SECTION 2 - PHOTOSYNTHESIS AND WATER RELATIONS

Plant responses to water stress

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Water stress is the most limiting environmental factor of growth of plants and agricultural productivity. Plant species or ecotypes are adapted to moist or dry localities, plants or their organs are acclimated to macro- and microclimatic conditions in which they grow, and plants respond to actual changes in air humidity or soil moisture. Different mechanisms were developed during plant evolution leading *a*) to avoidance of dangerous water deficit, and *b*) to tolerance of mild water deficit.

Water efflux is mainly controlled by the stomatal opening. The opening is regulated (feedback control) by the leaf water content. With the decrease of bulk leaf water potential below threshold value, the stomata close, even if other environmental conditions are optimal. In addition, two types of the feedforward control (the stomata are closed sooner than the mesophyll water potential is decreased) have been determined. The first of them is the direct response of stomata to air humidity which is associated with the peristomatal transpiration. The second one is response of stomata to the decrease of soil moisture and the rate of water absorption by roots (*e.g.* Jones 1990, Zhang *et al.* 1990, Meinzer and Grantz 1991). The changes in gradients of water potential (especially of pressure potential) between guard cells and other epidermal cells, between epidermis and mesophyll, *etc.* operate in all types of stomatal regulation. Even the decrease in pressure potential in root xylem due to soil water deficit can be rapidly transmitted (pressure changes in connected bodies of water are transferred at the speed of sound) through the xylem to leaves and through apoplast to neighbourhood of guard cells (Matsuda and Rayan 1989, Boyer and Nonami 1990). In addition, chemical regulation of transport of osmotically active substances (mainly K^+) through guard cell membranes or regulation of production of osmotically active substances in guard cells take part in the control of stomatal opening. In this regulation the most important is abscisic acid (ABA) and the root system can sense soil drying effectively by producing substantial amounts of ABA. Also further growth hormones, auxins and cytokinins (*e.g.* Mansfield and Atkinson 1990) or Ca^{2+} ions (*e.g.* McAinsh*et al.* 1990) can operate in conjunction with ABA. Water deficit, and in consequence the production of ABA, also modifies the response of stomata to other factors, *e.g.* increases the sensitivity to internal CO_2 concentration. Similarly stomatal response to blue radiation is decreased under low air humidity.

The decrease in transpiration rate is always accompanied with the decrease in CO_2 influx. According to the theory of Cowan and Farquhar (1977) the stomatal regulation maximizing daily photosynthetic CO_2 influx for a given H_2O efflux is considered to be "optimal". High water use efficiency is important in crop monocultures. For wild plants, relatively low CO_2 influx is often sufficient, even if restricted CO_2 influx due to water stress supports photoinhibition under high irradiance. C_4 pathway of CO_2 fixation is advantagenous for growth of plants under dry conditions, while under extremely dry conditions CAM predominates. In addition to decrease in CO_2 influx severe water stress also inhibited chloroplast activity. The mathematical models frequently used for distinguishing the stomatal and non-stomatal limitations to photosynthesis were based on the assumption that the stomatal conductance is the same over the whole leaf blade. However, under certain environmental conditions (also under water stress or application of ABA) heterogeneity in stomatal opening ("stomatal patchiness") was found (*e.g.* Raschke 1990). In conseqence, the stomatal limitation to photosynthesis is probably more important than it was assumed using the above mentioned models (*e.g.* Terashima *et al.* 1988).

Regulation of transpiration rate is significant also from the point of view of leaf energy balance in which an important role is played by leaf movements. The transpiration rate is also restricted by inhibition of shoot growth, eventually by fall of old leaves. Simultaneously, the root growth is much less affected by water stress and so absorption capacity of roots per leaf area unit is increased (*e.g.* Sharp 1990).

Growth and many other physiological processes are mostly affected by pressure potential. Thus for plant tolerance to water stress the "osmotic adjustment" is important. Water potential is decreased due to a decrease in osmotic potential (by influx of mineral ions or production and transport of osmotically active organic substances) and not by decrease in pressure potential. Also "elastic adjustment" was found (*e.g.* Richter and Kikuta 1989). The stable pressure potential during water stress is reached by the changes in cell wall elasticity.

Drought can induce the transcription of mRNAs coding of a large number of proteins which under well watered conditions are not synthesized. Among them osmotin and its derivatives are the best known (Singh *et al.* Takeda *et al.* 1991).

Since about 2000 papers dealing with plant water relations and relative topics are published every year (see Lösch 1991, Pospíšilová and Solárová 1992), only few chosen problems may be mentioned in this brief survey. From the new books *e.g.* Cherry (1989), Jones *et al,* (1989), Kreeb *et al.* (1989), and Davies and Jeffcoat (1990) can be recommended.

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Photosynthesis of the leaf and plant in a future, higher CO_2 atmosphere

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Anthropogenic increase of carbon dioxide content in atmosphere has manifold impact on individual plants, plant communities and the whole ecosystems (*e.g.* Bazzaz 1990, 1992). Serious impact on soil microbiota, on nitrogen fixing symbionts and rhizobia, and on herbivorous insects is expected. Thus, there is an increasing concern in plant response to higher CO_2 among plant ecologists, climatologists, soil microbiologists and entomologists. Changes in both crop and forest management are anticipated. The effect of increasing level of CO_2 in the atmosphere on physiology of carbon fixation and on closely related processes is summarized here.

