

## Influence of N,P,K nutrition on sunflower plants.

### 1. Chlorophyll fluorescence in relation to photosynthesis

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#### SUMMARY

The effects of long term application of N, P and K nutrients on sunflower (*Helianthus annuus* L.) hybrids NS-H-43 and NS-H-52 in stationary field experiment have been studied. Photosynthetic rate and chlorophyll *a* fluorescence parameters were simultaneously measured in sunflower leaves as plant growth progressed from the stage of flower bud to maturity. The results show that rate of photosynthesis depends on light intensity, leaf age, genotype and the treatment. The highest rates were detected in fully developed 25th leaves of NS-H-52 plants during the anthesis in most treatments. The efficiency of photosystem II photochemistry and quantum yield of photosynthesis were the highest in the leaves of NPK supplied plants and the lowest in the leaves of K-treated plants of both genotypes, suggesting that PSII photochemistry can be modified by mineral nutrition of plants. Photosynthetic processes were more affected by nitrogen or NPK nutrition in NS-H-43 plants than in NS-H-52 plants.

#### INTRODUCTION

The important role of nitrogen and phosphorus nutrition for growth and production of sunflower has been demonstrated and reviewed in several publications (1, 2, 3) Nitrogen supply influences the size, structure and activity of photosynthetic apparatus (4), regulates content and activity of Rubisco (5) and level of plant hormones (6). One of the most important effects of phosphate deficiency is in the reduction of photosynthetic rate of leaves (7). Orthophosphate deficiency may limit photosynthesis by decreasing ATP synthesis, by affecting translocation of triosephosphates from chloroplasts (8), by decreasing carboxylation efficiency and apparent quantum yield for CO<sub>2</sub> assimilation (9). Peoples and Koch (10) have reported a clear effect of potassium on the rate of CO<sub>2</sub> assimilation, they showed that K<sup>+</sup> promoted *de novo* synthesis of Rubisco and decreased the diffusive resistance for CO<sub>2</sub> in the mesophyll.

In the present investigation we grew two sunflower hybrids NS-H-43 and NS-H-52 in the stationary field experiment, under optimal levels of nitrogen, phosphorus and potassium supply. We compared the effects of long term application of individual nutrients and their combination on the photosynthetic rate and the efficiency of primary photochemical reactions and photosynthesis in the leaves of the examined hybrids in several stages of plant development.

## MATERIALS AND METHODS

Sunflower hybrids (*Helianthus annuus* L.) NS-H-43 and NS-H-52 were grown in stationary field experiment which was started in 1966 at Rimski Šančevi, Novi Sad. Soil (chernozem, 100g contains: 0.170-0.181g total N, 15-23mg P<sub>2</sub>O<sub>5</sub> and 23.7-29.0 mg K<sub>2</sub>O) was either not supplied (control), or supplied with 100 kg/ha N (N), 100 kg/ha P<sub>2</sub>O<sub>5</sub> (P), 100 kg/ha K<sub>2</sub>O (K), or with their combination (NPK). There were five replicates of each treatment.

Simultaneous measurements of photosynthetic oxygen evolution (polarographic) and chlorophyll fluorescence yield were done on the 25th leaf, on a leaf disc (10 cm<sup>2</sup>) in a closed chamber (LD2, Hansatech) at 25°C and 5% CO<sub>2</sub> (11). The leaf disc was illuminated by increasing photon flux densities (PFDs) until steady-state rate of photosynthesis was achieved at each irradiance. At steady-state photosynthesis pulses of saturating light (5000 μmol quanta m<sup>-2</sup>s<sup>-1</sup>, 990 ms) were given every 20 s to fully reduce the primary electron acceptor of photosystem II (PSII) and remove photochemical quenching. Rates of photosynthesis, photochemical (qp) and nonphotochemical (qN) components of fluorescence quenching, efficiency of excitation energy capture by open PSII reaction centres  $\Phi_{exc}$  and quantum efficiency of PSII electron transport  $\Phi_{II}$  (12) were determined as described by Plesničar and Panković (13) at five PFDs. The quantum yield of photosynthesis  $\Phi_s$  was calculated by dividing the gross rate of O<sub>2</sub> evolution by the incident photon flux density (0-967 μmol quanta m<sup>-2</sup>s<sup>-1</sup>).

