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**GENOTYPIC DIFFERENCES IN THE PHOTOSYNTHETIC ACTIVITY AND CHLOROPLAST ULTRASTRUCTURE OF SUNFLOWER LEAVES**

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**SUMMARY**

Genotypic differences in photosynthetic activity and chloroplast structure of the leaves of CR-2 restorer and CH-0 cytoplasmic male sterile sunflower lines /parental lines of the single-cross registered hybrid VIKI/ have been determined. In the CR-2 leaves in the beginning of the irradiation the lag-phase of the oxygen evolution was shorter, the fluorescence quenching of chlorophyll-a was faster, the rate of de-epoxidated violaxanthine and the starch accumulation was lower than in the CH-0 leaves. The greater photosynthetic activity of CR-2 may be related to: a/ a larger surface of the chloroplast inner membrane system, b/ a higher ratio of the nonstacked membrane, c/ a more frequent occurrence of multi-thylakoid grana, d/ an increased swelling of the intrathylakoidal space in darkness.

**INTRODUCTION**

When the green leaves of CR-2 and CH-0 sunflower lines are irradiated under identical conditions intraspecific differences can be observed in the activity of xanthophyll cycle, photosynthetic O<sub>2</sub> evolution, carbohydrate partition, induction of fluorescence, etc.

These processes take place in the chloroplast. The question is that

- a/ how these functional deviations in the EM structure of the chloroplast are expressed
- b/ what kind of role these deviations may have in the biomass production of sunflower genotypes.

**MATERIALS AND METHODS**

Up to the age of 4 to 6 weeks, the CR-2 and CH-0 inbred sunflower lines were grown in the mixture of sand-perlite 1:1, with nutrient solution of Hoagland. The temperature in the phytotron climate chambers was 23-1°C and the photon flux density 200  $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  during 16 hours light period.

In fully developed leaves the following parameters were measured: the speed rate of violaxanthin conversion /Maróti and Gábnai, 1971; Maróti, 1982; Maróti and Pataky 1983/, oxygen evolution /Delieu and Walker, 1981/, the slow fluorescence of chlorophyll-a /Solymosi and Lehoczki, 1983/, carbohydrate content /Dubois et al. 1956/. A detailed biometry of ultrastructure of chloroplast was performed.

**RESULTS**
**Dry matter accumulation**

The weight of leaves per plant and total dry matter were always higher with CH-0 than with CR-2. /Fig.1./ The dry matter accumulation of both lines, when grown at increasing phosphate concentration show significant differences in their response to P concentration.

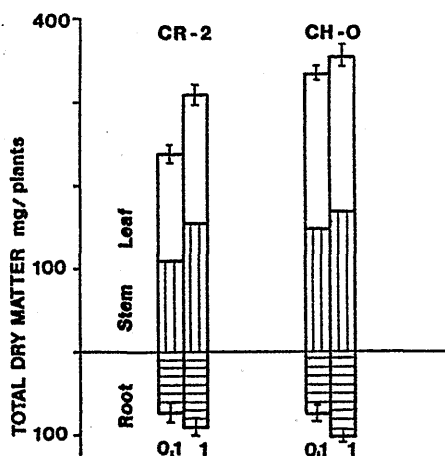


Fig.1. Total dry mass accumulated in two inbred lines of sunflower, grown in phytotron for 37 days in Hoagland nutrient solution modified with 0,1 and 1 mM  $H_2PO_4^-$  concentration.

#### Carbohydrate content

The soluble sugar and starch content of the leaf are influenced by phosphate concentration and internal factors, e.g. the genotype. Significantly higher amount of starch was found in the leaf of the CH-0 line both 0,1 and 1 mM phosphate application compared to the CR-2 leaf /Table 1./.

Table 1. Carbohydrate content in leaves of two inbred lines of sunflower grown for 37 days in Hoagland nutrient solution modified with 0,1 and 1 mM  $H_2PO_4^-$  concentration.

