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VERY-LONG-CHAIN ALKYL ESTERS IN *CEREUS PERUVIANUS* WAX

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Key Word Index—*Cereus peruvianus*; Cactaceae; wax; very-long-chain alkyl esters.

Abstract—Very-long-chain alkyl esters were detected in the wax from *Cereus peruvianus* and their individual molecular species up to C_{62} were analysed by means of reversed-phase HPLC, TLC and capillary GC-mass spectrometry. More than 80 isomeric alkyl wax esters were identified.

INTRODUCTION

Very-long-chain alkyl esters are characteristic components of cuticular waxes of the leaves in some species [1–3]. Recently, we studied the molecular species of wax esters in *Cereus peruvianus* [4] by GC-mass spectrometry and found that they were composed of fatty acids (up to 30:0) and alcohols (up to 32:0); branched (both *iso*- and *anteiso*-) fatty acids and fatty alcohols were also found. The present work reports results from a continued investigation of these wax esters.

RESULTS AND DISCUSSION

The composition of the molecular species of wax esters with very-long-chain acids and alcohols is presented in Table 1. In contrast to our previous report [4], we have now identified alcohols, predominantly branched, up to C_{34}. For enrichment of long-chains, we prepared a fraction containing more than 52 carbons by means of reversed-phase HPLC. (see Experimental). Argentation-TLC and urea crystallization were as previously described [4–6].

Esters from C_{53} to C_{62} were detected. Major esters contained *n*-acid-monounsaturated alcohol, up to 35.6% of the total very-long-chain alkyl esters, see, for example, peak 58 in Table 1. Such an ester was found previously [4] but now one major and two minor peaks were detected; two minor peaks (26–32 and 28–30) overlapped.

The range of esters of *br*-acid-monounsaturated alcohol was expanded. The C_{53} peak was now resolved into two compounds. The major compound was *br*-acid-unsaturated alcohol 25–28 (93% of total), the minor one 23–30 comprising ≈7% of the total. The C_{35} ester was also separated into two compounds.

It is interesting to note combination of the longest fatty acid (30:0) and alcohol (*br* 34:0) was not detected. We suppose that these chains are not suitable for esterification for two reasons. Firstly, such compounds have very high insolubility and, secondly, they would be transported only with difficulty. We have identified more than 80 very-long-chain wax ester isomers from the leaves of *Cereus peruvianus*. Proportions of the individual isomers of identical chain-lengths were quantified by means GC-mass spectrometry as described previously [4, 7].

EXPERIMENTAL

Cereus peruvianus cv. Monstrosus was obtained from the Plant Adaptation Unit, Institute of Desert Research and was collected in September 1993. Extraction and sepn of wax esters by TLC was as described previously [4]. Total wax esters were sepd by means of reversed-phase HPLC on a RP-18 column. After injection, wax esters were eluted with hexane–THF (4:1); eluates before elution of hexacosanyl-hexacosanoate (R_t 27 min) were discarded. Alternatively, eluates after this R_t were collected and, after evapn, the residue was applied to TLC plates. All other analytical procedures are as described previously, with one exception. For GC-MS analysis, on-column injection (100°) and a fused-silica capillary column (Supelcowax SPB-1, 15 m × 0.25 mm ID × 0.10 μm film thickness) was used. The temp. programme was as

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Table 1. Composition of very-long-chain wax esters isolated from *Cereus peruvianus*

Carbon-number	*n*-Acid-monounsatd alcohol	%*	%†	%‡	Monounsatd-acid-*hr*-alcohol	%*	%†	%‡	*n*-acid-*n*-alcohol	%*	%†	%‡	Monounsatd-acid-monounsatd alcohol	%*	%†	%‡
53	21–32	1.8			24–29	100.0	12.4		25–28	23.8						
	23–30	17.5							26–27	14.7						
	25–28	37.1							27–26	45.0						
	27–26	43.6	16.2						28–25	11.0						
									30–23	5.5	30.4					
54	22–32	3.5			20–34	7.2			26–28	30.0			22–32	3.3		
	24–30	41.9			22–32	10.2			27–27	1.2			26–28	54.0		
	26–28	26.5			24–30	82.6	45.1		28–26	56.4				42.7	83.3	
	28–26	23.6							30–24	12.4	48.4					
	30–24	4.5	59.8													
55	23–32	13.6			26–29	100.0	2.2		27–28	39.4						
	25–30	40.6							28–27	24.2						
	27–28	45.8	3.5						30–25	36.4	4.6					
56	24–32	20.4			22–34	19.4			28–28	7.5			24–32	48.7		
	26–30	17.2			24–32	52.8			30–26	92.5	14.8		26–30	51.3	15.4	
	28–28	19.4			26–30	27.8	23.5									
	30–26	43.0	16.4													
57	25–32	40.0														
	27–30	60.0	0.6													
58	26–32	17.0			24–34	85.1			30–28	100.0	1.8		26–32	100.0	1.3	
	28–30	25.5			26–32	14.9	14.6									
	30–28	57.2	2.8													
59	27–32	100.0	0.1													
60	28–32	25.0			26–34	100.0	2.2									
	30–30	75.0	0.5													
62	30–32	100.0	0.1	35.9				9.7				15.2				15.9

Table 1—Continued.

Carbon number	*br*-Acid-monounsatd alcohol	%*	%†	%‡	*n*-Acid-*br*-alcohol	%*	%†	%‡	*br*-Acid-*br*-alcohol	%*	%†	%‡	Monounsatd-acid-*n*-alcohol	%*	%†	%‡	*br*-Acid-*n*-alcohol	%*	%‡	%‡
53	a23–30	6.9			19–i34	6.2			a19–i34	6.7			26–27	100.0	23.6		b25–28	100.0	100.0	0.1
	a25–28	93.1	65.9		21–i32	4.5			a23–i30	40.0										
					23–i30	21.2			i24–a29	53.3	33.3									
					24–a29	62.8														
					28–a25	5.3	20.2													
54	i24–30	100.0	18.2		20–i34	5.8			i20–i34	20.0			26–28	100.0	76.5					
					22–i32	12.9			i24–i30	80.0	66.6									
					24–i30	76.7														
					25–a29	1.4														
					30–i24	3.2	49.7													
55	a23–32	5.3			21–i34	13.9														
	a25–30	94.7	10.6		23–i32	22.3														
					25–i30	33.3														
					26–a29	22.2														
					30–a25	8.3	6.7													
56	i24–32	100.0	3.0		22–i34	27.3														
					24–i32	53.7														
					26–i30	18.2														
57	a25–32	100.0	2.3	5.1	27–a29	0.8	23.4	11.9				2.6				3.6				

* Values obtained by mass spectrometry [single ions, i.e. ions $RCOO^+$, $RCOOH_2^+$ (from acid) and $R'\text{-}1^+$ (from alcohol)].

† Groups quantified by capillary GC-MS (total ion current).

‡ Groups separated by urea crystallization and Ag-TLC.

follows: 100° for 1 min, then increased at 20° min^{-1} to 240° and at 4° min^{-1} to 320° then at this temp. maintained for 10 min. The carrier gas was H_2 at a flow-rate of 150 cm s^{-1}. NH_3 (0.6 torr) was used as the CI (positive and/or negative mode) reagent gas. MS were scanned between *m/z* 200–900.

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