In C_3 plants, the following processes were identified as the main physiological targets of the CO_2 increment: 1) short-term and long-term regulatory changes in carbon fixation, 2) changes in dark and light utilization of assimilates, sucrose or other assimilates allocation into expanding sink capacities, and 3) both direct effect of CO_2 and indirect effect of increasing assimilate pools on respiration. Consequently, changes in root/shoot ratio, increase in growth rate, rise in C/N ratio, enhancing quantity and changes in quality of root exudates and acceleration of plant development are observed. All these processes can be modulated by phosphorus or other nutrients availability (Harley and Sharkey 1991) as well as by concomitant global warming (Farrar and Williams 1991).

Appart from knowing the huge amount of experimental data, addressing plant response to higher CO_2, two basic responses can be postulated: (i) Reducing photosynthesis to the processes controlled largely by behaviour of single enzyme (ribulose-1,5,-bisphosphate carboxylase/oxygenase - 'rubisco'), an increase in photosynthesis rate can be expected. Under saturated irradiance (and, thus, under non-limiting regeneration of ribulose-1,5,-bisphosphate, RuBP) and non-limiting nutrients, the increase in net photosynthesis rate (P_N) should follow the Michaelis-Menten hyperbola known from enzyme kinetic theory. In the shaded plants, where the constraints by RuBP regeneration are expected, the increase in P_N can be explained by increasing carboxylation/oxygenation ratio as the substrate concentration for carboxylation rise. The underlying mechanism is inherent for rubisco the evolution of which occurred under at least ten-times higher CO_2 concentrations in atmosphere until cretaceous (Ehleringer *et al.* 1991). (ii) Viewing photosynthesis as a catena of many backward-regulated processes producing assimilates for plant growth and maintenance, one could find many negative feedback loops leading to complete plant insensitivity to increasing CO_2 in the extreme case; *e.g.* capacities for water uptake and transport can be insufficient and 'selfdamage' of expanding plant body in dry air could be envisaged: regulation of leaf internal CO_2 concentration by stomata closure mediated perhaps through photosynthate level is frequently observed: blocking of triose phosphate utilization can act as an end-product inhibition *etc.* Of course, combinations of many backward mechanisms are probable besides the hormonal direct action and regulation of gene expression. As a result, 'CO_2 acclimation' response (a long-term decrease in P_N below the value observed immediately after the CO_2 concentration has been risen) is often reported.

Stomata response to increasing CO_2: Usually, guard cells are regarded to be very sensitive to CO_2 through its inner cell side exposed into substomatal cavity (Mott 1990). CO_2 response of stomata combines probably two mechanisms opposing in their effect: first, inhibition of K^+ accumulation mediated by CO_2 effect on membrane permeability which is dominant mode of action, and second, supply of CO_2 as substrate for PEP-carboxylase producing malate in guard cells. The first mechanism results in stomata closure, the second one in stomata opening under increasing CO_2 levels. The dominancy results usually in stomata closure (Mansfield 1983). However, the analyses of relations among photosynthesis rate, stomatal conductance and internal CO_2 concentration as well as quantification of the CO_2-feeedback gain of stomata lead to conclusions about low (if any) sensitivity of stomata to CO_2 (Wong *et al.* 1985, Farquhar *et al.* 1978). Stomata should response more to photosynthate stimuli transferred via epidermis then to CO_2 change transmitted through intercellular gas phase.

Short-term CO_2 response in P_N and carboxylation: During several minutes after the CO_2 content was increased, dumping oscillatory response in P_N is usually observed followed by an increased steady state value in P_N (Sage *et al.* 1988). Higher steady state P_N reflects reduced competition from O_2 for rubisco and, thus, reduced photorespiration rate. Surprisingly, activation state of rubisco has dropped in the Sage's experiments. The regulation of rubisco activity is not a direct effect of higher CO_2, which could cause *e.g.* carbamylation of lysine in reaction centre resulting in activation of rubisco. The drop in activity probably reflects limitation in RuBP regeneration induced by defficiency of ATP in shaded plants or by phosphate defficiency due to restricted triose phosphate utilization (Sage *et al.* 1988).

Long-term CO_2 response in P_N and in utilization of assimilates: The initial mean increase in P_N, induced by doubling of CO_2 concentration in atmosphere, was calculated to be 52 % for C_3 plants. However, the P_N stimulation has dropped to 29 % in the average after a period of the high CO_2 acclimation (Cure and Acock 1986). Besides the above mentioned drop in activation state of rubisco, decrease in total rubisco concent was observed (Rowland-Bamford *et al.* 1991). Carboxylation rate transiently exceeds the rate of sucrose synthesis and export. Accumulation of starch grains in chloroplasts and chloroplast damage was also reported (Ehret and Jolliffe 1985). Appart from the widely accepted concent of the CO_2 acclimation, we can find the results of long-term (years) experiments which argue that the acclimation is an experimental artifact resulting from nutrient defficiency and from an inadequate space for the development of the root system (Ziska *et al.* 1990, Idso and Kimball 1991). Cure *et al.* (1987) did not find any acclimation response for soybean after three weeks of plant exposure to 700 $\mu l\ CO_2\ l^{-1}$. The rise in assimilates production was compensated by their utilization in dark after three days of exposition and then also by their utilization in light. Allocation into expanding sinks (root and stems) was observed in parallel.

