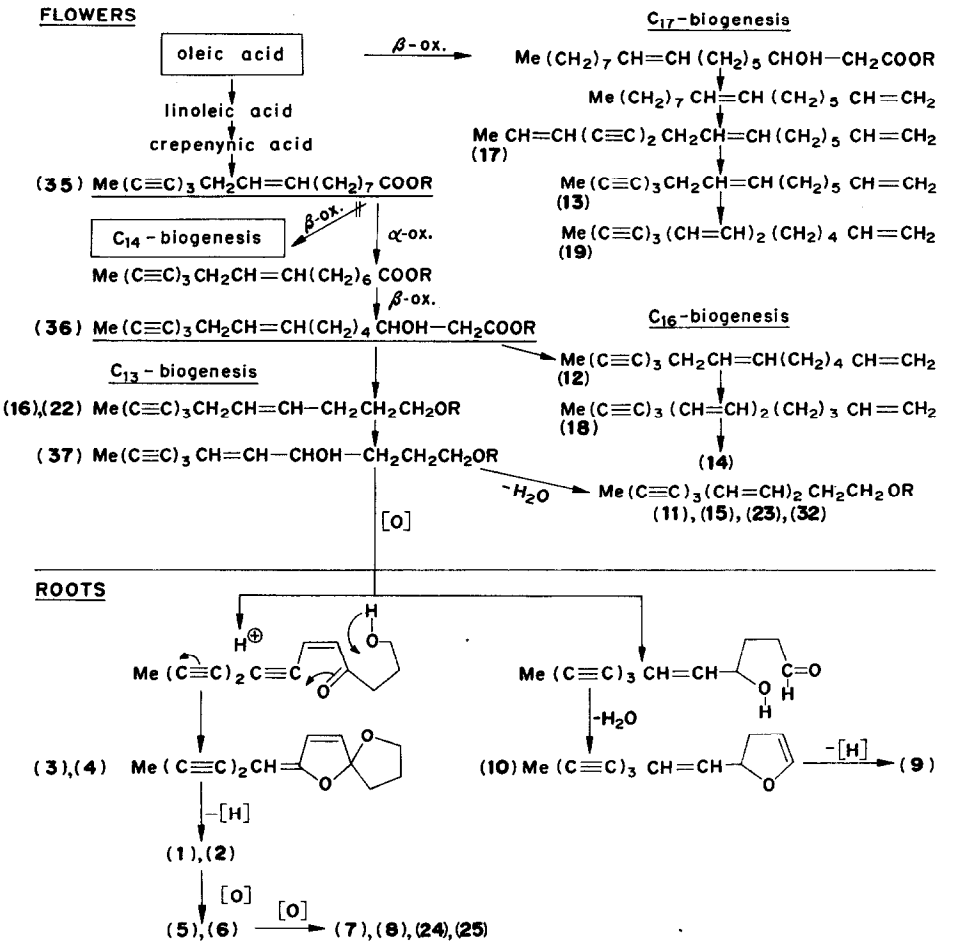



Fig. 7. Possible biogenetical pathways in *Chrysanthemum leucanthemum*.

dehydration of (**12**), analogous to $C_{13}$-biogenesis leads to (**18**) which, by oxidation, could be transformed into the epoxide **14**. It is still not clear, however, whether $C_{16}$-biogenesis follows the main biogenetic route to the triyne intermediate (**36**) or, as with $C_{17}$-biogenesis, starts with an α-oxidation at the oleic acid step. It should be noted that the $C_{16}$-compounds (**12**, **14** and **18**) have been isolated only from plants having the analogous $C_{13}$ compounds (**11** etc.) whereas the $C_{17}$-hydrocarbons are also known from plants without $C_{14}$-compounds. This suggests a closer relation between $C_{13}$- and the $C_{16}$-biogenesis than between the $C_{14}$- and the $C_{17}$-biogenesis.

A stronger indication of the proposed pathway is that while the $C_{16}$-hydrocarbons with triyne- and dienetriyne chromophores (**12** and **18**) and found in much greater amounts than the corresponding $C_{17}$ homologues (**13** and **19**), $C_{16}$-homologue corresponding to the hydrocarbon (**17**) with an ene-diyne chromophore were not detected.

The main route, however, precedes from (**36**) via a repeated β-oxidation to the $C_{13}$-compounds (**16**) and (**22**). Although the latter compound has only recently been found to occur naturally [1], and the corresponding alcohol (**16**) was not previously found, extensive investigation with labeled (**16**) and (**22**) has revealed their importance in the formation of $C_{13}$-compounds [14, 21, 22]. The isolation of both compounds from *C. leucanthemum*, a typical $C_{13}$-producing plant, thus offers good support for the biogenetic route shown in Fig. 7.

While in other *Chrysanthemum* species (**16**) may be transformed into aromatic compounds [14], here a rearrangement into the ene-triyne compound (**37**) presumably takes place. The biogenetic importance of this acetylene has been demonstrated by labelling experiments [14], Although no (**37**) was apparently present in our extracts, some fractions contained minute amounts of acetylenes with ene-triyne chromophores. It is suggested that (**37**) either may be formed in the aerial parts of the plant and dehydrated to compounds (**11**), (**15**) and (**23**), or it may be transported to the roots. In the roots it could undergo oxidation in either of two ways. One possibility is the oxidation of the primary alcohol group to an aldehyde, which may be cyclisized via a semiacetal and dehydration to the furan derivatives (**9**) and (**10**) [14]. The other possibility is oxidation of the alcohol group (in allylic position) to the corresponding ketone, which may cyclize to the spiro compounds (**3**) and (**4**) (Fig. 7) [14]. Dehydrogenation of the latter would lead to (**1**) and (**2**) [14, 22], which are presumed to be the precursors of other spiro compounds in the plant.

