

RELATIONSHIP BETWEEN NON-STRUCTURAL CARBOHYDRATE CONTENT OF THE LEAF AND GROWTH IN MAIZES GROWN IN SHORT AND LONG LIGHT-DARK PERIODS

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Abstract

The dry mass accumulation as well as soluble sugar and starch content of the leaf lamella of P165 and F2 inbred maize lines were studied in 16-8 hours, 30-15 mins and 15-7.5 mins light-dark periods (LDPs), on the 16th, 32nd and 48th days counted from sowing.

It was determined that the growth rate of the two maize lines also differed in the 16-8 hours LPD: first the dry mass of the P165 was greater, then that of the F2 line.

The starch content of the 2. leaf lamella in the second week following its complete development increased in the case of the P165 maize, that of the 4. leaf decreased; it barely changed in the case of the F2 maize in 16-8 hours LDP.

The short LDPs generally decreased the dry matter and leaf carbohydrate content, but the 30-15 min. LDP — even in case of the more sensitive P165 — produced an opposite effect sometimes: the dry mass of leaf of the 16 day old plants (and the 32 day old F2) as well as the starch content of the developed 2. leaf increased. The decrease of total non-structural carbohydrate content of the leaf firstly inhibited the growth of the root.

The relative starch content was higher in the basal part of the leaf blade than that of in the apex.

Key words: maize line, light-dark period, dry mass, soluble sugar and starch content

Introduction

The growth of a plant greatly depends on its total non-structural carbohydrate (TNC) content, distribution as well as the intensity of its transport and metabolism. GENT (1984) found tight positive correlation between the TNC content and relative growth ratio of young tomato plants. Relationship was found between the growth per organ of the biomass and the carbohydrate distribution pattern (SCHULZE et al. 1983). There is a negative connection between the starch content of the leaf and the relative mass of the root (HUBER, 1983).

The TNC content of the leaf and within this the ratio of the starch and sugars are influenced by external and internal factors, e.g. the genotype (HUBER, 1983; AVIGAD, 1982). During the increase of the assimilation rate the starch synthesis shows a more abrupt rise than that of the sucrose thus their ratio changes in the leaf of bean plants (SHARKEY et al. 1985).

In young maize the fixed carbon incorporates into reductive sugars in the lower leaves, and into sucrose in the upper ones (MOROT-GAUDRY et al. 1979).

The level of transport sugar increases during the expansion of the leaf (PHARR and SAX, 1984). The starch content in the developed source leaf inversely depends on the ratio of sink and source parts of the plant (MAYORAL, 1985). There is a difference between the utilization and transport, resp., of the sugars and starch during the dark period: in the lower leaves there is a greater decrease in the starch content, while in the upper leaves the greater decrease is found in the sugar content (CHANG, 1980). From the effects of the environment the shortening of the light period causes an increase in the starch content of the leaf (CHATTERTON and SILVIUS, 1979).

The earlier studies performed in alternating light-dark periods (LDPs) demonstrated that if the plant receives the daily light amount in short periods alternated with dark, this generally has an unfavourable effect (SAGER and GIGER, 1980). The dry mass and carbohydrate content (MARÓTI and MIHALIK, 1984; MARÓTI and MARGÓCZI, 1984); the amount of pigments in the leaf (MARÓTI, 1982); the grana area ratio in the chloroplasts (TAKÁCS and MARÓTI, 1984) decrease. These changes are firstly determined by the length of the light period, but also depend on the genotype (MARÓTI et al. 1981), and the above mentioned publications occasionally describe positive effects as well. There are no data available in regard to the behaviour of various aged plants and leaves, resp., during short LDP.

The present paper studies two maize lines of dissimilar behaviour in two short LDPs, which often differ in their effects. Analysis is given of the distribution of the dry mass at 3 ages, as well as of the soluble sugar and starch contents of the 2. and 4. leaves in their fully developed and older state. Our aim was to search for correlations between the carbohydrate content of the leaf and the growth, furthermore, to analyse and interpret the differing behaviour of the maize lines.