## RESULTS AND DISCUSSION

A long term study conducted in a stationary trial at Rimski Šančevi showed that over the period of 20 years fertilization affected significantly the level of mineral nitrogen and available phosphorus in the soil, while total nitrogen and available potassium were not affected significantly (14). The addition of nutrients to the soil had a similar stimulatory effect on the growth of plants of both examined NS-hybrids (15).

Our results show that rate of photosynthesis depends on leaf age, genotype and the treatment (Fig. 1). The highest rates were detected in the leaves 20 - 25 (counting from the base) during the anthesis (16). Rate of photosynthesis decreased with leaf age in all treatments, more so in NS-H-52 than in NS-H-43 plants. Leaves of NS-H-43 plants grown on the soil that was supplied with NPK (300 kg/ha), or with 100 kg N/ha, exhibited higher rates of photosynthesis compared to the control plants, while the NS-H-52 plants, which generally exhibited higher rates of photosynthesis, did not react to these treatments. Supply of potassium alone (100 kg K<sub>2</sub>O/ha) was inhibitory for photosynthetic processes in examined leaves of both hybrids.

Chlorophyll fluorescence measurements were employed to monitor changes in the redox state of the photosynthetic electron transport chain and in thylakoid energization in the examined leaves. The yield of chlorophyll *a* fluorescence is determined by two distinct quenching processes, qp (photochemical quenching), which is caused by the use of energy for

the photosynthetic electron transport and  $q_N$  (non-photochemical quenching) which arises from nonradiative dissipation of excitation energy (17).

During steady-state photosynthesis  $q_p$  decreased with increasing irradiance in all treatments (Fig. 2). With both genotypes plants that were supplied with NPK nutrients had the lowest values of  $q_p$ , control plants (NS-H-52) and K-treated plants (NS-H-43) had the highest  $q_p$  values. This suggests that the primary electron acceptor of PSII remained more reduced in the leaves of NPK supplied plants despite the higher rates of photosynthesis, indicating higher photosynthetic capacity of these plants; when saturating irradiance was applied, electron acceptor of PSII became more oxidized than in most other treatments. Non-radiative dissipation of excitation energy ( $q_N$ ) was the lowest in leaves of NPK supplied plants and it was the highest in the leaves of N- and K-supplied plants of both genotypes. As a consequence, the efficiency of excitation energy capture by open PSII reaction centres, as well as the efficiency of PSII electron transport were the highest in NPK supplied plants and the lowest in N- and K-treated plants. Correspondingly, photosynthetic light response curves demonstrate the highest rates of photosynthetic  $O_2$  evolution and the highest quantum efficiency of photosynthesis in the leaves of NPK-treated plants (Fig.2). High level of  $q_N$  allows PSII to remain more oxidized in K-treated plants even though the rate of photosynthesis is reduced compared to the control. The leaves of K-treated plants exhibited the lowest quantum efficiency of photosynthesis. High photosynthetic rates in the leaves of N-treated plants, which exhibited low efficiency of PSII electron transport, are most probably caused by higher activities of photosynthetic enzymes (5) and, therefore, increased efficiency of RuBP regeneration. The results suggest that PSII photochemistry can be modified by mineral supply (18).

