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Effect of the change of nitrogen source on photosynthesis and respiration of Scots pine (Pinus silvestris L.) seedlings\*

#### J. ZAJACZKOWSKA

Institute of Sylviculture, Warsaw Agricultural University (Received: October 4, 1973.)

### Abstract

Rates of photosynthesis and respiration of Scots pine seedlings, grown in water culture under conditions of nitrogen deficiency and supplied with nitrogen in ammonium or nitrate form, were investigated. Besides, the rates of photosynthesis and respiration of the seedlings were measured, after changing the form of supplied nitrogen. A rapid and different response of nitrogen deficient seedlings was found independent on the form of supplied nitrogen. The investigations also showed a change in rates of photosynthesis and respiration of seedlings after changing the form of nitrogen.

#### INTRODUCTION

It was found in previous works that Scots pine seedlings grown in water culture with either ammonium or nitrate form of nitrogen accumulated different amounts of organic substances as well as exhibited their various distribution in particular organs (Łotocki and Żelawski, 1973; Zajączkowska, 1973). Slight differences in rates of photosynthesis and almost the same respiration rates were observed in the above-mentioned investigations. However, as the plants were investigated in later stages of vegetation, the results have not shown what is the immediate effect of both nitrogen forms introduced into the nutrient solution. One of the ways leading to the explanation of this problem is the investigation of photosynthesis and respiration response to the ammonium or the nitrate form of nitrogen in plants showing symptoms of nitrogen deficiency. Another way of

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approach is the investigation of the response of plants, grown in good conditions of nitrogen supply, to the change of the provided form of nitrogen.

# MATERIAL AND METHODS

Scots pine seedlings were grown in water culture under greenhouse conditions during winter as in the previous work (Zajączkowska, 1973). They grew in the nutrient solution after Ingestad (1962/63). Photosynthesis and respiration measurements were carried out under laboratory conditions with the use of an infrared  $CO_2$  analyzer like in the previous work. Five plants were examined, in a chamber, at one time. Chlorophyll a and b content was determined spectrophotometrically in  $80^9/_0$  acetone extract of fresh needles (Mac Kinney, 1941; Bruinsma, 1963).

In the first experiment two and a half months old seedlings were transferred from complete Ingestad nutrient solution to that without nitrogen also recommended by the same author. First measurements were carried out two weeks after changing the solution. Rates of photosynthesis and respiration of seedlings, growing in solution lacking nitrogen were measured at the beginning of the experiment. Then the plants were transferred to the solution containing either ammonium or nitrate form of nitrogen and rates of photosynthesis and respiration were measured again. Measurements were carried out 10 minutes, 3 hours, 24, 48 and 72 hours after changing the nutrient solution. Root respiration was measured at the end of the three days experiment, immediately after cutting the shoots off. Measurements for each experimental variant were repeated four times.

In the second experiment three months old pine seedlings were transferred for a three month period from the complete nutrient solution to that lacking nitrogen. Plants were placed again in the nutrient solution containing either ammonium or nitrate form of nitrogen after the three months period of time. Gas exchange and chlorophyll a and b content were examined 1, 2 and 3 weeks after the plants were transferred to various solutions. Measurements of both variants at each time of examination, were made in two repeats.

The third experiment was carried out on three months old seedlings grown from the beginning in nutrient solutions containing nitrogen in either ammonium or nitrate form. Photosynthesis and respiration rates were measured 10 minutes and 5 hours after transfer of the plants from the solution containing the ammonium form of nitrogen to that containing the nitrate form, or vice versa. There were four repeats of each experimental variant.