Materials and methods

P165 and F2 inbred maize lines were used in our experiments. The maize grains were sown in the mixture of sand and perlite in a ratio of 1:1. 3-3 plants were placed in each 600 cm³ sized plastic pot. The humidity and nutrient content of the medium was ensured by modified HOAGLAND nutrient solution (see composition MARGÓCZI and MARÓTI, 1985). The moisture content of the medium (80% of the whole water capacity) was maintained daily by watering with distilled water. The plants received 20 ml of nutrient solution twice a week per pot.

The temperature in the phytotron climate chambers was 21 ± 2 °C, ensuring $185 \mu\text{mol m}^{-2} \text{s}^{-1}$ light intensity during the light periods with F33 type light tubes.

Three kinds of light treatments were applied: in the first chamber 16 hours light and 8 hours dark (16-8 hours LDP), in the second 30 min. light and 15 min. dark (30-15 min. LDP), and in the third 15 min. light and 7.5 min. dark (15-7.5 min. LDP) alternated. The plants were analysed at the age of 16, 32 and 48 days. 4-4 repetitions were formed from the 12-12 plant per treatment. The 2. leaves of the 16 day old plants, on which the lingules had just appeared (this indicating the complete development), were immediately fixed with heat following cutting (105 °C, 5 min.) and dried at 60 °C. The soluble sugar and starch contents were measured separately in the basal and apex part of the dried leaf blades (without the midrib). The just developed 4. and 2. leaves of the 32 day old plants, as well as the older 4. leaves of the 48 day old plants were analysed in similar manner. The dry mass of the other parts of the plants was measured per organ, after drying at 60 °C.

The carbohydrate content was determined according to the earlier description (MARÓTI and MARGÓCZI, 1984): the sugars were extracted with hot water, the starch from the remaining substance with perchloric acid. The carbohydrate content in the extracts was measured by the colorimetric method of DUBOIS et al. (1956). Soluble sugar + starch = total non-structural carbohydrate (TNC). The obtained results were evaluated by two-factored variancy-analysis (SVÁB, 1981).

Results

1. DISTRIBUTION OF DRY MASS PER LEAF STAGE AND DRY MASS RATIO OF THE ROOT

The dry mass of the 1., 2. and 3. leaf blade was less in the case of the F2 line compared to that of the P165 maize, and its increase was also slighter. The mass of the 4. leaf was close to similar in both lines, but the 5. and 6. leaves had greater mass in the F2 line and also grew faster in the 16–8 hours LDP (Fig. 1).

The 30–15 min. LDP exerted positive (or indifferent) effect on the accumulation of dry mass of the leaf in every leaf stage at 16 day old age, and the 15–7.5 min. LDP did not cause significant decrease either, compared to the 16–8 hours LDP. At the age of 32 days a considerable decrease was observed in the dry mass of the 4. leaf, having most of all source nature. This was also the case concerning the 5. leaf of the P165 line. However, the 5. leaf of the F2 had significantly greater mass in the short LDP, compared to the 16–8 hours LDP (Fig. 1b). Even at the age of 48 days the dry mass of the largest, chiefly source-character leaves showed the greatest decrease in the short LDPs (Fig. 1c). Between the age of 16 and 48 days there was a decrease in the root dry mass ratio of the plants (root dry mass/total dry mass) (Fig. 1). In the case of the F2 line this decrease was greater than in that of the P165.

The short LDPs — especially the 30–15 min. LDP — reduced the dry mass ratio of the root.

2. CARBOHYDRATE CONTENT IN THE 2. AND 4. LEAF BLADES

In the 16–8 hours LDP the leaves of P165 and F2 lines accumulate the carbohydrates differently: significantly higher amount of starch was found in the 2., older leaf of the P165 line compared to the developed leaf (Fig. 2a), while this was not so in the case of the F2 maize leaf (Fig. 2b). The carbohydrate transport from the 2. leaf of the F2 line was probably more intensive. On the contrary, the starch level slightly decreased (but significantly) in the older 4. leaf of the P165 maize compared to the developed leaf, but that in the 4. leaf of the F2 line did not (Fig. 2b); i.e. here the transport was possibly more intensive from the 4. leaf of the P165 line.

