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Adaptation of the Photosynthetic Apparatus in Maize Leaves as a Result of Nitrogen Limitation[1]

Relationships between Electron Transport and Carbon Assimilation

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ABSTRACT

In maize (*Zea mays* L., cv Contessa), nitrogen (NO_3^-) limitation resulted in a reduction in shoot growth and photosynthetic capacity and in an increase in the leaf zeaxanthin contents. Nitrogen deficiency had only a small effect on the quantum yield of CO_2 assimilation but a large effect on the light-saturated rate of photosynthesis. Linear relationships persisted between the quantum yield of CO_2 assimilation and that of photosystem II photochemistry in all circumstances. At high irradiances, large differences in photochemical quenching and nonphotochemical quenching of Chl a fluorescence as well as the ratio of variable to maximal fluorescence (Fv/Fm) were apparent between nitrogen-deficient plants and nitrogen-replete controls, whereas at low irradiances these parameters were comparable in all plants. Light intensity-dependent increases in nonphotochemical quenching were greatest in nitrogen-deficient plants as were the decreases in Fv/Fm ratio. In nitrogen-deficient plants, photochemical quenching decreased with increasing irradiance but remained higher than in controls at high irradiances. Thermal dissipative processes were enhanced as a result of nitrogen deficiency (nonphotochemical quenching was elevated and Fv/Fm was lowered) allowing PSII to remain relatively oxidised even when carbon metabolism was limited via nitrogen limitation.

Strong positive correlations have been found between the photosynthetic capacity of leaves and their nitrogen content, most of which is used for synthesis of components of the photosynthetic apparatus (1, 6, 23, 26). Indeed, the availability of nitrogen limits growth in most environments but the restricted development of nitrogen-deficient plants is usually due to a lower rate of leaf expansion rather than a decline in rate of photosynthesis per unit leaf area (22). Evans and Terashima (6) found that the photosynthetic properties of spinach thylakoid membranes were virtually independent of nitrogen treatments. In this case, nitrogen nutrition affected the amount of thylakoids per unit leaf area but not the properties of the membranes (6, 27). In contrast, studies with other species have found decreases in the light-saturated rate of photosynthesis (15, 17, 23, 26, 27, 30), and the incident quantum yield (17) when nitrate was limiting. In developing maize leaves nitrogen deficiency has been found to result in a significant decrease in photosynthesis with a selective reduction in the levels of phosphoenolpyruvate carboxylase, pyruvate orthophosphate dikinase, and ribulose 1,5-bisphosphate carboxylase and a concomitant decrease in level of their respective mRNAs (26). Nitrogen-limited *Chlamydomonas* cells were found to have a 70% reduction in the Chl content and in this case the Chl *a*/*b* ratio increased as a result of N-limitation (19). Nitrogen-limited plants also accumulate large amounts of carotenoids (12, 19). Shade-grown plants grown with limiting nitrogen have been found to be more susceptible to photoinhibition than nitrogen-replete controls (8, 25). Damage to the PSII reaction center occurs when the absorption of excitation energy exceeds the capacity for dissipation (9). The extent of photoinhibitory damage may be viewed as the net difference between the rate of damage and the rate of repair or recovery (13). In circumstances of nitrogen limitation it is probable that it is the repair processes which are limited (8, 13, 26).

The yield of Chl *a* fluorescence from photosynthetic organisms is determined by two distinct quenching processes, qQ[2], which is due to operation of photosynthetic electron transport, and qNP (2, 9, 11, 14, 20, 24). Nonphotochemical quenching arises from nonradiative dissipative processes that regulate the quantum yield of PSII (14, 28), and prevent damage to the photosynthetic apparatus by excess light. qNP is composed of at least three types of quenching processes which function in the protection of the membranes and also in the regulation of PSII photochemistry. These are: (a) the thermal dissipation process activated by the formation of ΔpH known as qE (3), (b) the regulation of the absorption cross section via the state transition, qT (29), and (c) the thermal dissipation process correlated with the conversion of violaxanthin to zeaxanthin,

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[2] Abbreviations: qQ, photochemical quenching coefficient; ϕCO_2, quantum efficiency of CO_2 assimilation; ϕPSII, quantum efficiency of PSII photochemistry; Fv/Fm, efficiency of open PSII centers; Q_A, primary acceptor of PSII; qE, thermal dissipation associated with the formation of ΔpH; qI, thermal dissipation due several processes including the xanthophyll cycle; qNP, nonphotochemical quenching coefficient; qT, radiationless dissipation due to the state transition.

Table I. *Shoot Characteristics of 3 Week Old Maize*

Plants, without endosperm, were grown for 2 weeks on sand periodically watered with a complete nutrient solution containing various concentrations of nitrate.

