The effect of nitrogen fertilization on flag leaf and ear photosynthesis and grain yield of spring wheat

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ABSTRACT

In a three-year field experiment the assessment of leaves and ears photosynthesis rate in spring wheat was made using a LI-COR 6400 portable photosynthesis system. The photosynthetic rate of spring wheat was affected by cultivars, nitrogen fertilization and weather conditions. We generally found a negative correlation between the yield of spring wheat and the rate of photosynthesis in flag leaves in phases 39–55 BBCH (the strength of this effect depended on the level of nitrogen fertilization). Strong negative correlation occurred for cv. Bryza in phases: 39–51 BBCH in treatment fertilized with lower dose of nitrogen and in phase 39–41 BBCH for dose 120 kg/ha. There was a significant negative correlation for cv. Tybald only in phase 39–41 BBCH for higher dose of nitrogen and 52–55 BBCH for lower dose. Our studies show that the photosynthetic activity of flag leaves decreased from the booting (39–41 BBCH) to heading stage (52–55 BBCH), and their function was taken over by ears. Contrary to flag leaf, in wheat ears the intensity of photosynthesis correlated positively with grain yield for most of the studied period (52–65 BBCH, with highly significant correlation at 56–57 BBCH and – only for high nitrogen treatments – at 59–61 BBCH; a negative correlation was generally observed at a later phase, i.e. at 65–69 BBCH). Contrary to flag leaf photosynthesis, the intensity of this process in wheat ears (at the heading and flowering stages) seems highly relevant for grain yield. High positive correlation was noted in 56–57 BBCH and 61–65 BBCH. However, the study was done in field conditions and for definitive conclusions observations over a longer period would be desirable.

Keywords: gas exchange; Li-COR; spring cereals; *Triticum aestivum*

Crop yield is determined by the efficiency of photosynthesis, assimilates transport and distribution. Nitrogen ions play a key role in these processes. Nitrogen fertilization contributes to leaf area expansion, and affects leaf habit and longevity, thus influencing the course and efficiency of photosynthesis (Uzik and Zafajowa 2000).

Leaf photosynthesis rate is affected by leaf position on the plant and in winter wheat the flag leaf and the second leaf are photosynthetically the most active (Olszewski et al. 2008). The flag leaf stays longest on the plant and makes a major contribution to the grain yield in cereals. Flag leaf damage caused by pathogenic fungi results in yield decrease. Flag leaves are characterized by long-term photosynthetic activity, which is particularly important during grain filling when older leaves die off (Loss and Siddique 1994, Turner

1997). Under favorable conditions, approximately 70–90% of the total grain yield is derived from the photosynthates accumulated during grain filling (Inoue et al. 2004).

The photosynthetic activity of flag leaves decreases with age, and their function may be taken over by ears. During the flowering stage, photosynthesis of the ears has a stronger impact on grain yield, compared to the flag leaves.

In view of the facts above, the aim of this study was to determine the photosynthesis rate of flag leaves and ears as well as the grain yield in two spring wheat cultivars (awned and awnless) grown under different nitrogen fertilization levels.

MATERIAL AND METHODS

A two-factorial experiment with four replications was carried out at the Agricultural Experiment Station in Bałcyny (northeast Poland), in 2007–2009 designed as split-blocks with 4 replications. A field experiment was established on Haplic Luvisoil originating from boulder clay (IUSS 2006). The soil had very high available phosphorus levels, moderately high potassium levels, and high to very high magnesium levels (Table 1). The experimental factors were two cultivars of spring wheat (*Triticum aestivum* L.), Bryza (awned) and Tybalt (awnless), and two levels of nitrogen fertilization, 60 and 120 kg N/ha.

Nitrogen fertilizer was applied as 34% ammonium nitrate, pre-sowing at a rate of 60 kg N/ha in all treatments. Treatments with higher fertilization levels received a supplemental (top-dressing) rate of 60 kg N/ha at the stem elongation stage (31–32 BBCH). Phosphorus-potassium fertilizers were applied before sowing according to relevant guidelines and recommendations. The forecrop for cereals was winter rapeseed in the first and third year of the study, and potato in the second year. Seeding depth was approximately 2–3 cm, row spacing was 11 cm, seeding rate was 500 germinating kernels of spring wheat per meter.