The soluble sugar and starch content in the 2. and 4. leaf blade of the P165 line showed extrem reaction to the short LDP treatment (Fig. 2a). The sugar content decreased considerably and significantly in every case compared to the 16–8 hours LDP, similarly decreased the starch content in the older 2. and developed 4. leaves of the 32 days old plants.

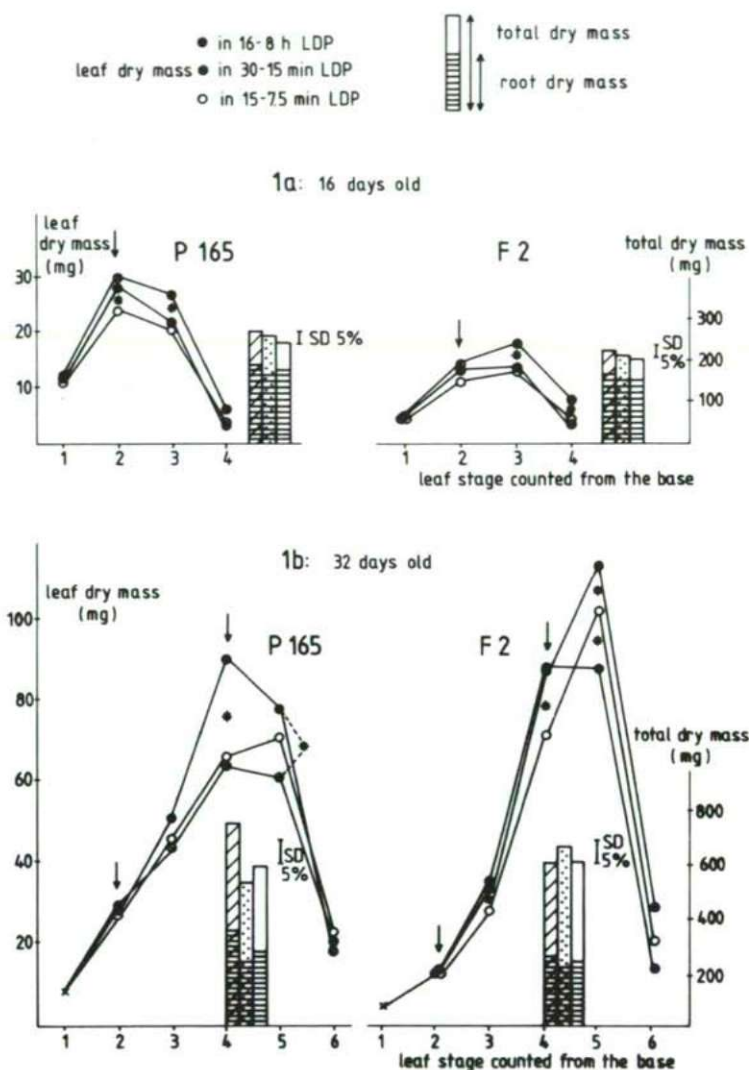


Fig. 1. a—b

The starch content of the just developed 2. leaf was strikingly high in the 30–15 min. LDP compared to the 16–8 hours LDP (Fig. 2a). This was presumably due to the fact that this LDP enhances the carbohydrate transport of the seed (and perhaps even that of the leaves), this is why the leaves become bigger here at the age of 16 days. This has been assumed in the case of other maize genotypes, too (MARGÓCZI, 1984).