Treatment	Shoot Mass	Chl	Net photosynthesis	NO_3^- Concentration	Reduced N
mM	*g dry wt plant^{-1}*	*mg g^{-1} fresh wt*	*μmol h^{-1} mg^{-1} Chl*	*μmol g^{-1} dry wt*	*mg g^{-1} dry wt*
12.0	0.106	2.47	78	700	38.9
0.8	0.098	2.38	82	30	39.0
0.2	0.061	1.66	114	20	28.2
0.05	0.045	1.04	156	10	17.1

qI (4, 5, 18). These energy dissipation processes, which are mediated largely at the level of the antenna complexes, result in a pronounced increase in the rate of radiationless dissipation of the antenna Chl. As a result, they compete effectively with excitation energy transfer processes to the PSII, leading to reduced quantum yield.

Nitrogen-(NO_3^-) nutrition influences photosynthesis in three main ways. (a) Nitrogen deficiency may affect the quantity, structure and composition of the photosynthetic apparatus since this accounts for most of the nitrogen in the leaf (6, 27). (b) Assimilation of NO_3^- by the cell requires the NAD(P)H, reduced ferredoxin, ATP, and carbon chains produced by photosynthesis, so that NO_3^- is in competition with CO_2 for assimilatory power. (c) Accumulated NO_3^- may alter chloroplast function by way of modulation of sucrose synthesis (1).

To study the interactions between nitrogen and carbon metabolism in maize, we have used Chl *a* fluorescence analysis to moniter changes in the regulation of the photosynthetic apparatus of plants undergoing nitrogen limitation. To avoid any alteration in carbon metabolism of N-deficient plants due to changes in the NO_3^- concentration in the tissues and not to changes in nitrogen utilization, we chose 0.8 mM nitrate-grown plants as controls since they exhibited maximal biomass production and normal nitrogen characteristics in the absence of NO_3^- accumulation in the leaves (16).

MATERIALS AND METHODS

Plant Material

Maize (*Zea mays* L. cv Contessa) seeds were germinated for 3 d and then grown in a growth chamber on aerated 0.2 mM $CaSO_4$ for an additional 3 d. After subsequent removal of the endosperm, treatments were initiated by transplantation of the seedlings into pots of sand (8 plants per pot of 3.6 L) for an additional 2 weeks. The growth environment consisted of (dawn/day/dusk/night) cycle of 19/22/20/17°C, with dawn/day/dusk length of 2.5/7.5/6 h at 140/200/65 μmol m^{-2} s^{-1} PAR respectively (cool-white fluorescent lamps, General Electric, FG6 PG 17/C W) and 85% RH. The nutrient solution (pH 6.0) was automatically delivered at the rate of 100 mL every 30 min in the light period and 100 mL every 2 h during the night. The whole experiment was carried out twice. Data are the mean of four measurements with four different plants per treatment.

Treatments, Sampling, and Assays

Maize seedlings were supplied with 0.05, 0.2, 0.8, or 12.0 mM NO_3^- as Ca^{2+} salt plus 1.0 mM KH_2PO_4, 0.2 mM NaCl, 0.1 mM K_2HPO_4 and, either 0.7 mM K_2SO_4; 0.4 mM $MgSO_4$; 1.4 mM $CaCl_2$; in addition to micronutrients: 24 μM H_3BO_3, 16 μM FeEDTA, 9 μM $MnSO_4$, 3.5 μM $ZnSO_4$, 1 μM $CuSO_4$, and 0.1 μM $(NH_4)Mo_7O_{24}$. Plants were used for Chl *a* fluorescence measurements after 14 d treatment and for biochemical assays the day after. In the latter case, plants were harvested 4 h after the beginning of the photoperiod. The third leaf of each plant was rapidly excised, weighed, and frozen in liquid nitrogen for subsequent determination. Chl, nitrate, and total nitrogen were assayed as described by Khamis and Lamaze (16). Ammonium and amino acid extraction was done in 3% sulfosalicylic acid (5 mL for 1 g fresh weight). Amino acids and ammonium were determined using a Biotronik LC 5001 analyzer (physiological program run with lithium buffers, detection at 570 and 440 nm after postcolumn derivatization with ninhydrin) and a SP 4090 Spectra-Physics integrator after baseline subtraction.

Table II. *Free Amino Acid Content in the Third Maize Leaf*

The experimental conditions were as those of Table I.