The response of growth rate and of dry matter accumulation to elevated CO_2 is probably modified by the plant strategy. From 27 species native in British islands only competitors were highly sensitive to higher CO_2. The CO_2 effect decreased toward stress-tolerant or ruderal species (Hunt *et al.* 1991).

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Interrelationship between uptake of mineral ions and photosynthesis in submerged aquatic plants

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In four higher aquatic species with different ecological requirements for total alkalinity ("carbonate hardness") and different affinities to HCO_3^- use, uptake of six ion species by leafy shoots was studied as dependent on a high or a very low photosynthetic rate, *i.e.*, at a medium (7.5) or a high pH (10 or 9 or 8.8), and in darkness (pH 7.5). Apical segments (6-10 cm long) of *Callitriche hamulata* (growing in soft waters, HCO_3^- non-user), *Elodea canadensis* (wide range of waters, HCO_3^- user), *Ranunculus aquatilis* (soft waters, HCO_3^- user), and *R. trichophyllus* (hard waters, HCO_3^- user) were used. The uptake of HPO_4^{2-}, NO_3^-, Cl^-, K^+, Ca^{2+}, and Mg^{2+} was measured as a decrease in ion concentration of the solution (mM: $NaHCO_3$ - 1, $CaCl_2$ - 0.1, $MgSO_4$ - 0.1, KNO_3 - 0.3, Na_2HPO_4 - 0.25) in 2-h periods, after which the solution was partly renewed. The pH of the solution in a stirred chamber (80 cm^3) was kept constant (±-0.05), bubbling by gaseous CO_2 and CO_2-free air and a pH-stat were used. The species growing in hard waters (*E. canadensis* and *R.trichophyllus*) took up all the ion species at a higher rate at a low photosynthetic rate than the soft-water species (*C. hamulata, R. aquatilis*) did, it also held for HPO_4^{2-} and NO_3^- at a high photosynthesis. This difference in ion uptake at a high pH (very low uptake of carbon) could have an ecological significance when soft-water species compete strongly for inorganic carbon, especially in HCO_3^- non-users. In all species, the uptake of HPO_4^{2-}, NO_3^-, Cl^-, and Mg^{2+} did not depend markedly on the photosynthetic rate. The uptake of K^+ in all species was distinctly higher at a high photosynthesis and even a profound K^+ efflux from plants was found in darkness. Except of *C. hamulata*, a small uptake of Ca^{2+} prevailed at a high pH, whereas a Ca^{2+} efflux prevailed at the pH of 7.5 both in light and darkness. Obviously, the Ca^{2+} fluxes in both directions included mostly the apoplast ($CaCO_3$ incrustation of cell walls).

Effect of water stress and different mineral nutrition on plant water relations of cereals

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Effect of different stressors (polyethylenglycol, mannitol) was studied in laboratory experiments on young spring barley plants (*Hordeum vulgare* L.) and on some other cereals. Changes of plant water parameters are quantified (transpiration, water content, leaf diffusion resistance, water saturation deficit) in relation to the growth rates of the individual organs of the plant. Deficit of the main mineral elements (N,P,K) influenced more expressively the initiation and growth of the leaves, their active life duration. leaf diffusion conductance than N,P,K surplus. The possibilities for formulation of monitoring of the load levels by stressors through expeditive methods as well as possibilities for the elimination of negative ecological situations and stability of plant physiological processes are discussed.

Photosynthesis under conditions of *in vitro* culture

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Photosynthetic activity of plantlets grown autotrophically or mixotrophically on agar substrate *in vitro* is frequently limited by low carbon dioxide concentration and low irradiance during the light period, and affected by high air humidity, increased ethylene concentration, *etc*. The limitation of net photosynthetic rate (P_N) by low CO_2 concentration then leads to a considerable reduction of growth (Solárová 1989). Daily carbon gain (24 h) of autotrophic plantlets cultivated *in vitro* using a standard procedure decreased - in dependence on saccharose concentration in the medium - to 25 - 10 % of that in control plantlets cultivated under atmospheric CO_2 concentration (tobacco, carnation). CO_2 concentration in cultivation vessels for about 80 % of light period approached CO_2 compensation concentration (Γ) and P_N was near to zero. Considerable limitation to P_N resulted in a similar limitation to growth rate measured as the rate of dry matter accumulation. In spite of very low P_N in cultures *in vitro* (measured *in situ*) the photosynthetic apparatus was fully developed. The limitation to P_N by CO_2 deficiency was confirmed also by a 200 % increase in dry matter accumulation after increasing CO_2 concentration in cultivation vessels. The difference in dry matter accumulation between plantlets grown under atmospheric and increased CO_2 concentration was more pronounced on medium without saccharides. Limitations to photosynthesis under conditions *in vitro* were studied in model experiments: Environmental conditions in cultivation vessels were simulated by growing French bean (*Phaseolus vulgaris* L.) plants in nutrient solution in closed vessels under high relative humidity of air (90%) and CO_2 concentration decreasing with the development of photosynthetic activities during plant ontogeny (1200 to 300 mg m^{-3}). The ontogeny of model plants was more rapid, primary leaves reached photosynthetic maturity 2 to 3 d earlier and their life span was 7 to 14 d shorter. Photosynthetic activity *in situ* of these plants was limited, after reaching "photosynthetic maturity", similarly to plants grown *in vitro*. When measured *ex vitro*, however, 50 to 70 % higher P_N was found in leaves of different ages as compared with plants grown *in vitro* under atmospheric CO_2 concentration (600 mg m^{-3}) and lower relative humidity of air (40 to 60 %). The increase in P_N was connected with a very high epidermal conductance for CO_2 transfer and with low reactivity of stomata. The rates of dark and light respiration as well as CO_2 compensation concentration were 10 to 30 % lower in plants grown under conditions simulating the culture *in vitro*.