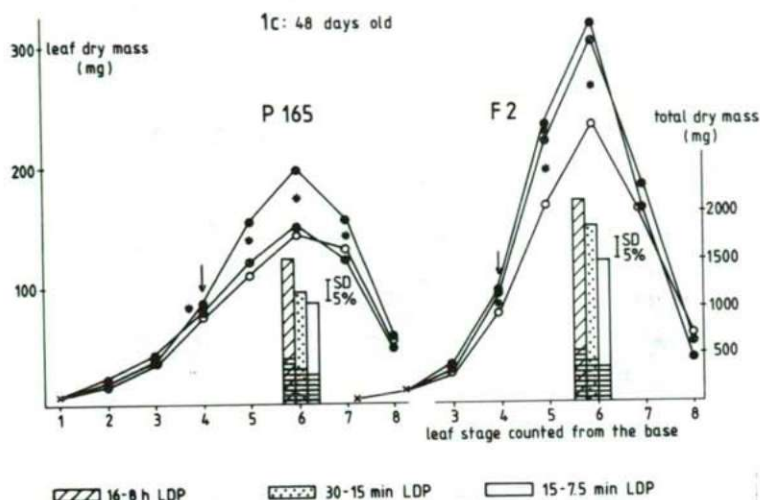


Fig. 1. Effect of short LDP treatment on the dry mass of the leaves per leaf stage; on the dry mass of the whole plant, and within this on the root share in the case of a: 16, b: 32, c: 48 day old plants.
 *: Significant difference at the level of $P = 0.05$. Carbohydrate content determination from the leaves labelled by arrows.
 $SD_{5\%}$: Significant difference at the level of $P = 0.05$ between the values per treatment of the total dry mass.

In the case of the F2 genotype an elevated starch level in the 2. leaf could hardly be observed on the effect of 30-15 min. LDP (Fig. 2b). This line was at a disadvantage at the age of 16 days in comparison with the P165 line (Fig. 1a), i.e. the reserved utilization of the seed was less intensive and was only slightly increasing on the effect of 30-15 min. LDP.

The carbohydrate content in the studied leaves of the F2 line only occasionally showed significant decrease on the effect of both short LDP treatments (e.g. the sugar content of the 4. leaf, Fig. 2b). The starch level of the leaf blade was only decreased in places significantly by the 15-7.5 min. LDP. It has also been found to be generally valid in the case of other maizes that the short LDP produces a greater decrease in the soluble sugar, than in the starch level of the leaf (MARGÓCZI, 1984).

3. TNC CONTENT OF THE LEAF AND RELATIVE GROWTH

The relative growth of the root and the young leaves was compared in the given time period with the TNC content of a leaf stage which had stopped growing, thus being of source nature and exporting photosynthates (Fig. 3).