During the growing season, the rate of flag leaf photosynthesis was determined using a LI-COR 6400 portable gas analyzer (Lincoln, USA), at a constant CO_2 concentration of 400 ppm and light intensity of 1000 µmol/m^2/s. The photon source was a LED Light Source lamp with the main spectrum peak at 670 nm and a second peak at 465 nm. Flag leaf measurements were performed in five replications, on randomly selected plants from each replicated plot, at three growth stages: booting (39–41 BBCH), first awns visible (49–51 BBCH) and beginning of heading (52–55 BBCH). Ear net photosynthesis (on main stem) was measured at the following growth stages: beginning of heading (52–55 BBCH), middle of heading (56–57 BBCH), end of heading (58–59 BBCH), beginning of flowering (59–61 BBCH), full flowering (61–65 BBCH), end of flowering (65–69 BBCH), watery ripe (69–71 BBCH) and early milk (71–73 BBCH). The results were presented as average values for a given growth stage (BBCH scale).

The data for grain yield, gas exchange parameters were processed statistically using a multiple range test to identify homogeneous subsets of means at a significance level of α = 0.05. Correlations between grain yield and the net photosynthetic rates of spring wheat leaves and ears at the analyzed growth stages were also determined. All calculations were done with Statistica software, version 10 (www.statsoft.com).

RESULTS AND DISCUSSION

Slightly higher net photosynthetic rates in flag leaves were noted in treatments with a higher nitrogen fertilization level (Figure 1) which is in accordance with previous reports suggesting that the rate of photosynthesis decreases under nitrogen deficiency (Gyuga et al. 2002).

The highest rate of photosynthesis in flag leaves of both cultivars of spring wheat was observed at the boot stage (flag leaf sheath extending and swelling), and it decreased at successive growth

Table 1. Selected physicochemical properties of soil used in the experiment

Year of investigation	N total (g/kg)	pH (1 mol KCl/L)	Available forms of macronutrients (mg/kg)		
			P	K	Mg
2007	1.09	5.9	122.8	206.8	87.0
2008	1.01	6.5	113.4	198.5	91.0
2009	0.89	6.0	125.4	186.2	82.0

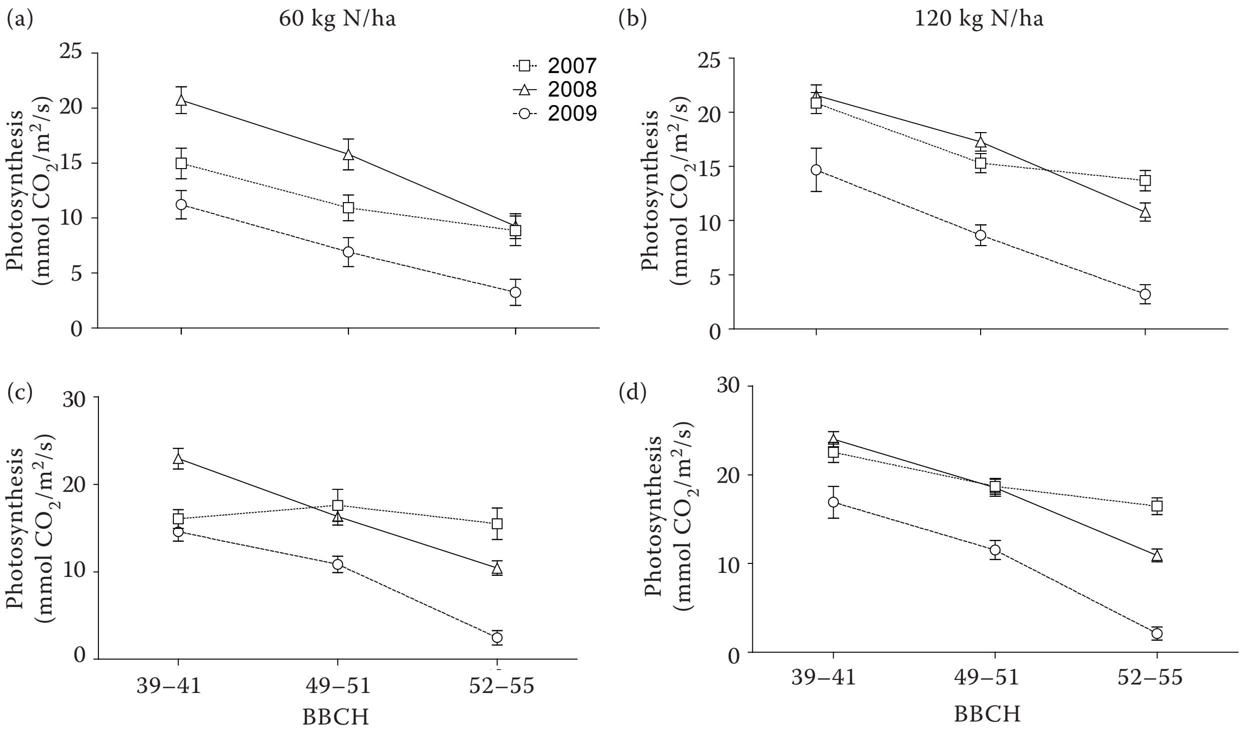

Figure 1. Flag leaf photosynthesis of spring wheat: (a, b) cv. Bryza; (c, d) cv. Tybalt. Bars show ± standard deviation