## RESULTS

Slight decrease of the rate of photosynthesis in plants, showing symptoms of nitrogen deficiency, supplied with nitrogen in nitrate form is observed 10 minutes after changing the solution (Table 1). Further decrease of photosynthesis rate in these plants is observed after 3 hours of the experiment. The photosynthetic rate continued to decrease during three days and after that time it amounted to 69% of the rate observed before changing the nutrient solution. A slight increase of photosynthetic rate in plants, transferred from solution lacking nitrogen to that containing the ammonium form of nitrogen, is already observed 10 minutes after the transfer. Differences in photosynthetic rate, before changing the nutrient solution and 10 minutes after the change, are slight in both variants. Nevertheless the reproducibility of four repeats within each variant indicates the differences to be true. The rate of photosynthesis in plants of two repeats remained during three hours on the same level, as it was after 10 minutes of the experiment, while a slight decrease of the photosynthetic rate occurred in plants of the other two repeats. Further decline in the rate of photosynthesis was observed after that period of time. The rate of photosynthesis of plants 72 hours after supplying the ammonium form of nitrogen, amounted to  $84^{\circ}/_{\circ}$  of the value found before the changing of solution, so the decline was slower than in plants supplied with the nitrate form of nitrogen. The decline in the rate of photosynthesis could partially result from the state of plants getting worse during the three days period of staying in the experimental chamber.

Respiration rates of shoots of both experimental variants were similar during three days of experiment. Root respiration rates measured on the third day after changing the nutrient solution was considerably higher in seedlings with nitrate form of nitrogen than in those supplied with the ammonium form of nitrogen.

The ratio of photosynthesis to respiration estimated for the whole plant from the values obtained on the third day after changing the nutrient solution was distinctly higher in plants supplied with the ammonium form than in those transferred to solution containing the nitrate form of nitrogen.

Higher rates of photosynthesis were observed in plants transferred from nitrogen lacking solution to that containing the ammonium form than in the control plants (Table 2), while plants transferred from solution lacking nitrogen to that with the nitrate form exhibited lower rates of photosynthesis than the control plants. The results of this experiment indicate that the previously observed increase of photosynthetic rate, already after 10 minutes from the moment of providing the plants with the ammonium form of nitrogen, as well as the

Table 1

Rates of photosynthesis and respiration before and after the change of solution

	Pho	Photosynthesis mg CO2.h-1.g of dry weight-1	mg CO2.	h⁻¹•g of c	ry weight	.1	Respira	tion mg (	02.h-1.g	Respiration mg CO2.h-1.g of dry weight-1	ight-1	Dotion
7/424			after the	after the change of solution	solution				shoot		root	nhotosyn-
Experimental	before the					1000	before the	afi	ter the cha	after the change of solution	ution	thesis/res
variant change of solution	change of solution	10 minutes	3 hours	24 hours	48 hours	72 hours	change of solution	24 hours	48 hours	72 hours	at the end of the experiment	piration whole plant
NO <sub>3</sub> -	16.2	15.8	14.6	13.3	11.9	11.2	1.2	1.2	1.2	1.0	3.1	3.5
NH <sup>4</sup> +	16.3	17.0	16.9	15.9	14.8	13.8	1.0	1.1	1.2	1.1	2.0	5.5

Table 2

Rates of photosynthesis and respiration 1, 2, and 3 weeks after the change of solution

	After	one week sol	After one week from the change of solution	ange of	After t	After two weeks from the change of solution	ks from the cl solution	nange of	After tl	After three weeks from the change of solution	eks from the c	change of
Experimental variant	mg C	mg CO <sub>2</sub> ·h <sup>-1</sup> ·g of dry weight <sup>-1</sup>	of dry	Ratio: photo-	mg C	mg CO <sub>2</sub> ·h <sup>-1</sup> ·g of dry weight <sup>-1</sup>	of dry	Ratio: photo-	mg C	mg CO <sub>2</sub> ·h <sup>-1</sup> ·g of dry weight <sup>-1</sup>	of dry	Ratio: photo-
- a q r	photosyn- thesis	shoot res- piration	photosyn- shoot res-root respi- thesis piration ration	respiration whole plant		photosyn- shoot res-root respi- thesis piration ration	root respi- ration	syntn./ respiration whole plant		photosyn- shoot res- root respi- thesis piration ration	root respi- ration	synth./ respiration whole plant
NO3-	8.7	1.4	1.6	2.6	9.2	1.9	1.9	2.1	7.1	1.4	1.9	2.0
NH <sub>4</sub> + Control	13.8	1.5	1.7	4.1	10.1	2.0	2.0	2.2	11.1	1.6	1.9	5.6
(without N)	10.0	1.6	1.2	3.0	10.0	1.9	13	2.7	8	1.4	7	26