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Stomatal behaviour of an invasive annual (*Impatiens parviflora* DC.) in natural habitats, SW Slovakia

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Impatiens parviflora is an erect annual of Central-Asiatic origin, introduced into Europe in the 19th century, and now invading forest communities in Central Europe. It was considered as a shade-tolerant plant or as a facultative sun plant, being an extremely phenotypically plastic species. In summer months (July, August) of 1984-1987, stomatal resistance (r_S) was measured by *Li-Cor* porometer, in course of several sunny days, in plants growing in some *Impatiens* populations near Bratislava (Malé Karpaty Mts: Železná Studienka, Biely Kríž) and at Báb (formely I.B.P. Forest Research Site), SW Slovakia. The *Impatiens* populations in forests, at forest margins (edges) and on clear-cut areas differed by size (height, biomass) and architecture of plants but also in density, size and arrangement of stomata (hypostomatic *versus* amphistomatic leaves). Stomatal resistance of the *Impatiens* species varied predominantly in relation to light conditions of the habitats (sunny places, light-flecks, semi-shade, deep shade) and water-saturation status of the leaves. In all habitats daily r_S-curves were close to the U-type-curves but r_S-values were usually lower in plants growing at forest margins than in forest-growing plants. In a forest stand, noon values of r_S varied around ca 5 s cm^{-1}, while at forest margins they were usually lower than 4 s cm^{-1}. The relationship between stomatal conductance (k_S) and photosynthetically active radiation (PhAR, $\mu mol\ m^{-2}\ s^{-1}$) was analysed by the boundary-line method. It was found that at similar PhAR values, higher k_S occurred in plants growing at forest margins (maximum k_S - values were higher than 0.5 cm s^{-1}), in comparison with plants growing in the forest stand (k_S max. *ca.* 0.25 cm s^{-1}). In hot summer days (drought, high temperature) plants growing in sunny places (forest margins, woodland clearings) were stressed and higher r_S - values were found than in forest-growing plants.

Stomatal conductance and transpiration rate of tall poplar trees in a flood-plain forest by the Danube river, SW Slovakia

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In an 11-year-old and 28.6 m high (in average) stand of poplar (*Populus euramericana*, cv. Robusta) at Gabčíkovo (locality Královská lúka, SW Slovakia), transpiration rate (TR) and stomatal conductance (g_S) were measured by LI-1600 Steady State Porometer in crowns of tall trees in the courses of 10 d from July to October 1991. To measure the TR and g_S in crowns of the trees, a 30 m high steel-tube tower was built up around the sample poplar tree ($d_{1,3}$ = 30.7 cm, h = 31.5 m, total leaf area 249 m^2). Variations in measured parameters within the crown were analysed in relation to aspects (S, E, N, W), levels (upper, lower), current-shoot types (long, short) and sides of amphistomatic leaves (abaxial, adaxial). Maximum TR and g_S was usually found in the upper crown level, west (July to August) and south (August-September) aspects and in the long shoots, and in abaxial epidermis (lower side of a leaf), and it was usually higher than 10 $\mu g\ cm^{-2}\ s^{-1}$ and 12.4 $mm\ s^{-1}$, respectively. In the lower crown levels, TR and g_S were usually lower than 5 $\mu g\ cm^{-2}\ s^{-1}$ and 7.5 $mm\ s^{-1}$, respectively. Hourly values of transpiration per tree increased up to 80 l (at noon) and daily values of transpiration per tree varied, in summer, between 256 and 661 l of H_2O. It is evident that the annual summ of transpiration of the poplar stand (stand density was 247 trees per ha) is higher than 1200 mm H_2O. The measurements have been a part of monitoring of the forest environment changes induced by construction of the Gabčíkovo Dam on the River Danube. The results will be used by the Centre of Environment Monitoring at the Slovak Commission of Environment (SKŽP) in Bratislava.

Stomatal regulation of the winter wheat transpiration daily course

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It follows from Penman-Monteith equation for transpiration, that transpiration rate (E) falls sharply as stomatal resistance (r_L) increases, if atmospheric conditions and soil moisture content are kept constant. There is, however, a lot of experimental data in literature showing that daily maxima of E do not correspond to daily maxima of r_L. The aim of the paper is to analyse and support this paradox using a verified mathematical model of winter wheat water regime. Daily courses of r_L for low and high air evaporative demands (water vapour pressure deficit maximum during the day 1000 and 2000 Pa) were analysed. It follows from the analysis, that the daily course of r_L at nonlimiting soil moisture content is simple, with one minimum between 9.00 and 10.00 a.m. If we decrease soil moisture content during the simulation, a strong local maximum of r_L daily course appears at about midday. In spite of this, transpiration is maximal at midday as well. Such situation occurs, because the relationship between E and r_L is not functional. From 6.00 to 9.00 a.m. E falls as r_L increases, but later it increases. The increase of r_L is accompanied by sharp increase in leaf water potential absolute value, which causes the maximum E to be at midday. It also follows from the analysis, that changes in leaf water potential depend on changes in stomatal resistance and not *vice versa*.