stages in all years of investigation (Figure 1). The photosynthetic function of ageing flag leaves was probably taken over by ears (Figure 2). In the first and third year, ear photosynthesis was similar in both spring wheat cultivars and at all growth stages (Figure 2). In the second year, at the flowering stage (59–69 BBCH), the rate of photosynthesis was higher in the ears of cv. Bryza. A higher level of nitrogen fertilization enhanced ear photosynthesis only during flowering. The rate of photosynthesis decreased steadily at later stages. At the watery ripe stage (69–71 BBCH), CO_2 release into the air exceeded CO_2 uptake, as shown by the negative values for net photosynthesis. Such patterns were also observed by Olszewski et al. (2008). In a study by Jiang et al. (2006), the rate of ear photosynthesis in awnless barley dropped to zero which – according to the authors – points to the significance of awns for proper grain carbohydrate balance. However, in our own studies the photosynthetic activity of ears in awnless spring wheat cv. Tybalt did not differ from that measured in awned cv. Bryza, therefore we conclude that the observation made by Jiang et al. (2006), does not hold true across other barley cultivars.

Tambussi et al. (2007) showed that ears photosynthesis could contribute substantially to grain-filling. Various parts of ear spike (glumes, awns, etc.) show different photosynthetic capacity determined by their morphology, development, and metabolic capacity.

In our experiment, the highest grain yield was achieved in 2007 and 2009 (Table 2). Throughout the experiment, the awnless form of cv. Tybalt was characterized by a significantly higher (by 9–16%) grain yield than cv. Bryza. Higher level of nitrogen fertilization made bigger yield of spring wheat grain.

No positive correlation was found between the grain yield of the studied cultivars of spring wheat and the rate of photosynthesis in flag leaves (Table 3), regardless of nitrogen fertilization. We found a significant negative correlation in cv. Bryza with 60 kg N/ha fertilization (at all growth stages) and in stage (39–41 BBCH) for two spring wheat cultivars under high nitrogen fertilization. There was no such relationship at the other growth stages, or a negative correlation was encountered, pointing to the low contribution of the photosynthesis to the yield-forming process. This is very interesting because earlier studies revealed that photosynthesis of leaves is responsible for yield (Loss and Siddique 1994, Turner 1997, Inoue et al. 2004). The current study was preceded by a preliminary analysis which revealed that leaf photosynthesis rates decreased rapidly from the middle of heading. It was assumed

