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LUMINESCENT INVESTIGATION OF THE COMPONENTS OF SUGAR CANE

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A luminescent analysis of sugar cane has been made, and the dependence of the amounts of the components investigated on the organ, variety, and age of the plant has been established. Rising relative amounts of lignin have been found in the sequence leaves, stem, and trunk with increasing age of the plant, and this to a greater degree for the variety I 60–5 than for S 374–72.

Sugar cane is a very important source not only of raw sugar but also of cellulose, lignin processing products, and biomass. Its practical significance dictates the need for studying the productivity of sugar cane and the influence of various factors on it. In the study of the biosynthesis, composition, and structure of plants, wide use is made of spectroscopic, and, especially, luminescent methods [1, 2], but in the case of sugar cane they have been used inadequately [3, 4].

In the present paper we give the results of an investigation of the fluorescence and fluorescence-excitation spectra of sugar cane and analyze the changes in its component composition according to the organ (leaves, stem, trunk), variety (I 60–5, S 374–732), and age (9 and 15 months) of the plant.

The nature of the spectral curves shows that the fluorescence is the sum of the emission of several centers.

In the interval of 400-600 nm on excitation at λ_E = 300-360 nm a broad structureless emission band was observed in the region of 430-480 nm (Fig. 1). The form and position of the band changed according to the organ and age of the plant. In the spectrum of a young plant (9 months) a band was observed at 430-440 nm, and with an increase in age (15 months) it shifted to 445-450 nm. In the case of the lignified parts (stem, trunk) the absorption maximum shifted in the long-wave direction, to 450-480 nm In the full-grown plant the pattern was the opposite – a small short-wave shift to 430-440 nm. With an increase in λ_E to 420 nm, the fluorescence maximum shifted to 485-530 nm. In the excitation spectrum with recording at λ_R = 435-470 nm a band was observed with its maximum at 345-375 nm. But, while in the excitation spectrum of the leaves there was a single maximm at 345 nm, in the spectra of stems and trunks an additional band appeared at 375 nm. With increasing age of the plant, this band became predominating and, in a number of cases, the only one. On recording at λ_R = 500-520 nm the excitation spectrum was enriched with maxima at 430 and 465 nm. According to the literature [1], the emission of green leaves in the interval of 435-450 nm (excitation at 345 nm) has been identified as the fluorescence of reduced forms of pyridine nucleotides, NADH and NADPH, and that at 500-520 nm (excitation at 445 nm) as the fluorescence of the oxidized forms FMN and FAD. In the lignified parts of the plants, intense emission in the 440-470 nm region (excitation at 340-350 nm) has been determined as the luminescence of lignin [5, 6]. The difference in the positions of the maxima in the fluorescence and excitation spectra of different parts of sugar canes of different ages can be explained by the presence of several centers of emission in this interval and a change in their ratio.

In the interval of 600-800 nm (λ_E = 420 nm) there were two fluorescence bands, with maxima at 675-695 and 705-750 nm. According to the literature [1, 2, 7, 8], chlorophyll *a* (Chl *a*) molecules fluoresce at 600-800 nm. The short-wave fluorescence band (675-695 nm) relates predominantly to the Chl *a* of photosystem 2 (PS 2), and the long-wave band (705-750 nm) predominantly to the Chl *a* of photosystem 1 (PS 1). The fluorescence-excitation spectra of this region have a complex structure. In addition to the Chla band at 420 nm (Soret band) there are bands of auxiliary pigments – chlorophyll

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TABLE 1. Relative Intensities of the Fluorescence of the Sugar Cane Specimens Investigated

Plant variety	Age, months	Material	λ_E = 345 nm	λ_E = 420 nm	
			I_{435}/I_{680}	I_{520}/I_{680}	I_{730}/I_{680}
I 60—5	9	Leaf	6.54	0.65	0.96
		Stem	13.0	0.49	0.85
		Trunk (outer part)	14.59	4.22	0.58
		(inner part)	64.60		
	15	Leaf	9.05	0.65	1.07
		Stem	48.62	1.14	0.54
		Trunk (outer part)	67.27	3.37	0.22
		(inner part)	73.60		
C 374—72	9	Leaf	2.25	1.22	1.22
		Stem	5.50	0.82	0.89
		Trunk	12.77	1.18	0.46
	15	(outer part)	4.79	0.83	1.09
		(inner part)	35.63	0.70	0.33
		Leaf	65.04	6.70	0.23