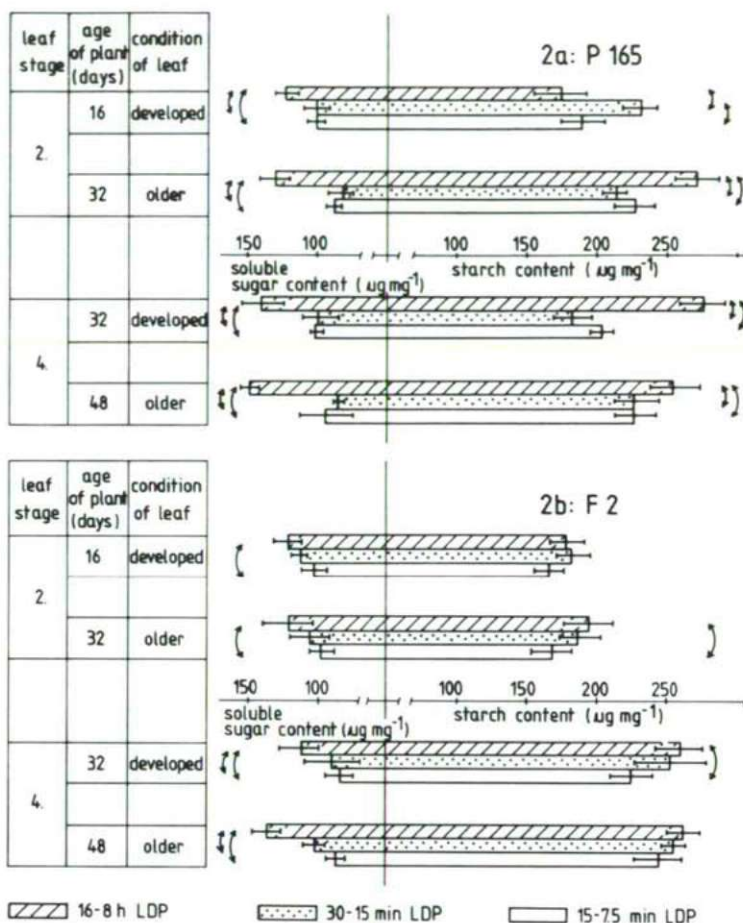


Fig. 2. Soluble sugar and starch content in the blades of the 2. and 4. leaves related to dry mass unit, in the case of leaves of two different stages of development in long and two short LDPs. a : P165 line, b: F2 line.

(The data are averages of measurements made separately from the basal and apex parts of the leaf lamella). The difference between the data pairs indicated by arrows is significant at the level of $P = 0.05$.

It was determined that the lower TNC content of the studied leaf was generally accompanied by slighter relative root growth in the short LDPs, however, the relative growth of the leaves was unchanged. That is, it is presumable that the carbohydrate flows from the source leaf firstly towards the growing leaves and the possible insufficiency hinders the growth of the root.

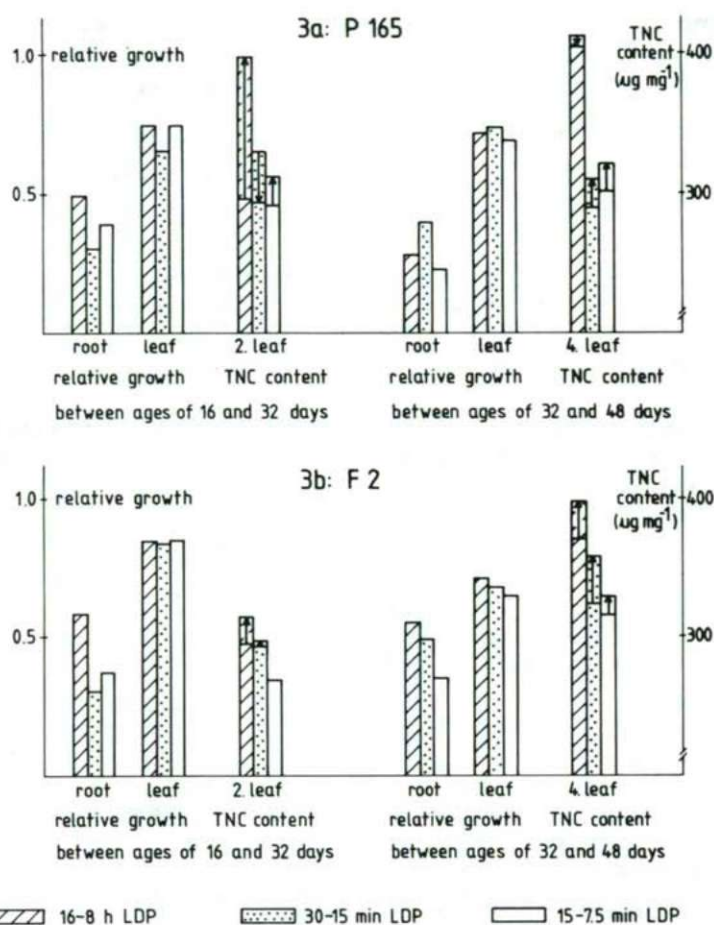


Fig. 3. Relative growth of the root and every leaf between the age of 16 and 32, as well as 32 and 48 days.

$$\text{Relative growth} = (W_2 - W_1) / W_1 \quad \text{where } W_1 = \text{dry mass measured at beginning period} \\ W_2 = \text{dry mass measured at end of period}$$

furthermore, the TNC (= soluble sugar + starch) content in the source leaf — 2. leaf between the age of 16 and 32 days and 4. leaf between 32 and 48 days of age — being the most characteristic of the two time periods. The direction and degree of the change in TNC content from the beginning till the end of the period is indicated by arrows. a: P165, b: F2.

