

EPICUTICULAR WAX COMPOSITION OF *ECHINOCHLOA CRUSGALLI**

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Previous analyses of waxes from grasses of the subfamily Panicoideae [1–4] suggested that there might be noticeable differences between these waxes and those from the subfamily Festucoideae [5, 6]. Preliminary investigation of wax from *Echinochloa crusgalli* (L.) Beauv., barnyard grass, which is an annual weed over much of North America, indicated an unusual composition. To obtain larger amounts of wax, however, a cultivated variety *E. crusgalli* var. *frumentacea* (Roxb.) W. F. Wight (Japanese millet) was grown and the wax extracted.

Yield of wax (Table 1) was much lower than that usually found [5, 6] and there was no single major component. Other grass waxes [5, 6] often contain a single free alcohol or β -diketone as principal component. The large unidentified fraction was not one component but many compounds distributed throughout the fractions obtained by column chromatography.

The compositions of the fractions (Table 2) were unusual in that each contained a wide range of chain lengths and only in the aldehyde fraction did one component exceed 50%. Thus free alcohols range in chain length from C_{24} to C_{32} whereas alcohols from waxes of most members of the Festucoideae previously examined [5, 6] contain C_{26} or C_{28} alcohol as principal constituent. The free alcohol composition is similar to that of sugar cane wax [3] and of two *Andropogon* species [4]; also C_{32} alcohol is a major component of wax of *Zea mays* [1].

The amount of free acids (10%) is appreciably larger than that found in festucoid grass waxes [5, 6] but sugar cane wax contains 11.6% [3] and wax of two varieties of *Sorghum vulgare* [2] contains much larger percentages although this is probably unusual. Free acids contain a much smaller proportion of C_{14} to C_{22} acids than is usually observed but combined acids, with major C_{20} to C_{24} components, are similar to those of many other grass waxes [5, 6]. Aldehydes, which form 12% of the wax, resemble free acids more closely than free alcohols suggesting a closer biosynthetic relationship with the acids.

Ester composition, C_{40} to C_{56} with maximum at C_{46} , was similar to that of most other grasses. The composition of the combined alcohols obtained on hydrolysis, however, was C_{20} to C_{30} with C_{24} the major component, considerably different from that of the free alcohols, C_{24} to C_{32} with C_{32} the dominant component. Usually combined and free alcohols have similar compositions with the same major component [5, 6]. GC–

Table 1. Composition and yield of epicuticular wax from *Echinochloa crusgalli**

Component	%
Hydrocarbons	17
Esters	20
Aldehydes	12
Free acids	10
Free alcohols	10
Unidentified fractions	31
Yield (% dry wt)	0.06

* Determined from weights of components obtained by silicic acid chromatography.

MS analysis showed that the esters contained most of the possible combinations of the combined acids and alcohols (Table 2) but the percentages of the longest chain esters are lower than those which would have resulted from random esterification. Esters formed randomly from alcohols with the free alcohol composition would have had longer chain lengths, mainly C_{50} to C_{60} . Such longer chain esters are usually only found in waxes of tropical plants [5, 7] and might be too high melting for plants growing in cooler climates. Possibly enzymes specific for only the shorter alcohols are involved in interesterification or different 'pools' of alcohols exist.

Thus, in general, the composition reported here for epicuticular wax of *E. crusgalli* agrees with previous observations that waxes of panicoid grasses have a distinctly different composition from those of festucoid grasses [4].

EXPERIMENTAL

E. crusgalli var. *frumentacea* cv Chiwapa was grown outside from seed supplied by Plant Materials Center, U.S. Department of Agriculture, Coffeerville, Mississippi. Only vegetative growth occurred but plants were cut and wax extracted [4] 105 days after germination. Wax was fractionated by column chromatography and fractions identified by GLC and TLC as previously described [4]. Esters and aldehydes were obtained as a combined fraction and were separated by TLC; aldehydes were identified by methods reported earlier [6]. Chain lengths and structures of the esters, free acids and combined alcohols were confirmed by GC–MS analysis [8].

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Table 2. Composition of wax fractions from *Echinochloa crusgalli**

No. of carbon atoms	Hydrocarbons	Esters	Hydrolysis products of esters		Aldehydes	Free acids	Free alcohols
			Acids	Alcohols			
16	—	—	2	—	—	—	—
18	—	—	9	—	—	—	—
20	—	—	26	2	—	—	—
22	—	—	32	19	—	2	—
23	1	—	—	—	—	—	—
24	—	—	23	36	4	21	2
25	4	—	—	—	—	—	—
26	—	—	4	17	15	20	8
27	15	—	—	—	—	—	—
28	—	—	2	21	55	24	33
29	11	—	—	—	—	—	—
30	—	—	2	5	15	12	19
31	28	—	—	—	—	—	—
32	—	—	—	—	4	8	38
33	30	—	—	—	—	—	—
34	—	—	—	—	—	4	—
35	8	—	—	—	—	—	—
36	—	—	—	—	—	4	—
40	—	1	—	—	—	—	—
42	—	9	—	—	—	—	—
44	—	23	—	—	—	—	—
46	—	32	—	—	—	—	—
48	—	16	—	—	—	—	—
50	—	7	—	—	—	—	—
52	—	5	—	—	—	—	—
54	—	1	—	—	—	—	—
56	—	1	—	—	—	—	—
Unidentified	3(7)	5(8)	—	—	7(5)	5(8)	—

* Components with emergence temperatures which indicated structures which were not straight chain and saturated and for hydrocarbons did not contain an odd number of carbons, and for esters, acids, alcohols and aldehydes did not contain an even number of carbons have been listed as unidentified; components present in less than 0.5% have been omitted.

REFERENCES

1. Bianchi, G. and Salamini, F. (1975) *Maydica* **20**, 1.
2. Bianchi, G., Avato, P., Bertorelli, P. and Mariani, G. (1978) *Phytochemistry* **17**, 999.
3. Stranský, K., Zajic, J., Streibl, M., Doubravová, L. and Ubik, K. (1978) *Seifen-Öle-Fette-Wachse* **104**, 21.
4. Tulloch, A. P. and Hoffman, L. L. (1979) *Phytochemistry* **18**, 267.
5. Tulloch, A. P. (1976) in *Chemistry and Biochemistry of Natural Waxes* (Kolattukudy, P. E., ed.) p. 235. Elsevier, Amsterdam.
6. Tulloch, A. P. and Hoffmann, L. L. (1977) *Can. J. Botany* **55**, 853.
7. Tulloch, A. P. (1973) *J. Am. Oil Chem. Soc.* **50**, 367.
8. Tulloch, A. P. and Hogge, L. R. (1978) *J. Chromatogr.* **157**, 291.