decrease of this rate, after providing the plants with nitrate form, are not transient and can last for a longer period of time.

Shoot respiration was found for each experimental variant to be similar. Roots of plants transferred to nutrient solution containing either the ammonium or the nitrate form of nitrogen showed similar respiration rates, while control plants showed considerably lower rates of this process. The ratio of photosynthesis to respiration of the whole plant was the lowest in seedlings supplied with nitrogen in nitrate form.

Table 3
Chlorophyll content 1, 2, and 3 weeks after the change of solution

	mg of	chlorophyll.	g of needle fre	esh weight <sup>-1</sup>		
Experimental	one week		two weeks		three week change of	
variant –	a+b	a b	a+b	a b	a+b	$\frac{a}{b}$
NO <sub>3</sub>	2.5	2.5	2.7	2.6	2.3	2.6
NH <sub>4</sub> <sup>+</sup> Control	2.8	2.5	2.7	2.5	2.8	2.5
(without N)	2.4	2.5	2.4	2.6	1.9	2.3

Plants grown in nutrient solutions with either the ammonium or the nitrate form of nitrogen contained more chlorophyll (a  $\pm$  b) compared with control plants (Table 3). Plants supplied with ammonium nitrogen contained more chlorophyll than those supplied with nitrate nitrogen. Chlorophyll a/b ratio was similar in all experimental variants.

Table 4

Rates of photosynthesis and respiration before and after the change of solution

		Photosynthesis		Shoot re	spiration	Root res-
Type of the		mg CO <sub>2</sub> .	h <sup>-1</sup> ⋅g of dry	weight-1		piration
change in nitrogen source	before the change of solution	ten minu- tes after the change	five hours after the change	before the change of solution	five hours after the change of solution	$mg$ $CO_2 \cdot h^{-1}$ • g of dry weight <sup>-1</sup>
from NO <sub>3</sub> to NH <sub>4</sub> <sup>+</sup>	19.4	20.2	20.7	1.5	2.1	3.3
from NH <sup>+</sup> <sub>4</sub> to NO <sup>-</sup> <sub>3</sub>	20.4	20.4	20.1	1.7	2.3	3.5

Plants grown in nutrient solution containing the nitrate form showed 4% increase of photosynthetic rate already 10 minutes after transferring them to the solution with the ammonium form and 6.5% increase 5 hours later (Table 4). Plants transferred from ammonium to nitrate containing solution did not show changes of photosynthetic rate during the 10 minute period and showed slight decrease of the rate of this process after 5 hours. Respiration rate increased during 5 hour period, after changing nutrient solution, and its value was similar in both variants. Roots of seedlings transferred from ammonium to nitrate containing solution showed somewhat higher respiration rates compared with those transferred from nitrate to ammonium containing solution.

#### DISCUSSION

Rapid response of photosynthesis to the supplied form of nitrogen was found in Scots pine seedlings showing symptoms of nitrogen deficiency. Slight increase of photosynthetic rate was already observed after 10 minutes from the moment of providing the nutrient solution containing the ammonium form of nitrogen, while a slight decrease was observed after providing the nitrate form. Thus, the response of photosynthesis to ammonium and nitrate form of nutrition was different.