Treatment	Amino Acid										
	Asp	Asn	Glu	Gln	Gly	Ser	Ala	Pro	Thr	Val	Met
mM	*nmol g^{-1} fresh wt*										
12.0	1055	63	1645	525	959	780	2728	89	179	139	16
0.8	1120	50	1585	470	594	528	3648	67	140	98	13
0.2	610	32	1024	238	342	245	1437	57	79	81	10
0.05	312	0	810	41	177	117	352	24	43	63	8

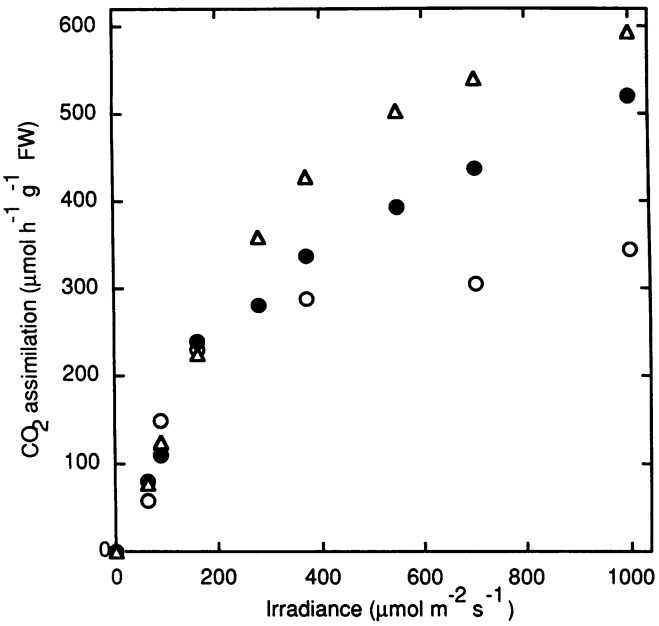

Figure 1. Light saturation curves of CO_2 assimilation in maize plants grown with 0.8 (△), 0.2 (●), and 0.05 mM (○) nitrogen supply.

Chl *a* Fluorescence and CO_2 Assimilation Measurements

Measurements of room temperature Chl *a* fluorescence in air were made on the attached fourth leaf using a pulse amplitude modulation fluorometer (PAM-101, H. Walz, Effeltrich, FRG). CO_2 assimilation measurements were made both simultaneously with fluorescence analysis and also the day after in the growth chamber using infra-red gas analysis (ADC and LCA-2, Analytical Development Co., Hoddesdon, England) and the transpiration rates assessed with a hygrodynamic sensor (Elcowa, 1100, DP). The quantum efficiency of electron transport through PSII was estimated from Chl fluorescence quenching analysis using the light doubling technique (2) as described by Genty *et al.* (10) as follows

$$\phi PSII = (Fm - Fs)/Fm$$

where Fm is the level of Chl a fluorescence when all the PSII traps are closed, *i.e.* during a saturation light pulse, and Fs is the steady-state level of Chl *a* fluorescence.

Photochemical quenching of Chl *a* fluorescence qQ was estimated as follows

$$qQ = (Fm - Fs)/Fv$$

where variable fluorescence, Fv = Fm − Fo. Fo is the level of fluorescence measured when all the PSII traps are open following a dark incubation.

Nonphotochemical quenching of Chl *a* fluorescence (24) was calculated as follows

$$qNP = (Fm' - Fm)/Fm'$$

where Fm′ is the maximum level of Chl *a* fluorescence measured during a saturating light pulse immediately following a dark incubation period of 30 min.

Carotenoid Estimation

Carotenoid pigments were separated by nonaqueous reversed phase high pressure liquid chromatography using a gradient of 3 to 40% dichloromethane in a mixture of acetonitrile and methanol (70/30, v/v) on a Dupont Zorbax octadecyl silica (ODS) column. Continuous measurement of the pigment spectrum was made using a Hewlett-Packard 1040A spectrophotometer. The Hewlett-Packard 1040 A diode array spectrophotometer allowed accurate pigment absorption spectra to be obtained without the need to stop solvent flow. Quantitative determination of the pigments was obtained after external calibration of the DPU multichannel integrator and detector with available standards. The extinction coefficients used were as described previously.

RESULTS

Plants grown at 0.8 mM NO_3^- had similar growth characteristics, reduced nitrogen and Chl contents as plants grown with higher nitrate concentrations in the nutrient solution (Table I). The free amino acid contents were slightly lowered with the exception of alanine and aspartate which were increased, as a result of the restriction in NO_3^- availability in the plants grown at 0.8 mM NO_3^- when the NO_3^- content of the shoot was almost zero (Table II). When the nitrate available to the plants was lowered to 0.2 and 0.05 mM, a large decrease in shoot growth was observed (Table I) which was accompanied by lower Chl and reduced N contents and by a greatly altered amino acid composition (Table II). Due to the large decrease in Chl content accompanying N limitation the rate of photosynthesis expressed on a Chl basis was double that of the control in the plants grown with 0.05 mM NO_3^- (Table I). However, the light-saturated rates of photosynthesis