Certain deviations from the foregoing could be experienced in the case of the P165 line in the 30-15 min. LDP: the high leaf TNC content and dry mass observed at the age of 16 days showed relatively slighter increase till 32 days of age, moreover there was a decrease in the TNC content (Fig. 3a). Reaching this age the carbohydrate flow from the seed had stopped and the earlier growth rate was not maintained by

the CO₂ assimilation. The relative growth (mainly of the root) was again found to be more intensive between the age of 32 and 48 days, but the TNC content of the 4. leaf was still low, but increasing in the given period, indicating the probably resatisfactory CO₂ fixation (Fig. 3a).

4. CARBOHYDRATE CONTENT IN THE BASAL AND APEX PARTS OF THE LEAF BLADE (Table 1)

There were firstly differences between the basal and apex parts of the blade of the studied maize leaves in the ratio of the soluble sugar and starch contents. The relative starch content was higher in the basic part of the leaf blade. Furthermore, the short LDP treatment was found to increase the relative starch content in general (cp. p8).

The relative starch content was higher in the case of the P165 maize in the 2. leaf while in the case of the F2 line, it was higher in the 4. leaf, particularly at the basal part.

Table 1.: Relative starch content in the basic and apex part of the studied leaves.

(Relative starch content = $\frac{\text{starch content}/\mu\text{g mg}^{-1}}{\text{TNC content}/\mu\text{g mg}^{-1}}$; LDP : Light-dark periods)

LEAF STAGE	LEAF AGE	LDP	P165			F2		
			BASE	APEX	DIFF.	BASE	APEX	DIFF.
2nd	developed	16-8 h	0.60	0.58	+0.02	0.61	0.58	+0.03
		30-15 min	0.69	0.71	-0.02	0.63	0.60	+0.03
		15-7.5 min	0.65	0.67	-0.02	0.61	0.61	0
	older	16-8 h	0.71	0.63	+0.08	0.65	0.58	+0.07
		30-15 min	0.74	0.70	+0.04	0.67	0.60	+0.07
		15-7.5 min	0.75	0.69	+0.06	0.66	0.59	+0.07
4th	developed	16-8 h	0.69	0.65	+0.04	0.78	0.61	+0.17
		30-15 min	0.70	0.60	+0.10	0.76	0.67	+0.09
		15-7.5 min	0.71	0.64	+0.07	0.74	0.65	+0.09
	older	16-8 h	0.68	0.58	+0.10	0.72	0.58	+0.14
		30-15 min	0.75	0.70	+0.05	0.78	0.64	+0.14
		15-7.5 min	0.72	0.69	+0.03	0.80	0.64	+0.16

Discussion

Our results proved the earlier observation according to which the P165 and F2 maize lines react differently to the short LDP treatment. The variancy-analysis demonstrated the interaction of the light-dark period and the genotype factor in the majority of the data on both dry mass and carbohydrate content. However, even in the case of the P165 maize more sensitive to the short LDP, several points were found where significant positive changes were detectable on the effect of the 30-15 min. LDP. (For example the starch content of the developed 2. leaf, the leaf-dry mass of the 16 days old plant). According to our assumptions, the different behaviour of the two maize lines at very young age is caused by the genetically varying transport and utilization of the seed reserves, later, however, the different photosynthesis of the leaves was determinative. The latter has been studied in detail by Pataki and Maróti (1985) in both maize lines with the finding that compared to the leaves of the F2 maize, in those of the P165 the quenching of the slow fluorescence is faster; the lag phase of the O₂ evolution is shorter; the ratio of the maximal oxygen evolution is higher; the violaxanthin de-epoxidation in the intrahylakoidal space is slower; furthermore the mesophyll chloroplasts contain higher amount of stroma membranes.