Line	Phosphate supply mM dm <sup>-3</sup>	Soluble sugar	Starch content
		$\mu\text{g mg}^{-1}$ dr. wt.	$\mu\text{g mg}^{-1}$ dr. wt.
CR-2	0,1	148 <sup>+12</sup>	202 <sup>+18</sup>
	1,0	103 <sup>+8</sup>	228 <sup>+21</sup>
CH-0	0,1	135 <sup>+8</sup>	341 <sup>+20</sup>
	1,0	168 <sup>+14</sup>	325 <sup>+10</sup>

#### Oxygen evolution

In the evolution of photosynthetic oxygen, the time /phase/ between the onset of illumination and the attainment of this maximum steady state rate is called the induction period.

The oxygen evolution rate of the leaves of sunflower /CR-2, CH-0/ is characterized by a "burst" at the beginning of the induction period. This is often followed at CH-0 by a decrease, after this the ratio of oxygen evolution exponentially increases /Fig.2./.

the leaf of CR-2,  $O_2$  "burst" is very strong. In the maximum rate of oxygen evolution, there is a significant difference as well.

#### Fluorescence induction kinetics

The slow phases of the fluorescence induction curves of sunflower (CR-2, CH-0) are different from each other /Fig.2./. The fluorescence decreases through states P, S, M and T, where P is an initial peak, M is a transitional peak, S is minimum level between P and M, T is the final steady-state level. On the induction curve of sunflowers the transition S-M is absent. The fluorescence quenching of leaf CR-2 is faster than that of CH-0 /Fig.2./. In the fluorescence decrease of chlorophyll-a two main components take place: photochemical quenching qP /Bradbury and Baker, 1984/ and non-photochemical qnP /Horton, 1983/. The qP is in connection with the re-oxidation of PS II electron acceptors Q, and qnP with the formation of the transthylakoid  $H^+$  gradient. According to Brandbury and Baker /1984/, between peaks P and M reoxydation of Q /qP/ has the main role in the change in fluorescence yield, on the other hand, from M to T, the qnP / $\Delta pH$  quenching/ significantly increases.

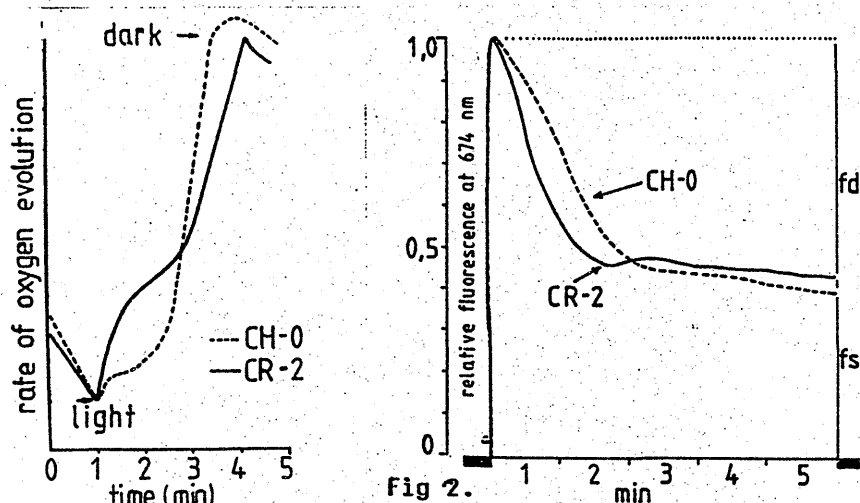


Fig. 2. Oxygen evolution rate and relative fluorescence at 674 nm of leaves of CR-2 and CH-0. In leaves of CR-2 at the onset of illumination the lag phase of oxygen evolution is shorter, the  $O_2$  "burst" is stronger, fluorescence quenching of chlorophyll-a is faster than in CH-0 leaves.