The relationship between  $\Phi_{II}$  and  $\Phi_s$  (Fig. 3) changed under the influence of plant nutrition but in all examined cases it showed lower quantum efficiency of PSII electron transport, which corresponds to higher quantum efficiency of photosynthesis, in the leaves of NS-H-52 than in leaves of NS-H-43. This behaviour is characteristic for light-limited range of light response curve and it does not apply to the examined leaves of N-treated plants, which show similar relationship for both genotypes. The observed differences in photosynthetic rates and photosynthetic efficiencies between the two hybrids are genetically determined and most probably caused by the differences in morphology and in leaf area duration. Plants of NS-H-43 hybrid have smaller number of larger leaves, longer vegetation and greater LAD than NS-H-52 plants. Our results indicate that 25th leaf of NS-H-52, according to its photosynthetic characteristics, corresponds more to the 20th leaf than to 25th leaf of NS-H-43 (16, 13).

#### CONCLUSIONS

A study of the effects of long term application of N, P, K nutrients on sunflower plants in stationary field experiments indicates that:

- rate of photosynthesis depends on the leaf age, genotype and the treatment,
- rates of photosynthesis and quantum yield of photosynthetic  $O_2$  evolution in 25th leaf of NS-H-52 plants were higher than those in the leaf at the same position on NS-H-43 plants for most treatments,

- under the influence of NPK or N nutrients photosynthetic  $O_2$  evolution rates were increased in the examined leaves of NS-H-43 plants, while they did not differ significantly between the nutrient-supplied and control NS-H-52 plants,
- the efficiency of PSII photochemistry and quantum yield of photosynthesis were the highest in the leaves of NPK supplied plants and the lowest in the leaves of K-treated plants of both genotypes, suggesting that PSII photochemistry can be modified by mineral nutrition of plants,
- the observed differences in photosynthetic rates and photosynthetic efficiencies between the NS-H-43 and NS-H-52 plants are genetically determined and most probably caused by the differences in plant morphology and in leaf area duration.

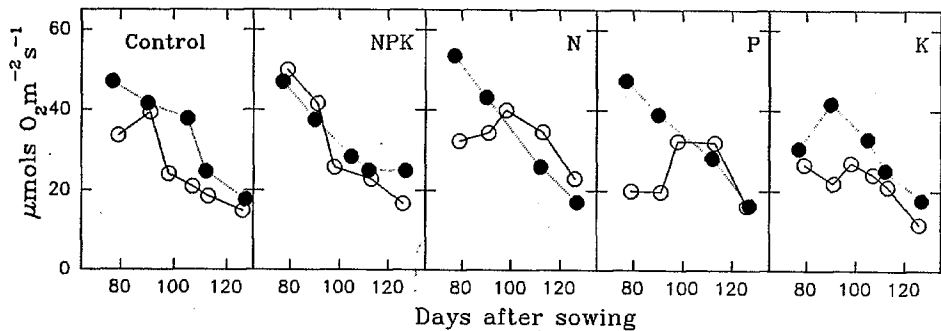


Figure 1. The effect of N, P, K nutrition on the rate of photosynthesis in sunflower leaves. Photosynthetic  $O_2$  evolution was measured at saturating irradiance, on the 25th leaf of NS-H-43 (○), or NS-H-52 plant (●) in several stages of plant development.