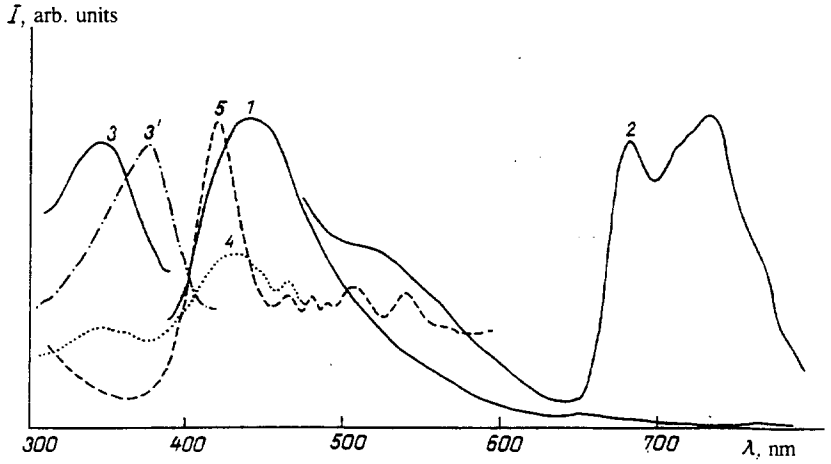

Fig. 1. Fluorescence and excitation spectra of the leaves (*1-5*) and stem (*3'*) of a sugar cane of the variety I 60-5 (15 months): *1*) λ_E = 345 nm; *2*) λ_E = 420 nm; *3,3'*) λ_R =445 nm; *4*) λ_R = 520 nm; *5*) λ_R = 680 nm.

b (Chl b) at 465 nm and of carotenoids at 480-540 nm [1, 2, 8]. The presence of maxima in the region of absorption of Chl b and carotenoids reflects the participation of the auxiliary pigments in the absorption of light and the transmission of this energy to Chl *a*.

Not only the shape and position but also the integral intensity and the ratio of peak intensities of the fluorescence bands depend on the organ, variety, and age of the plant (Table 1). Thus, the integral intensity of fluorescence in the 600-800 nm interval (λ_E = 420 nm) decreases in the sequence leaf, stem, trunk. This result is normal, since it is in this region that the Chl *a* molecule fluoresces. The ratio of the intensities of the fluorescence bands at 730 and 680 nm (I_{730}/I_{680}) also decreased in the same sequence. With increasing age of the plants of both varieties a decrease in the I_{730}/I_{680} ratio was also observed for all the components investigated (with the exception of the leaves of I 60−5). The greatest dependence on ripeness was observed for the lignified parts of the plant and was practically the same for the two varieties. According to the assignment of these bands, the results obtained indicated a fall in the relative amount of the long-wave forms of Chl *a* in the seuqence leaf, stem, trunk, and also in all the components (with the exception of the leaves of I 60−5) with an increase in the age of the plant. The dependence on age was greatest for the lignified parts and was practically the same in the two varieties.

So far as concerns the intensity of fluorescence at 520 and 680 nm (λ_E = 20 nm), no general laws were observed here. This is possibly due to a complex origin of the 520 nm band with equivalent contributions of the different components and dissimilar tendencies of variation.

Conversely, the integral intensity of fluorescence in the 400-600 nm interval (λ_E = 3435 nm) and the relative intensity of the band at 430-440 nm (I_{435}/I_{680}) increase in the sequence leaf, stem, trunk and for each part of the plant with increasing age. The dependence of the intensity ratio I_{435}/I_{680} on the age of the plant was greatest for the lignified parts and was far smaller for the leaves. This confirms the hypothesis of the predominant contribution of the fluorescence of lignin to the fluorescence of the lignified parts of the sugar cane in the 400-600 nm region. Regardless of the stage of ripeness, all parts of the sugar cane variety I 60−5 were characterized by higher values of I_{435}/I_{680} than the variety S 374−72. At the same time a powerful dependence of I_{435}/I_{680} on the age of the plant was observed for variety S 374−72 and a weaker one for variety I 60−5.

Since the predominant contribution to the band at 435 nm is made by the fluorescence of lignin, the relationships obtained reflect changes in the relative amount of lignin. A general characteristic for both varieties was an rise in the relative amount of lignin in the sequence leaf, stem, trunk, and also with increasing age of the plant. At the same time, variety I 60−5 was characterized by by a higher relative lignin content than variety S 374−72. The process of development of the plant was accompanied by considerable changes in the relative amount of lignin in the outer surfaces of the lignified parts of sugar cane variety S 374−72 and less pronounced ones in the leaves and in variety I 60−5.

EXPERIMENTAL

Fluorescence and excitation spectra were recorded on a Fica-55 spectrofluorimeter, with automatic correction of spectral sensitivity, by reflection at the same amplification parameters. The width of the excitation and recording monochromator slits was 7.5 nm.

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