Daily course differences of transpiration rate for soybean cultivars

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The transpiration rate (TR) was measured for 6 soybean cultivars (Dunajka, Sluna, Zora, Aida, McCall, Maple Arrow) in R-5 stage during summer sunlight day in field conditions by Steady-State porometer *LI-1600*. Values of PhAR, leaf and air temperatures and stomatal resistance were recorded simultaneously. Significant differences of water relations were obtained for tested cultivars. TR of Canadian cultivars were lower with smaller variations during day time than TR of the domestic one. All domestic cultivars showed the first highest maximum TR between 11-12 h a.m. Afternoon inhibition TR differ for individual cultivars. Cultivars Dunajka, Sluna and Zora were strongly inhibited (up to 40 % of maximum), whereas Aida, McCall and Maple Arrow were inhibited only a little. The second TR maximum was recorded at 16:30 or 17:30 h with high peaks for cultivars McCall, Sluna and Zora. Cultivars Aida, Dunajka and Maple Arrow had lower second peak of TR. These cultivars have very low values of stomatal resistances with small fluctuation amplitudes. The lowest alternations of leaf water content were obtained in cv. Dunajka and Sluna. Leaf temperatures were 1-2 °C higher than air temperatures during day, after 17:00 h leaf temperature was lower than the air temperature.

Chlorophyll content, CO_2-exchange and plastid structure of the saprophyte *Neottia nidus-avis* (L.) L.C. Rich.

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Neottia nidus-avis has been attractive for botanists since the last century because of its special appearance and biology. It is especially known for the rapid colour change of the inflorescence from brown to green following immersion in methanol or hot water. Stomata are reported to only be found on tepals. Our observations confirmed the presence of stomata on these same organs and then only in very low frequency. Some early work claims that this plant has low photosynthesis at low irradiance, however, a subsequent study questions this conclusion (Reznik 1958). Our study showed that whole plant photosynthesis actually reached negative values and the dark respiration also was low. In both normal (brown) and albino forms no CO_2 fixation occurred. These findings, together with the observation that both forms display essentially identical vitality led to the conclusion that *Neottia nidus-avis* is a true holosaprophyte (Reznik 1958). However, the same and other authors found only chlorophyll a in this species. In our measurements we also found chlorophyll *a* (0.7388 mg g^{-1}) but in addition, a very low level of chlorophyll *b* (0.058 mg g^{-1}) was detected. Structural studies of this species reveal several interesting features. Reznik (1959) observed the presence of starch in both the brown and the white forms of *Neottia*. Later observations demonstrated spindle-like plastids in tepals. EM studies showed coiled and branched thylakoids, plastoglobuli as well as starch in flowers and stalks. Our observations confirmed all of these structures in plastids of tepals. But apart from this, we found another type of unusual plastid in stalk scales. These plastids contained dark (electron dense) bodies which were penetrated by regularly distributed tubes. The function of these newly discovered structures is, as yet, unknown.

Reference:
Reznik, H.: Planta 51:694-704, 1958.

Leaf photosynthetical capacity of high-productive hybrids of maize (*Zea mays* L.) grown under field conditions during generative stage of ontogeny

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The place of maize leaf photosynthesis in productional process and its improvement by means of selection and breeding is a frequently discussed problem which is closely connected with knowledge of photosynthetical potentionality. Therefore in the years 1986-1988 we decided to analyse a term of high and low net photosynthetical rate (P_N) through the measurement of photosynthetical capacity (PC) on the middle part of leaves in the positions -2, +1, +4 (0 = the nearest leaf to the main ear). The selected maize hybrids (CE 185, DEA, CE 420) were grown in two different densities (84 000 and 119 000 plants per ha). The PC of intact leaves was measured in 3 cycles with an open gas exchange system with IRGA under constant laboratory conditions (photosynthetical photon flux density-PPFD-varied from 210 to 2130 μmol m^{-2} s^{-1}) during the generative stage of ontogeny. In another experiment in 1991, we measured the actual P_N of the middle part of leaves in the position +1 of the hybrid TA-TO 260 S by *LI-6000* in field conditions (3 densities, 3 levels of applied N, with and without irrigation). From the first experiment it resulted that PC of the measured leaves (all hybrids) in both densities was high and comparable. The highest value of P_N in the first experiment was 2.263 mg (CO_2) m^{-2} s^{-1} (PPFD 2130 μmol m^{-2} s^{-1}) and that in the 1991 experiment was 2.067 mg (CO_2) m^{-2} s^{-1} (PPFD 1725 μmol m^{-2} s^{-1}). In the first experiment, the term "high and low P_N" in the productional process is specified by means of "light curves" of photosynthesis. Comparison of the maximum P_N values in our experiments with those indicated in foreign works and measured under adequate conditions shows that the current high-productive hybrids probably do not differ in potential possibilities of P_N.