#### The different activity of xanthophyll cycle

When the green leaves of sunflower are irradiated a strong light / $800 Wm^{-2}$ / for two minutes, there is a great difference in the degree of violaxanthin decrease between the genotype pairs : CR-2, CH-0 /cf Table 2/.

Table 2. Light-dependent xanthophyll conversion (under strong light :  $800 \text{ Wm}^{-2}$ ) in sunflower leaves.

Plants	Xanthophyll content ( $\mu\text{g}/\text{mg}$ chlorophyll-a)				
	Antheraxanthin		Violaxanthin		%
	Dark	2 min light	Dark	2 min light	
Cr-2	16,5	19,7	72,3	66,7	- 7
CH-0	7,8	15,2	82,7	62,1	- 25

The leaves of the same age and position of genotype pairs were collected after 8 hrs darkness. Percentage is the decrease of violaxanthin, as compared with dark controls. In the CH-0 leaves in the first minutes of illumination, considerably more violaxanthin transforms than in CR-2 leaves.

#### Chloroplast structure

It is to be asked what kind of chloroplast structure the different photosynthetic activity is connected with. We have demonstrated that:

- a/ In the CH-0 chloroplast, the grana consisting of few /2,3,4,5/ thylakoids, occur, with higher frequency /Fig. 3./.

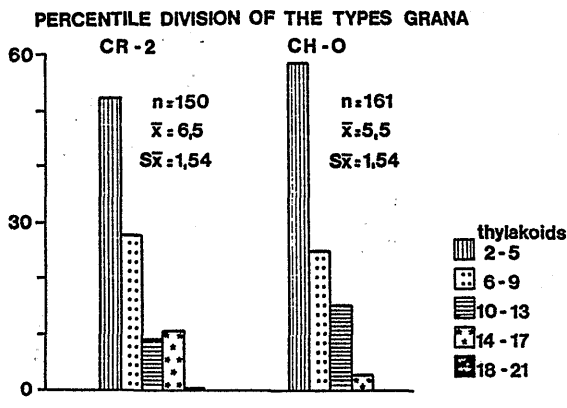


Fig. 3. Partition (%) of grana types in the CR-2 and CH-0 palisade chloroplasts. The data are the average values of 22-22 chloroplasts (section-surface) and 150-161 grana.

- b/ There is more appressed and non-appressed regions on a unit of section in CR-2 chloroplast than in those of CH-0 /Table 3/.

Table 3. Quantity of the thylakoid membrane system of CR-2 and CH-0 palisade chloroplasts.

Plants	Thylakoid membrane $\mu\text{m}/\mu\text{m}^2$		
	partition	stroma	terminal
CR-2	20,3	8,5	4,6
CH-0	18,7	5,3	3,6

c/ In dark, the loculus of the CR-2 thylakoids are much more swollen than the intrathylakoid spaces of CH-0.

d/ Starch accumulation for CH-0 in the chloroplast is much more important than in CR-2.

#### DISCUSSION

In the CR-2 leaves at the beginning of the irradiation a lower rate of violaxanthin de-epoxidates, the lag phase of oxygen evolution is shorter, the quenching of the fluorescence of chlorophyll-a is faster than those in the CH-0 leaves.

In the CR-2 chloroplast a lower rate of violaxanthin may be transformed because at the beginning of the illumination both the rate of  $\text{H}^+$  use /ATP formation/ and the initial rate of the noncyclic electron transport are quicker.

According to our measurements, the lower activity of xanthophyll cycle of CR-2 is in a positive correlation with a/ a larger surface of the thylakoid membrane system; b/ a higher ratio of non-appressed stroma exposed regions; c/ a more frequent occurrence of multi-thylakoid grana; d/ an increased swelling of the intrathylakoid space in darknees.

The role played by the above genotypic properties in the adaptation of plants to light is not properly known as yet.

#### ACKNOWLEDGENTS

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