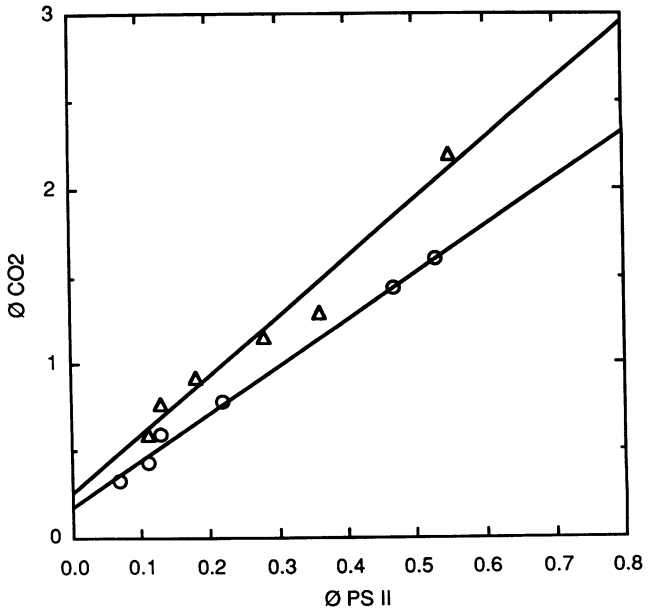

Figure 2. Relationship between the quantum efficiency of CO_2 assimilation (ϕCO_2) and the quantum efficiency of PSII (ϕPSII) in maize leaves grown with 0.8 (△) and 0.05 (○) mM nitrogen.

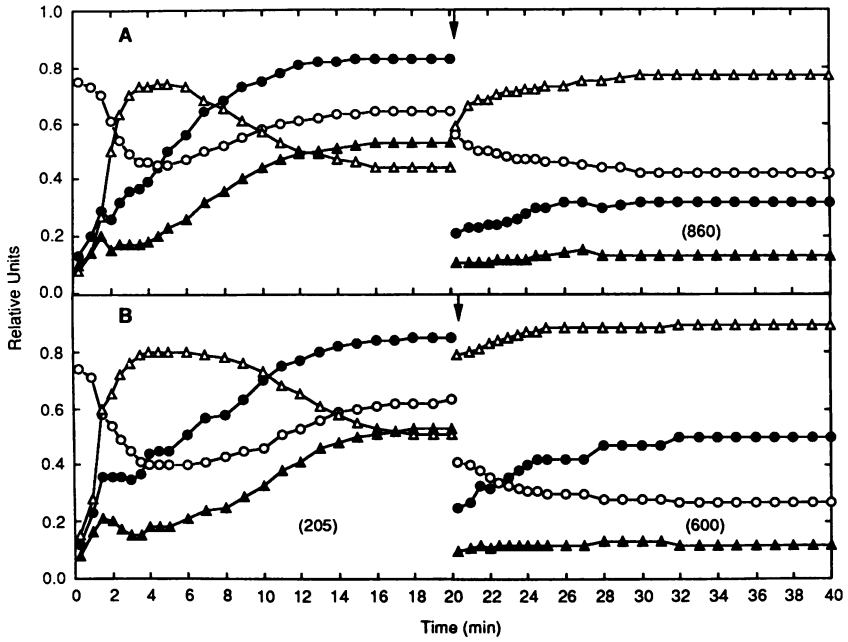

Figure 3. Effect of nitrogen supply on the qQ (●), qNP (△), Fv/Fm (○), and ϕPSII (▲) during the induction phase of photosynthesis at low irradiance (160 μmol m^{-2} s^{-1}) with a subsequent transition to high (1000 μmol m^{-2} s^{-1}) irradiance. The experiment was carried out on plants grown on either 0.8 (A) or 0.05 (B) mM nitrogen. Numbers in brackets in the figure represent values of the rates of CO_2 assimilation (μmol h^{-1} g^{-1} fresh weight). Arrows indicate the point of transition in irradiance.

on a leaf area or mass basis were decreased significantly by nitrogen limitation (Fig. 1). The quantum yield of CO_2 assimilation was only slightly diminished in the 0.05 mM NO_3^--grown plants compared to those grown at 0.8 mM NO_3^-. The relationship between the quantum efficiency of CO_2 assimilation and the quantum efficiency of PSII electron transport (ϕPSII = Fv/Fm·qQ) was linear in all cases (Fig. 2).