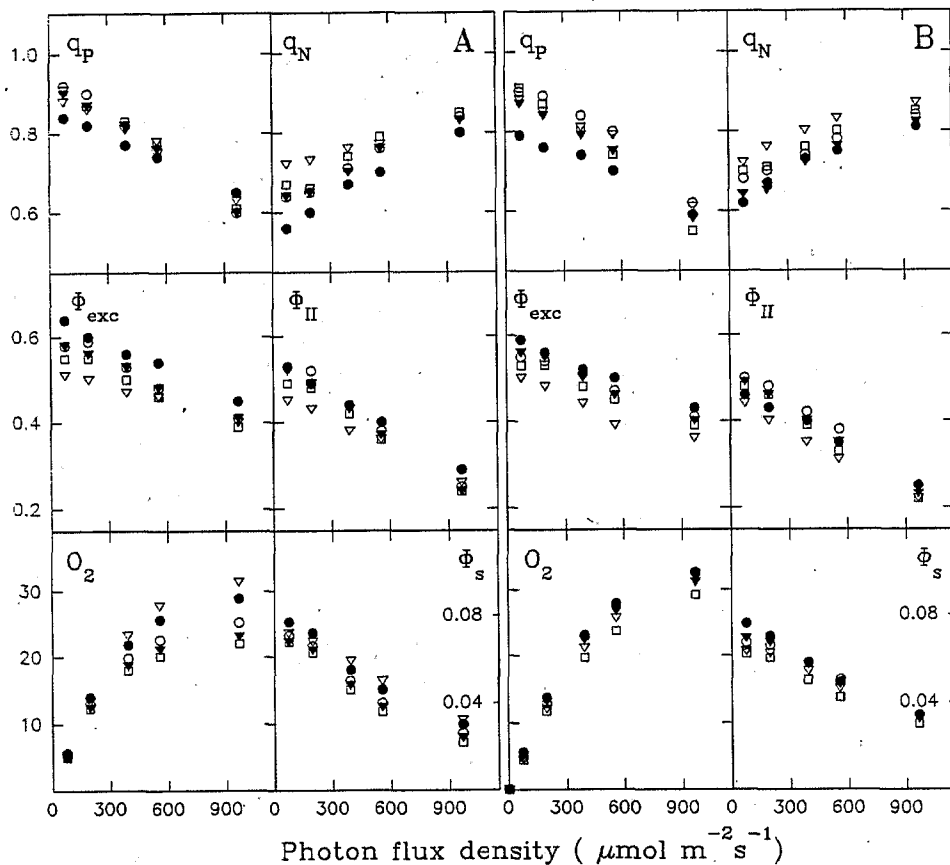


Figure 2. Light response curve of photochemical ( $q_p$ ) and non-photochemical fluorescence quenching ( $q_N$ ), photochemical efficiency of open PSII reaction centres ( $\phi_{exc}$ ), quantum efficiency of PSII electron transport ( $\phi_{II}$ ), rate of photosynthetic  $O_2$  evolution ( $O_2$ ) and quantum yield of  $O_2$  evolution ( $\phi_s$ ) on the 25th leaf of NS-H-43 (A) and NS-H-52 plant (B) under the influence of N, P, K nutrition. ○, Control; ●, NPK; ▽, N; ▼, P; □, K. The results are mean values of five determinations.

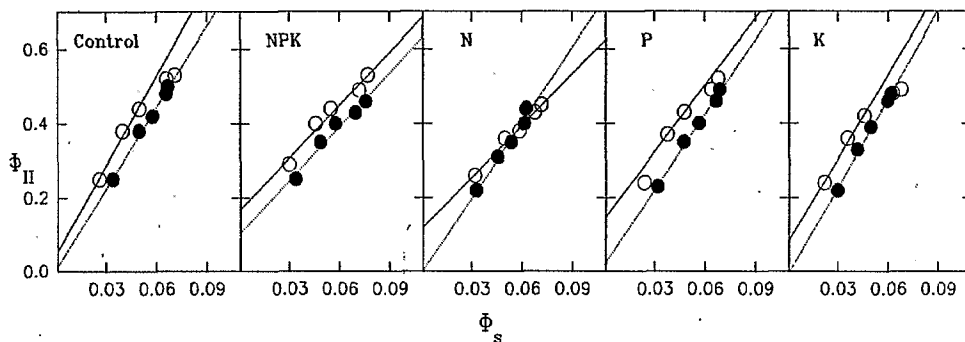


Figure 3. Relationship between the quantum efficiency of PSII electron transport ( $\Phi_{II}$ ) and the quantum efficiency of photosynthetic  $O_2$  evolution ( $\Phi_s$ ) in the leaves of NS-H-43 (○) and NS-H-52 plants (●) under the influence of N, P, K nutrition.

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