The comparison of the actual source leaf's TNC content and the growth showed that the relative growth of the leaves less depends on the carbohydrate content of the older leaves feeding them, however, that of the root is more dependent on this. The correlation is not on the whole unambiguous, other factors also play role. Such may be, for example, the enzyme function of the sucrose-phosphate-synthetase (HUBER et al. 1984), as well as the distribution of the inorganic phosphate in the cells (PREISS, 1982). It should be noted that (NAYLOR and GILES (1982) had also experienced the intensive growth of the young leaves of bean plants grown in short LDP, although these were almost completely dependent on the photosynthates of the older leaves, owing to the considerable chlorophyll destruction.

Since the earlier studies refer to the fact the chloroplast membranes of the plants grown in short LDP have become disorganized (MARÓTI and TAKÁCS, 1983); the pigment content of the leaves has decreased (MARÓTI, 1982), and the energization of the chloroplasts is of slighter degree (TAKÁCS et al. 1985); it is presumable that the plants fix less CO₂ within the same period, i.e. they make less use of the light received during the short periods. To compensate this, they need increasing the ratio of the photosynthetizing/nonphotosynthetizing organs, similarly to the plants grown under less light intensity (BJÖRKMAN, 1982). Perhaps this is why the plant ensures the growth of the leaves even on the account of the root.

It is also observed that where the dry mass of the root is more reduced by the short LDP treatment, the dry mass of the whole plant is less reduced, perhaps even increased: in both lines at the age of 16 days; in the F2 maize at the age of 32 days. Thus the reduced dry mass ratio of the root could be regarded as an effectual phenomenon of adaptation. The mechanism of regulation responsible for the development of this has not been known yet, nevertheless the carbohydrate level of

the leaf probably plays role. It seems that the appropriate TNC level of the leaf means a potential possibility for the growth, however, this possibility prevails through mechanisms of regulation known only imperfectly at present.