Data from the literature (see review: Nátr, 1972) indicate that prolonged nutrition with nitrogen in the ammonium form brings about accumulation of ammonia in leaves, which unfavourably affects chloroplast structure and consequently decreases net photosynthesis. The favourable influence of the ammonium form of nitrogen on photosynthesis, observed in the presented experiments, could result from using low concentrations of nitrogen, in the nutrient solutions and relatively short duration of the experiments. Therefore the ammonium was not accumulated in toxic amounts but was incorporated quite efficiently into metabolic processes leading to amino acid and protein synthesis. Also more efficient chlorophyll formation indicates no toxic effect of ammonium ion.

Although the rate of nitrate anion and ammonium cation uptake was not determined, the almost immediate change of photosynthetic rate, after providing nitrogen, indicates that these ions are absorbed very quickly. However, a number of authors, see e.g. Carrodus, (1969) and Yoshida, (1969) found quicker uptake of the ammonium than the nitrate form of nitrogen. Carrodus, (1969) found that the absorbed NH<sub>4</sub> cation was completly incorporated into organic substances already within four hours.

The presented results as well as those of previous works (Łotocki and Żelawski, 1973; Zajączkowska, 1973) did not show differences in the rate of shoot respiration between nutritional variants. However, the slightly higher root respiration rate of seedlings supplied with nitrogen in nitrate form as compared to those supplied with ammonium form as well as the increase of root respiration rate after substituting ammonium form with nitrate, give evidence for stimulating effect of the nitrate form of nitrogen on root respiration. There are works (Warburg, Negelein, 1920; Gilbert, Shive, 1945; Gumiński et al., 1957, see also review Street and Sheat, 1958) in which the respiration of plants supplied with nitrates was found to be more intensive than of those supplied with ammonium salts.

# CONCLUSIONS

- 1. The nitrogen form of nutrition exerts direct influence on the process of photosynthesis.
- 2. The photosynthetic rate of Scots pine seedlings changes quite rapidly after changing the form of nitrogen nutrition.
- 3. The response of photosynthesis to the form of nitrogen nutrition is different; an increase in photosynthetic rate is observed in the case of the ammonium form and a decrease in the case of the nitrate form of nitrogen.
- 4. More intensive root respiration was found in the case of nitrate nutrition whereas the change in nitrogen nutrition did not affect the shoot respiration.

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Author's address
Dr Jadwiga Zajączkowska
Institute of Sylviculture,
Warsaw Agricultural University,
ul. Rakowiecka 26/30
02-528 Warszawa, Poland

Wpływ zmiany źródła azotu na fotosyntezę i oddychanie siewek sosny zwyczajnej (Pinus silvestris L.)

#### Streszczenie

Badano intensywność fotosyntezy i oddychania siewek sosny zwyczajnej, wyhodowlanych w kulturze wodnej w warunkach niedoboru azotu, bezpośrednio po dostarczeniu azotu w amonowej bądź azotanowej formie. Ponadto oznaczano intensywność fotosyntezy i oddychania siewek po zmianie dotychczas dostępnej formy azotu. U siewek sosny z objawami niedoboru azotu stwierdzono zmianę intensywności fotosyntezy już po 10 minutach od wprowadzenia do pożywki amonowej bądź azotanowej formy azotu, przy czym zwiększyła się intensywność fotosyntezy siewek, którym dostarczono azot w formie amonowej. Zmiana formy azotu spowodowała również szybką zmianę intensywności fotosyntezy siewek. Nie stwierdzono wpływu zmiany pożywki na intensywność oddychania pędu. Stwierdzono natomiast nieco wyższą intensywność oddychania korzeni siewek przeniesionych z pożywki nie zawierającej azotu na pożywkę z azotem w formie azotanowej w porównaniu z siewkami przeniesionymi na pożywkę z azotem amonowym. Podobnie nieco wyższą intensywność oddychania wykazały korzenie siewek przeniesionych z pożywki zawierającej azot w amonowej formie na pożywkę z azotem w formie azotanowej w porównaniu z siewkami przeniesionymi z pożywki zawierającej azot azotanowy na pożywkę z azotem amonowym.