The biogenesis of the two $C_{10}$-compounds, dehydromatricariaester (**20**) and dehydromatricarianal (**21**), is not clear. The results of several experiments [14, 20, 23, 24] indicate that compounds with a Me–(C≡C)$_3$-group or $C_{18}$-compounds with a *cis*-double bond in the middle of the chain could be potential precursors of (**20**). It has been shown, however, that even compounds like Me(C≡C)$_3$(CH$_2$)$_n$COOH may be starting materials for the formation of dehydromatricariaester [24], although such compounds have not been found in the plants examined.

While $C_{13}$-biogenesis in Anthemideae thus seems to be clear from the triyne step onwards, it is not completely certain how compound (**36**) is formed from oleic acid, neither has the biogenesis leading to the $C_{16}$ and $C_{17}$ hydrocarbons been completely clarified.

## EXPERIMENTAL

Plants of *Chrysanthemum leucanthemum* L. were collected in Hasle, Aarhus, during mid-June 1971, a reference specimen being deposited in our laboratory. Plant material was divided into flower heads, green parts and roots (latter being washed and air dried). Each was ground and extracted, first with light petrol and then with $Et_2O$. On removal of solvents 0·7 kg of flower heads gave 6·9 g crude extract and 1·25 kg roots yielded 9·4 g crude extract. (Crude extract of green parts has not yet been fully examined.) Extracts were subjected to column chromatography [Si gel (Merck)], using light petrol (bp $<50°$) and light petrol containing increasing percentages of $Et_2O$ as eluting solvents. For further separation repeated preparative TLC applied. Hydrocarbon fractions were further separated by means of columns of 10% caffeine in Si gel or 5% for TLC.

Amounts of the isolated acetylenes were usually determined by UV. Amounts of compounds with triyne chromophores were, however, determined from the NMR-integrals as relative amounts of mixtures with dienetriyne chromophores before separation. Relative amounts of $C_{16}$ and $C_{17}$ hydrocarbons were determined by means of GLC after separation of triyne- and dienetriyne compounds on caffeine–Si gel. MS was with direct inlet and GLC–MS was used for some of the studies.

*Compounds isolated from the flower heads of* Chrysanthemum leucanthemum. 116 mg of *β*-farnesene, 11 mg of (**17**), 240 mg of (**12**), 90 mg of (**13**), 320 mg of (**18**) and 70 mg of (**19**) were eluted with light petrol. With 2% $Et_2O$ in light petrol: 1 mg of (**14**) and 40 mg of (**20**). With 5–20% $Et_2O$ and purified by TLC (by means of 5, 10 or 20% $Et_2O$ in light petrol) 2 mg of (**15**), 0·2 mg of (**21**), 150 mg of (**22**) and 500 mg of (**11**) were isolated. 6 mg of mixture of (**24**) and (**25**) were eluted with 40% $Et_2O$–light petrol. Finally, 60 mg of (**16**) and 80 mg of (**23**) were eluted with 80% $Et_2O$–light petrol.

*Compounds from the roots.* From the column the following compounds with increasing polarity were isolated: 336 mg of *β*-farnesene, 18 mg of (**9**), 2 mg of (**10**), 180 mg of (**1**), 6 mg of (**32**), 20 mg of (**2**), 14 mg of (**33**), 13 mg of (**5**), 2 mg of (**4**), 4 mg of (**34**), 5 mg of (**35**) and 50 mg of (**36**).

*Cleavage of the epoxide* (**14**). 1 mg of **14** was dissolved in 0·5 ml dioxane, 2 drops of $H_2SO_4$ were added and the soln was heated to 60° for 0·5 h. After reaction the product was subjected to UV inspection in $Et_2O$ ($\lambda_{max}$ 331, 310, 291, 274, 245·5 and 235 nm). The diol was treated with 5 mg $NaIO_4$ after addition of 2 drops of 2 N $H_2SO_4$ and kept at room temp for 15 min and the product was purified by TLC. The UV spectrum showed a bathochromic shift relative to the alcohol $\lambda_{max}$ 348, 326, 306, 288, 260 and 246 nm. Furthermore the product was identical with authentic dehydromatricarianal (**21**) on TLC.

*Preparation of the senecionate* (**15**). 300 mg of acetate (**11**) were dissolved in 60 ml MeOH and 25 ml 2 N NaOH added. Mixture was heated to 60°, for 10 min and subsequently evaporated to 15 ml. The product was extracted with $Et_2O$, dried ($CaCl_2$) and purified by TLC in $Et_2O$. Product [135 mg of (**23**)] was dissolved in 7 ml of anhydrous $C_6H_6$ and 0·5 ml of $C_5H_5N$ and 0·5 ml of senecionyl chloride added, under stirring. The product was dissolved in $Et_2O$ and soln washed with $H_2O$ to neutral pH and dried ($Na_2SO_4$). After evaporation of solvent, the product was purified by preparative TLC in 10% $Et_2O$–light petrol. Its UV, IR, NMR, MS and TLC data were identical with those of the naturally occurring compound (**15**).

*Hydrolysis of the acetate* (**5**). 8 mg of (**5**) in MeOH and 70 mg KOH was heated to boiling pt. It was then left at room temp. overnight, and evaporated to 2 ml; $H_2O$ was added and mixture extracted with $Et_2O$. Extract was dried ($Na_2SO_4$) and the product obtained proved to be identical with (**34**).

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