References

- AVIGAD, G. (1982): Sucrose-starch transformation. In *Plant Carbohydrates I.* (ed. by F. A. LOEWUS and W. TANNER). Springer Verlag, Berlin, Heidelberg, New York.
- BJÖRCKMAN, O. (1982): Responses to different quantum flux densities. pp. 57-75. In: *Physiological Plant Ecology II.* (ed. by P. S. NOBEL, C. B. OSMOND, H. ZIEGLER). Springer Verlag, Berlin, Heidelberg, New York.
- DUBOIS, M., GILLES, K. A., HAMILTON, J. K., ROBERTS, D. A. and SMITH, E. (1956): Colorimetric method for determination of sugars and related substances. — *Anal. Chem.* 28, 350-356.
- CHANG, C. V. (1980): Starch depletion and sugars in developing cotton leaves. — *Plant Physiol.* 65, 844-847.
- CHATTERTON, N. J. and SILVIUS, J. E. (1979): Photosynthate partitioning into starch in soybean leaves. — *Plant Physiol.* 64, 749-753.
- GENT, P. N. (1984): Carbohydrate level and growth of tomato plants. — *Plant Physiol.* 76, 694-699.
- HUBER, S. C. (1983): Relation between photosynthetic starch formation and dry weight partitioning between the shoot and root. — *Can. J. Bot.* 61, 2709-2716.
- HUBER, S. C., RUFTY, T. W. and KERR, P. S. (1984): Effect of photoperiod on photosynthate partitioning and diurnal rhythms in sucrose phosphate synthase activity in leaves of soybean and tobacco. — *Plant Physiol.* 75, 1080-1084.
- MARGÓCZI, K. (1984): A rövid fény-sötét periódusok hatása hibridkukoricák és szülői vonalaink morfológiájára és a levél szénhidrát tartalmának térbeli és időbeli megoszlására. (Effect of the short light-dark periods on the morphology of hybrid maizes and their parent lines, and on the spatial and temporal distribution of carbohydrates in the leaves. In Hungarian). — Ph.D. Thesis, Szeged.
- MARGÓCZI, K. and MARÓTI, I. (1985): Spatial distribution of carbohydrates in the leaves of maizes, grown in different light-dark periods. — *Acta Biol. Szeged.* 31, 87-96.
- MARÓTI, I. (1982): Effect of short light-dark cycles on the chlorophyll and carotenoid content of maize and tomatoes. — *Acta Biol. Szeged.* 28, 85-94.
- MARÓTI, I. and MARGÓCZI, K. (1984): Effect of the identical and alternating light-dark periods on the growth, dry matter production and carbohydrate content of maize leaves. — *Acta Biol. Szeged.* 30, 51-59.
- MARÓTI, I., MARGÓCZI, K., AL-SUBAI, M. Y., FÜLÖP, E., TAKÁCS, E. and MIHALIK, E. (1981): Effect of short periods of light and darkness on the histological structure of bean, mustard and pea. — *Acta Biol. Szeged.* 27, 117-126.
- MARÓTI, I. and MIHALIK, E. (1984): A rövid ritmusú megvilágítás hatása a kukoricák szárazsúlyára, oldható cukor, keményítő és cellulóz tartalmára. (Effect of the short rhythmical illumination on the dry weight, soluble sugar, starch and cellulose content of maizes. In Hungarian). — *Bot. Közlem.* 71, 117-186.
- MARÓTI, I. and TAKÁCS, E. (1983): Effect of the short periods of light on the membraneous system of corn mesophyll chloroplasts. — *Acta Biol. Szeged.* 29, 33-43.
- MAYORAL, M. L., ZVI, P. and REINHOLD, L. (1985): Effect of translocation-hindering procedures on source leaf photosynthesis in cucumber. — *Plant Physiol.* 77, 712-717.
- MOROT-GAUDRY, I. F., FARINEAU, J. and JOLIVET, E. (1979): Effect of leaf position and plant age on photosynthetic carbon metabolism in leaves of 8 and 16 day-old maize seedlings /W64A/. — *Photosynthetica* 13, 365-375.
- NAYLOR, A. W. and GILES, L. J. (1982): Growth, pigment synthesis and ultrastructural responses of *Phaseolus vulgaris* L. cv. Blue Lake to intermittent and flashing light. — *Plant Physiol.* 70, 257-263.
- PATAKY, SZ. and MARÓTI, I. (1985): Connection of different photosynthetic activity with the structure of chloroplasts. — Hungarian-Austrian Joint Conf. on Electron Microscopy, Balatonaliga p. 114.

- PHARR, D.M. and SAX, H.N.(1984): Changes in carbohydrate and enzyme levels during the sink to source transition of leaves of *Cucumis sativus* a stachyose translocator. — *Plant Sci. Lett.* 35, 187–193.
- PREISS, J.(1982): Regulation of the biosynthesis and degradation of starch. — *Ann. Rev. Plant Physiol.* 33, 431–454.
- SAGER, J.C. and GIGER, W.JR.(1980): Reevaluation of published data on the relative photosynthetic efficiency of intermittent and continuous light. — *Agric. Meteor.* 22, 289–302.
- SHARKEY, T.D., BERRY, J.A. and RASCHKE, K.(1985): Starch and sucrose synthesis in *Phaseolus vulgaris* as affected by light, CO₂ and abscisic acid. — *Plant Physiol.* 77, 617–620.
- SCHULZE, E.D., SCHILLING, K. and NAGAJARAH, S.(1983): Carbohydrate partitioning in relation to whole plant production and water use of *Vigna unguiculata* L. — *Oecologia*, 58, 169–177.
- SVÁB, J.(1981): Biometriai módszerek a kutatásban. (Biometrical methods in research. In Hungarian). — *Mezőgazd. K.* Budapest.
- TAKÁCS, E. and MARÓTI, I.(1984): Effect of the short light–dark cycles on the membrane system of bean chloroplasts. — *Acta Biol. Szeged.* 30, 61–73.
- TAKÁCS, E., TÉCSI, L. and MARÓTI, I.(1985): Energization of chloroplasts in palisade and spongy parenchymas. — *Hungarian–Austrian Joint Conference on Electron Microscopy, Balatonaliga*, p. 113.

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