Water potential of styles and unilateral interspecific incompatibility in *Solanaceae*

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In unilateral interspecific incompatibility, the pollen tubes of self-incompatible (SI) species grow through the styles of self-compatible (SC) species, while in reciprocal crosses the growth of pollen tubes of SC species is inhibited in the styles of SI species. The mechanism of these relations in the family *Solanaceae* was studied from the aspect of possible role of stylar water potential. Elongation growth of pollen tubes in intercellular spaces of transmitting tissues of styles requires a relatively large water uptake and is thus dependent on water potential gradient between the growing tube zone and the stylar tissue. In the SI apecies *Nicotiana alata* and *Petunia hybrida* the stylar water potential (measured by *Wescor Dew-Point Hygrometer*) was considerably lower and at the same time also the stylar water content (in % of fresh mass) was lower in comparison with the water potential and water content in the styles of SC species *Nicotiana sylvestris* and *Nicotiana tabacum*. During ontogeny of flower buds the stylar water potential in *N. alata* decreased more expressively than in *N. tabacum*. The pollen tubes of SI and SC species exhibited different requirements for concentration of osmotica (saccharose or polyethylene glycol) in medium during *in vitro* germination. The optimum growth *in vitro* of pollen tubes of *N. alata* and *P. hybrida* was achieved at lower water potential of medium than the optimal growth of pollen tubes of *N. sylvestris* and *N. tabacum*. The higher peroxidase activity determined in transmitting tissue of styles in SI species in comparison with SC species might be also connected with the lower stylar water potential in SI species than in SC species. Therefore one of the unspecific mechanisms of incompatibility in *Solanaceae* might consist in growth inhibition of pollen tubes of SC species in too "dry" transmitting tissue in the styles of SI species.

The direct effect of abscisic acid on photosynthesis is not an artifact

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The non-stomatal effect of abscisic acid (ABA) was established in *in vitro* cultivated plants under premise that ABA is not able to close stomata of *in vitro* cultivated plants. *In vitro* tobacco plants (*Nicotiana tabacum* cv. White Burley) cultivated on MS agar medium without phytohormones and sucrose in glass vessels covered with alluminium foil were used for experiments. The plants were cultivated under constant temperature 20 °C and 12 h photoperiod for 2 months. Photosynthesis (P_N) and transpiration (E) rates of adaxial side of the detached leaves were measured using an adapted *LI-6200* photosynthesis system (*LI-COR*, USA) with 250 cm^3 leaf chamber. Simultaneous leaf surface temperature measurement were done by IR camera (*NEC-SAN EI 6T62*, Japan) hermetically connected to the leaf chamber by a cylindrical glass adapter. The leaf exposed in the dark for 15-30 min was then illuminated. The kinetics of light induction of photosynthesis was typical for *in vitro* cultivated plants: P_N reached a maximum 8 to 15 min after switching the light on, then the constant or slightly decreasing P_N was observed for 30 to 50 min. In contrast to plants adapted to atmospheric conditions, plants growing *in vitro* showed nearly constant E for darkened and illuminated leaf. Thus, the kinetics of P_N reflected probably the process of light induction of the enzymes of Calvin cycle and was not modulated by stomata opening. The leaf surface temperature measured by IR camera in dark ranged in the central part of the leaf from 20.2 °C to 20.5 °C, after switching the light on, a non-uniform temperature increase was observed (the increase ranged from 2.4 to 3.4°C). Feeding with ABA (5 µl 10^{-2} M ABA solution into petiole of the leaf) has only very small effect on E as well as on leaf surface temperature (E increased for about 5% during 3 min following the ABA application and then fell down steadily to 82 % of its original value during the next 25 min). Temperature average over the leaf surface slightly decreased. In contrast to E and temperature, P_N showed dramatic transient fall followed by recovery to 70 % of the initial P_N values. Where the feeding with ABA was continuous, P_N fell down to zero without recovery. The results support the hypothesis, that stomata of *in vitro* cultivated plants are not sensitive to ABA and that ABA affects photosynthesis directly, not only through stomata closure. Mechanism of this effect is not known at present.

Response of photosynthetic apparatus to periodic CO_2 deficiency in two potato cultivars