The relative contributions of photochemical (qQ) and non-photochemical (qNP) quenching in determining ϕPSII during the induction of photosynthesis are given in Figure 3. At low irradiances, the steady-state values of qQ and qNP attained at the end of the induction period were similar in nitrogen sufficient and deficient plants and, hence, values of ϕPSII were comparable even though the rate of photosynthesis was lower in the nitrogen-deficient plants (Fig. 3). At high irradiance, ϕPSII values were much lower than those measured at low irradiances (Fig. 3). It is clear that the relative contribution of qQ and qNP to the ultimate value of ϕPSII were distinctly altered as a result of severe nitrogen limitation. At high irradiance, qNP remained high in steady-state compared with that obtained at low irradiance but qNP values were higher in the 0.05 mM NO_3^--grown plants than in plants grown at 0.8 mM NO_3^-. The values of qQ were larger in leaves suffering nitrogen deprivation compared to those of leaves grown with

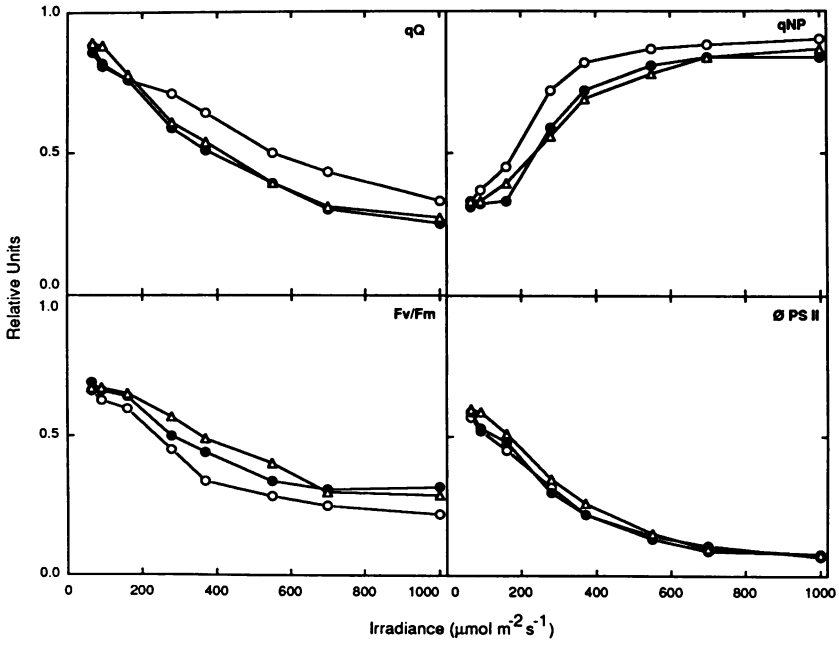

Figure 4. Light response curves of qQ, qNP, Fv/Fm, and ϕPSII in maize leaves grown at either 0.8 (△), 0.2 (●), or 0.05 (○) mM nitrogen.

adequate nitrogen, indicating that the primary electron acceptor of PSII, QA, is more oxidized in the former case than in the latter. Thus, a small change in qNP can facilitate a large change in qQ.

The change in the relative contributions of qQ and qNP to ϕPSII is clearly seen in the light response curves of those parameters (Fig. 4). During steady-state photosynthesis, the values of qQ (Fig. 4A) decreased with increasing irradiance in all types of plants. This is the normal observation of the response of qQ to increasing irradiances. The values of qQ obtained for plants grown at 0.2 mM NO_3^- were similar to those of plants grown at 0.8 mM NO_3^-. With plants grown at 0.05 mM NO_3^-, qQ initially decreased in the same way over the lowest light intensities to 160 μE m^{-2} s^{-1} (slightly below the growth irradiance). However, above this irradiance qQ remained higher in plants grown at 0.05 mM NO_3^- than in plants grown either at 0.2 or 0.8 mM NO_3^-. This suggests that the acceptor of PSII remained more oxidized at high irradiance in nitrogen-deficient plants despite the much lower rates of photosynthesis of these plants.

In the plants grown at 0.05 mM NO_3^-, qNP rose more quickly with respect to irradiance and attained higher maximum values than those observed in plants grown at 0.2 and 0.8 mM NO_3^- (Fig. 4B). The relative decrease in the Fv/Fm ratio, *i.e.* in the quantum efficiency of open centers, with increasing irradiance, was much greater in plants grown at 0.05 mM NO_3^- in comparison to those grown at higher nitrogen availability. This suggests that the regulation of PSII photochemistry was modified substantially as a result of severe nitrogen deficiency. ϕPSII was only slightly lower in the plants grown in limiting nitrogen (0.05 and 0.2 mM) than in those grown with adequate nitrogen (0.8 mM). Differences in the response of quenching components could be seen between the plants grown at 0.2 and 0.8 mM NO_3^- upon a transition from low to high irradiance (Fig. 5). The light transition caused a large, rapid, but transient fall in qQ in the plants

Table III. *Steady-State Xanthophyll Contents of Illuminated Maize Leaves*

Plants were grown under 200 μmol m^{-2} s^{-1}, PAR with 0.8 mM, 0.2 mM, and 0.05 mM NO_3^- and measurements were made 4 h into the photoperiod after 10 min under 1000 μmol m^{-2} s^{-1}.

Treatments	Zeaxanthin[a] + Antheraxanthin	Violaxanthin[a]	Chl[b] / Xanthophyll	Chl[b] / Carotenoid
mM				
0.05	21.6	6.1	4.8	3.7
0.20	15.8	8.0	4.9	4.3
0.80	8.1 (9.2)	16.1 (15.2)	5.6	4.7

[a] Values expressed as a percentage of the total leaf xanthophyll content. [b] Mol/mol. Numbers in parentheses represent values for plants grown with 12 mM NO_3^-.

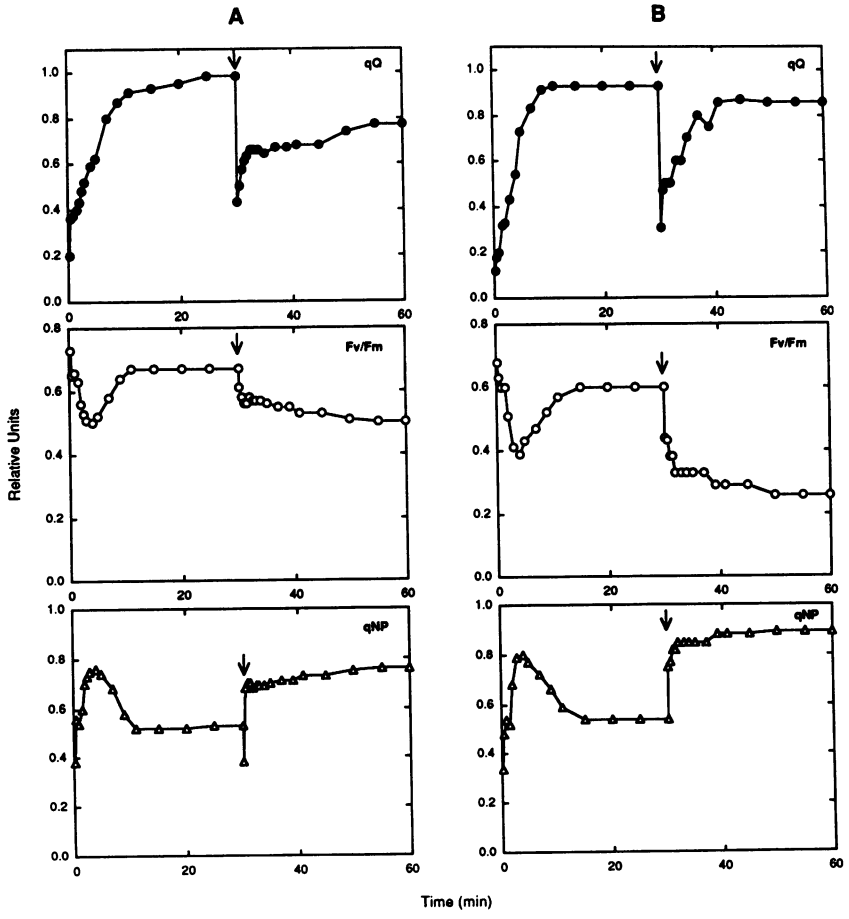

Figure 5. Effect of the transition from low (260 μmol m^{-2} s^{-1}) to high (1400 μmol m^{-2} s^{-1}) irradiances on qQ, Fv/Fm, and qNP values in maize plants grown with either 0.8 (A) or 0.2 (B) mM nitrogen. Arrows indicate the point of transition in irradiance.