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Two potato cultivars (*Solanum tuberosum* L., cvs. Kennebec and Jaerla) differ in their ability to survive a transfer from *in vitro* to normal *ex vitro* cultivation. Cultivar Kennebec (cv. K) acclimatizes very well while cv. Jaerla (cv. J) exhibits a great percentage of mortality. The periodically fluctuating CO_2 concentration in glass vessels during the light and dark periods was previously hypothesised (Šantrůček *et al.* 1991) to cause changes in radiant energy utilization. The explants were grown autotrophically in glass vessels covered with an alluminium foil for one month at 20 °C under photon fluence rate of 60 - 80 µmol m^{-2} s^{-1} during 12 h photoperiod. Then the vessels were hermetically closed. For 11 d, simultaneous measurements of CO_2 exchange and of chlorophyll fluorescence emission were made using *LI-6250* IR gas analyzer (*LI-COR*, USA) and *PAM-fluorimeter* (*H. Waltz*, Germany). The effect of O_2-free atmosphere was investigated. The inhibitory effect (IE) of O_2 on CO_2 fixation was very high (23-67 % of the photosynthetic rate in O_2-free atmosphere) immediately after the closing. It was valid for both CO_2 concentrations under which IE was investigated (close to Γ and 450 cm^3 m^{-3}). The CO_2 starvation resulted in the flattened CO_2 response of net photosynthetic rate (P_N), in an increase of CO_2 compensation concentration (Γ) and in a quick decrease (on the 5th d) and even in an elimination (on the 11th d) of O_2 effect measured at higher CO_2 concentration. The drop in IE was quicker then the depression in P_N. On the contrary, IE estimated at CO_2 concentrations close to Γ increased after the full stop of CO_2 supply. The fluorescence parameters Φ_e (the quantum yield of PS2 electron transport), q_0, q_E and q_I (the photochemical, non-photochemical and photoinhibitory quenching, respectively) were used to monitor differences between both cultivars in their photosynthetic activity. From the same starting value of Φ_e=0.52 for both cultivars decreased Φ_e on the 5th d to 0.38 (cv. K) and to 0.26 (cv. J). A higher reduction of Φ_e in cv. J brought about a decrease in q_0 (on the 11th day of 0.68 cv. J and of 0.86 cv. K). There was observed a significant difference in q_E (in cv. J every time higher by 20-30 %) and large decrease in q_I after the 5th d (from *ca.* 0.75 to 0.53). The adaptation mechanisms (*e.g.* fotorespiration and fotoinhibition) enable the plants to dissipate non-destructively excitation energy absorbed. Under the CO_2 deficiency, these mechanisms reflect the higher surviving ability of cv. Kennebec.

Reference:

Šantrůček, J., Šiffel, P., Polišenská, M., Pešková, H.: Photosynthetica 25: 203-208, 1991.

Effect of water and temperature stresses on some cereals

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Water deficit at the end of the stem elongation phase caused changes in flag leaf chloroplasts, influenced gradual formation of sclerenchymatous tissues and thickening of leaf epidermis. The diameter of tracheids was decreased. Water and temperature stresses during first phase of grain mass accumulation reduced the grain mass, during the second phase the grain mass reduction was depended on the species and cultivars and reached 20-35 %, and in the third phase the reduction was 5-12 %. The average maximal grain (sink) mass was decreased from 42.6 to 37.5 mg in barley and from 49.2 to 43.6 mg in winter wheat. The maximum sink size was in barley ear on the 8th and 9th grain and in winter wheat on the 7th and 8th spike. The liner correlation coefficient between ear length and ear grain number was decreased from 0.851 to 0.488 in water stressed plants closely before anthesis and repeated stress after anthesis reduced this correlation coefficient to 0.425.

The effect of global radiation intensity and water vapour pressure deficits on soybean (*Glycine max* (L.) Merr.) water regime characteristics

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Using a mathematical model, the relationship between meteorological factors and soybean water regime characteristics (leaf water potential, canopy resistance, actual transpiration to potential

transpiration ratio) was evaluated. Results after three years of measurements of leaf water potential, stomatal resistance and global radiation were used to find values of parameters in the model. Aim of this work was to describe the effect of water vapour pressure deficit and irradiance on soybean water regime. It was shown, that water vapour pressure deficit is an important ecological factor. Its relationship to canopy resistance is quadratic, and therefore it significantly affects soybean canopy water regime. Analysis of the relationship between water vapour pressure deficit and transpiration rate has shown that maximal transpiration rates were observed at water vapour pressure deficits 1000-1500 Pa. At lower or higher deficits transpiration rate falls and it cannot be changed by changing soil water content. Irradiance affects canopy or stomatal resistance differently at different soil water contents. In wet soil (soil moisture 30 %) stomatal resistance decreases with increasing irradiance, as expected. In a dry soil (soil moisture 15 %) stomatal resistance increases in parallel with irradiance.

Photosynthetic rate at different temperatures in relation to cold acclimation

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Plants of winter wheat (*Triticum aestivum* L., cv. Mara) were grown in Hoagland 3 nutrient solution in controlled conditions. The photoperiod was 14 h, with photon flux density of 300 μmol^{-2} s^{-1}, at a day/night temperature of 15/12 °C. When the area of the second leaf was fully developed, plants were transferred to 5 °C for cold acclimation. Other cultivation conditions were identical as described above. The CO_2 gas exchange rate was measured by the infra-red CO_2 analyzer *URAS 3G* in an open gas exchange system, the rate of transpiration was measured by a psychrometer. The CO_2 exchange rates were measured at the photon flux density of 500 μmol^{-2} s^{-1}, the leaf temperature of 5 °C and 20 °C, the O_2 concentration of 21 % and at the outlet concentration of 350 $cm^3(CO_2)$ m^{-3}. The aim of these measurements was to find out the time necessary for acclimated and nonacclimated plants to reach the photosynthetic steady-state rate at different temperatures. The plants were taken from the growth cabinet in dark period and ten attached fully developed second leaves were placed into the assimilation chamber for dark adaptation (60 min at 5 °C, 20 min at 20 °C). The nonacclimated plants reached the photosynthetic steady-state rate after 60 min of the light period at 5 °C (10 μmol^{-2} s^{-1}) and during the next 6 h no changes were found. The plants measured at 20 °C reached the photosynthetic steady-state rate in 20-40 min (19.5 μmol^{-2} s^{-1}). As the period of cold acclimation increased the time necessary for reaching the photosynthetic steady-state rate also went up. After 3-4 weeks of cold acclimation this time increased to 3 h, when measured at 5 °C (7.5 μmol^{-2} s^{-1}) and to 7 h, when measured at 20 °C (15.5 μmol^{-2} s^{-1}). In measurements at 20 °C the rates changed in the following way: after 1 h 21 %, after 2 h 43 %, after 3 h 64% of the photosynthetic steady state-rate. The prolonged time period necessary for the stabilization of the photosynthetic rate of cold acclimated plants measured at 5 °C is probably due to assimilate accumulation and due to increased viscosity in intracellular space. At the same time a two fold increase in dry mass per unit leaf area was found. Acclimated plants measured at 20 °C were affected in the same way, but also at the same time the increased temperature resulted in deacclimation. There is no problem when we measure the plants at the same temperature, as that in which they are cultivated, but if the temperature during measurement is different from cultivation conditions the plants are found to have adapted to the altered conditions. The adaptation of cold acclimated plants (at 5 °C) to 20 °C in the assimilation chamber is much faster than the adaptation of nonacclimatized plants (at 15 ° C) to 5 °C.