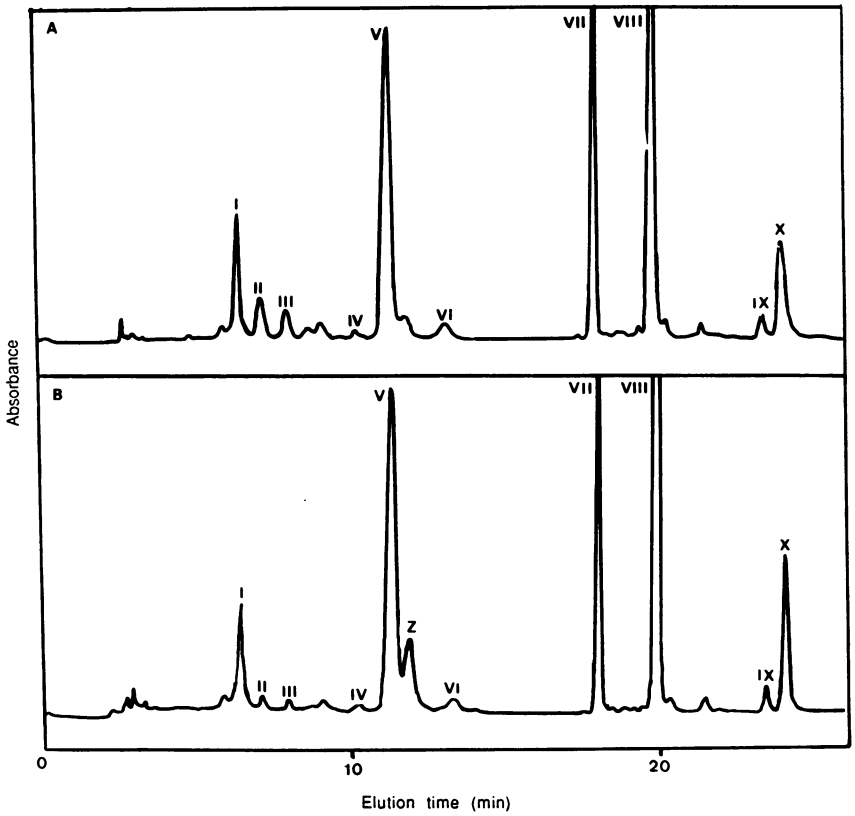

Figure 6. Effect of nitrogen limitation on the elution pattern of pigments from maize leaves measured at 437 nm following high pressure liquid chromatography. Leaves were taken 4 h after the beginning of the photoperiod (200 μmol m^{-2} s^{-1}) from plants grown with 0.8 mM (A) or 0.05 mM (B) NO_3. The peaks are identified as (I) neoxanthin, (II) violaxanthin, (III) luteoxanthin, (IV) antheraxanthin, (V) lutein, (Z) zeaxanthin, (VI) *cis*-lutein, (VII) Chl *b*, (VIII) Chl *a*, (IX) pheophytin a, and (X) β-carotene.

grown with 0.2 mM NO_3^- compared to those grown with 0.8 mM NO_3^-. Subsequently, qQ recovered to a value slightly higher in the 0.2 mM NO_3^- grown plants than those with 0.8 mM NO_3^-. Similarly, the decrease in Fv/Fm ratio following the light transition was much greater for the 0.2 than the 0.8 mM NO_3^--plants and values of qNP were slightly higher in the former than the latter. Thus, QA apparently attained a relatively oxidized state in the nitrogen deficient plants as a result of a decrease in Fv/Fm ratio and an increase in qNP. Similarly, it is clear that when photosynthesis is quickly shifted from a light-limiting to a light-saturating situation the immediate effect is a decrease in qQ but this can be rapidly reversed as a result of an increase in qNP and a decrease in the Fv/Fm ratio.

The xanthophyll composition of the leaves varied considerably with nitrogen availability. Figure 6 shows the changes in the relative levels of pigments in plants grown at 0.8 and 0.05 mM nitrate and demonstrates the pigment profiles obtained in these experiments. In all circumstances the leaf contents of violaxanthin plus antheraxanthin plus zeaxanthin amounted to approximately 24 to 27% of the total xanthophyll pool. Zeaxanthin was present in the leaves from plants of all types (Fig. 6; Table III). However, at the same irradiance, the leaves from plants grown with limiting nitrogen contained considerably more zeaxanthin and less violaxanthin than leaves of control (Table III). Similarly, the ratios of Chl/xanthophyll and Chl/carotenoid were decreased. The xanthophyll contents of plants grown at 12.0 mM NO_3^- were essentially similar to those of plants grown at 0.8 mM NO_3^-. There was no evidence to suggest that zeaxanthin was formed at the expense of β-carotene, or any carotenoid, other than violaxanthin (Fig. 6).

DISCUSSION

Maximal biomass production and normal nitrogen status can occur in maize in the absence of NO_3^- accumulation in shoots (16) and in the present work plants were grown with 0.8 mM NO_3^-. Our results allow a comparison of the photosynthetic characteristics of nitrogen-limited plants with those of control plants which have also very low NO_3^- levels in the tissues. It is thus possible to interpret the effects of NO_3^- deficiency in terms of N-limitation and to exclude the possibility of internal NO_3^- perturbation, *i.e.* NO_3^- considered in this latter case as an anion and as a major component of the cell osmolarity. This is important since the leaf NO_3^- level *per se* has been assumed to affect photosynthesis and the solute balance of the cell which, in turn, affect sugar accumulation (1).