Responses of stomata to ABA, IAA and carbon dioxide in isolated epidermis of *Vicia faba* L.

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Stomatal regulation is controlled by external and internal factors such as light, temperature, humidity, K^+ concentration, some phytohormones, *etc.* Contradictious views on the effect of phytohormones and their interactions expecially under different ambient conditions still exist. The effects of abscisic acid inducing the closure of stomata and β-indolyl-3-acetic acid inducing their opening are well known. Currently, ABA/CO_2, IAA/CO_2 and ABA+IAA/CO_2 relations still remain an open question. All experiments were made by the method of isolated abaxial epidermal strips incubated in standard medium of 10 mol m^{-3} MES buffer pH 6.15 in constant conditions (temperature 25 ± 1 °C, photon flux density 120 μmol^{-2} s^{-1}, relative humidity 98%) in the presence (350 μmol mol^{-1}) or absence of CO_2 during 3 h illumination. KCl (50 or 100 mol m^{-3}), ABA and IAA (10^{-5} to 1 mol m^{-3}) were incorporated singly or in combination into buffer. Epidermal strips being incubated in the buffer alone showed the stomatal apertures to be significantly smaller in the presence of CO_2, it acted as inhibitory agent on stomatal apertures. When CO_2 interacted with ABA stomata remained more open and the closing effect of CO_2 did not appear. ABA-induced closure of stomata was thus diminished. IAA stimulated stomatal opening; in CO_2 presence stomatal apertures were decreased in contrary to its interaction with ABA. The closing effect of CO_2 remained maintained but IAA was able to decrease it. At the simultaneous action of both phytohormones IAA limited the inhibitory effect of ABA but the way of interaction between CO_2 and ABA in the presence of IAA in incubation medium was maintained and its counteracting influence against ABA did not appear. The experiments revealed the existence of a complicated interaction among used phytohormones and CO_2, the possibility of changing in the way of these interactions by means of agent combinations as well as the fact that phytohormones were important regulators of the stomatal activity.

The changes of water relations and photosynthesis of barley after abscisic acid application

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Abscisic acid (ABA) was added to the hydroponic solution in total concentration of 10^{-4} M to the hydroponically grown young (14 - 28 d old) spring barley plants (*Hordeum vulgare* L.). Changes of leaf diffusion conductance, transpiration and photosynthesis rates were measured by *LICOR-6200* as well as water uptake and water content in individual plant organs. The onset of ABA starts immediately after addition with slight increase of P_N and conductance values followed by their decrease due to stomata closing. Individual cultivars react to ABA treatment differently. The most stabilized values had shown cv. Bonus, though its total decrease of P_N and conductance values was up to 40 % of its maximal values. Dwarf cultivar KM-341 reacts intensively to the ABA treatment.

Influence of water regime on the realisation of yield potential and on the length of potato vegetation period (*Solanum tuberosum L.*)

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The production of biomass and in this connection also the production of plants is very often limited by the water deficit. Concretely in potatoes it is one of the factors, which limit mostly the yield. Its lowering appears in three ways. It reduces the dry matter production directly and by means of the lowering of the rate of photosynthesis. It limits the development of the crop (the size of leaves) and by this indirectly the dry matter production. It accerelates also the senescence of the crop. The study of water regime was carried out in controlled conditions of pot experiments in greenhouse. For the experiment we chose 4 varieties with different length of growth period: Resy - very early, Radka - medium early, Désirée - medium late, Kamýk - late. During the vegetation period we submitted in different growing phases a part of plants to "a middle" (50% RSM - relative soil moisture) and to "a sharp" (35% RSH) stress. The drought delayed the emergence directly in proportion to its intensity. It lengthened growth phases and with this also the total length of vegetation period, if the water stress functioned at the beginning and in the further course of vegetation. On the contrary it shortened the vegetation period, if the drought took place at the end of vegetation period before the entrance into the physiological maturity of the crop. The drought at the beginning of vegetation period positively influenced the height of plants and the leaf area. It confirmed our previous estimation, that plants adapted to the drought in the phase of emergence are most productive. The water deficit in soil during the vegetation period caused more rapid old growing of haulm with the rate of photosynthesis proportionally to its height. The highest yields of tubers was estimated in variants with 50 and 35 % RSH in the phase of planting to the emergence.