The decrease in photosynthetic capacity reported here is consistent with the results of Sugiharto *et al.* (25) who showed that nitrogen-deficiency resulted in a selective decrease in the photosynthetic enzymes of the C4 cycle, as well as ribulose 1,5-bisphosphate carboxylase, in maize. Nitrogen deprivation, however, did not strongly alter CO_2 photoassimilation at low light intensities and thus the quantum yield of CO_2 assimilation was only slightly altered by N-deficiency. In contrast, nitrogen limitation results in a reduction in the rate of light-saturated photosynthesis and, at high irradiance, causes modifications in the photosynthetic apparatus and photochemis-

try. Thus, limited nitrogen nutrition in maize led to a reduction in photosynthetic capacity without a reduction in photosynthetic efficiency when plants were grown at low irradiance equivalent to deep shade for this species. As expected, a linear relationship between ϕCO_2 and ϕPSII was found although the slope of the relationship changed with nitrogen availability. This change may be related to differences in the PSII antenna resulting from nitrogen limitation, as described previously (10). The nitrogen-deprived plants have a much lower Chl content than the plants grown with an adequate nitrogen supply, resulting in increased rates of photosynthesis when Chl is used as the basis for expression. Thus the nitrogen-limited plants are much more efficient in terms of CO_2 assimilated per photosynthetic unit. Nitrogen deficiency resulted in a large increase in the zeaxanthin content of the leaves. Since the formation of zeaxanthin via the xanthophyll cycle has been consistently shown to correlate with the appearance of the nonphotochemical quenching process, qI (4, 5) and to influence the conditions necessary for qE formation (21), we are drawn to the conclusion that this change in the xanthophyll composition of the antenna makes a major contribution to the enhanced qNP observed in nitrogen-deficient maize leaves. More importantly, the increased capacity for excitation energy dissipation in PSII displayed by these shade-grown and nitrogen-regulated plants affords protection after transfer to potentially photoinhibitory conditions. Plants acclimate or adapt to the available light and nutrient environment and as a consequence can display a range of sensitivities to damage by high irradiances. It has been clearly demonstrated that shade plants grown with low nitrogen are more susceptible to photoinhibition than those that are nitrogen replete (8, 12). However, it is important to note that it is difficult to distinguish between light-induced damage to the PSII reaction center and the regulated decreases in PSII efficiency via the processes that facilitate thermal dissipation of excitation energy (4, 5, 21), using measurements of Fv/Fm and quantum yield alone. Recent advances in our understanding of the mechanisms by which the utilisation of excitation energy is controlled suggest that dissipation is a major cause of decrease in PSII quantum efficiency *in vivo* (9, 21).

It appears from the data presented here that qNP rises more quickly with increasing irradiance; *i.e.* thermal dissipation is higher for a given irradiance, and ultimately attains higher values in nitrogen-deficient plants compared to those grown with adequate nitrogen. This again may be linked to the enhanced zeaxanthin content since a possible dependence of energy-dependent quenching on xanthophyll composition has been demonstrated in thylakoids (21). In addition, the Fv/Fm ratio decreases more rapidly and drops to a lower level as a result of nitrogen-limitation. The low Fv/Fm ratio decreases ϕPSII (the product of Fv/Fm by qQ) to values that are comparable to those in nitrogen-replete plants despite the relatively higher qQ value. We conclude that it is the enhanced level of qNP that allows PSII to remain more oxidized at high irradiances in nitrogen-limited plants, even though the rate of carbon assimilation is greatly reduced compared to controls. Since values of qQ, qNP, and Fv/Fm are similar at low irradiance in the presence or the absence of nitrogen deficiency, it appears that the type of qNP which facilitates oxidation of qQ is initiated only at higher light intensities. These features can be readily explained in terms of regulation of PSII function by a reduction in the rate of excitation transfer to PSII because of the effective competition for excitation energy in the antenna between thermal dissipation processes and the reaction center. However, direct quenching of PSII photochemistry in the reactions centers, *e.g.* via a PSII cycle is also possible (7). The sustained oxidation of PSII at high irradiance must be seen as a protective mechanism since overreduction of PSII is a photoinhibitory condition (9). As a result of nitrogen limitation, the Chl content per unit leaf mass is reduced and the zeaxanthin content is increased suggesting that this relative pigment reorganization is a strategy for protection of PSII function. Thermal dissipation processes may overcompensate for the lowered CO_2 assimilation capacity at high irradiances leaving PSII much more oxidized than the normal condition. Loss of quantum efficiency in PSII is clearly traded for sustained oxidation on the acceptor side of PSII. This simple mechanism affords adaptation to the stress imposed by nitrogen-limitation when such plants are exposed to irradiances above the light-saturation point. Since the light saturation point is much reduced as a result of nitrogen deprivation, this is clearly an important protective strategy. Nitrogen limitation in maize reduces the activities of key photosynthetic enzymes (25) and clearly the photosynthetic light response is a reflection of this limitation. Surprisingly, the nitrogen-limited plants do not appear to suffer photoinhibitory damage following short-term exposure to light levels above saturation for photosynthesis and this appears to be related to zeaxanthin formation. Clearly, enhanced dissipation of excitation protects the PSII reaction center. However, we do not know if the nitrogen-limited chloroplast can conserve its membrane protein turnover under long-term irradiance stress.

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