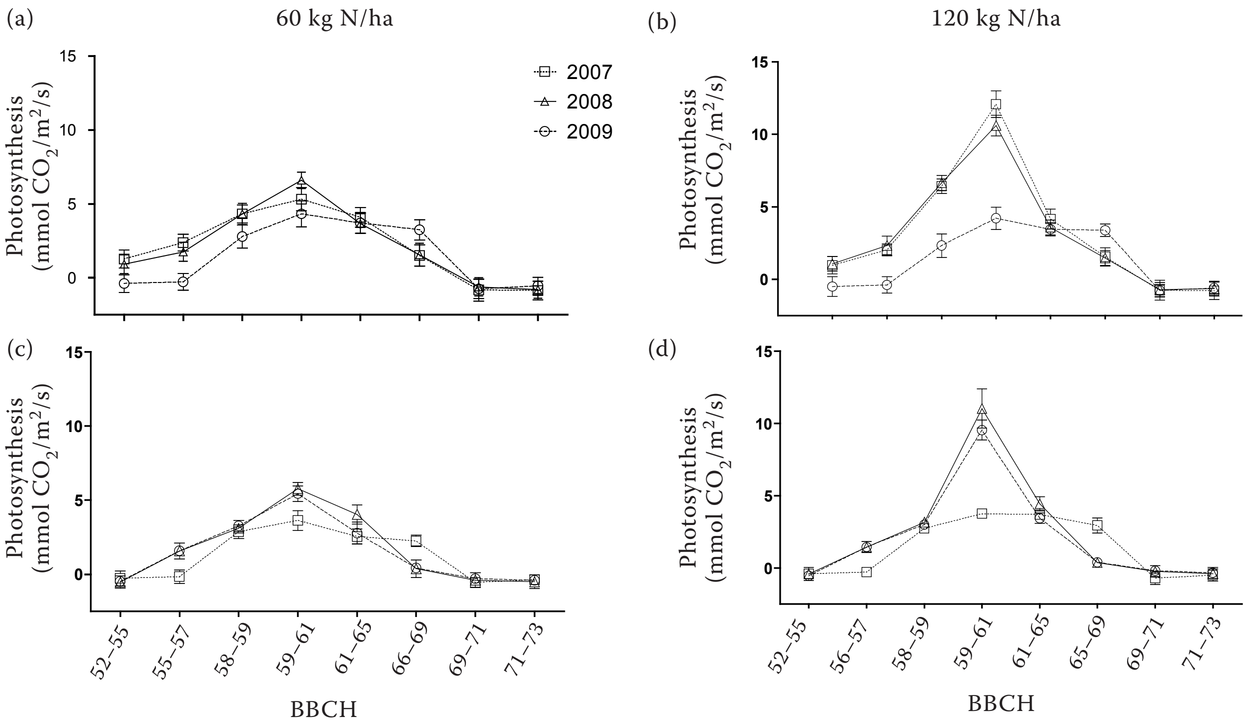

Figure 2. Ears photosynthesis of spring wheat: (a, b) cv. Bryza; (c, d) cv. Tybal. Bars show ± standard deviation

that low photosynthetic efficiency does not affect wheat grain yield, and therefore the photosynthetic activity of flag leaves was analyzed only until it reached a zero reading. No changes were noted in the rate of flag leaf photosynthesis at later growth stages, and so the data are not presented. A statistical analysis also revealed a positive correlation between the rate of ear photosynthesis and wheat yield. A significant correlation was observed between grain yield and the rate of photosynthesis in wheat ears at the heading (56–57 BBCH) and flowering (59–61 BBCH) stages (Table 3). At later phases after blossoming (anthesis) (61–65 BBCH) till water maturity (71 BBCH) (water rape) photosynthesis of ears slightly affected yield of crop (or did not affect at all). There are no publications presenting similar; results the therefore presented results need verification in completely controlled conditions.

Under low nitrogen supply, plants use photosynthesis to maintain life functions not related to the yield-forming process. Under high nitrogen supply, the photosynthetic activity of ears contributes to an increase in the grain yield. Similar results were reported by Abbad et al. (2004) who found that whole-ear photosynthesis correlated better with grain yield than flag leaf photosynthesis. Similar conclusions were drawn by Wang et al. (2001); in their study the net photosynthetic rates of winter wheat ears decreased at 12 days after flowering.

The contribution of ear photosynthesis to grain filling, although studied extensively, remains unclear (Tambussi et al. 2007, Maydup et al. 2010). Photosynthesis occurs mainly in flag and penultimate leaves, and ears. The assimilates can also be remobilized from stems (Álvaro et al. 2008, Ehdaie et al. 2008). According to Maydup et al. (2010) ear photosynthesis is an important contributor to wheat grain yield under both stress and nonstress conditions.

Table 2. Spring wheat grain yield (t/ha)

Treatment	Year of the study			
	2007	2008	2009	2007–2009
Mean values for cultivars				
Cv. Bryza	6.5[a]	4.9[a]	6.6[a]	6.0[a]
Cv. Tybalt	7.1[b]	5.7[b]	7.2[b]	6.7[b]
Mean values for nitrogen fertilization				
60 kg N/ha	6.2[a]	5.2[a]	6.3[a]	6.0[a]
120 kg N/ha	7.4[b]	5.4[a]	6.7[b]	6.5[b]

Homogeneous groups of cultivars and treatments (in years of investigation), *LSD* (α = 0.05)

Table 3. Coefficients of correlation between grain yield and photosynthesis rate in spring wheat leaves and ears in different growth stages (39–73 BBCH)

Cultivar	Nitrogen fertilization (kg N/ha)	Leaf			Ear							
		39–41	49–51	52–55	52–55	56–57	58–59	59–61	61–65	65–69	69–71	71–73
Bryza	60	–0.91**	–0.90**	–0.62*	0.31	0.86**	0.49	0.69*	0.40	–0.92**	0.18	–0.32
	120	–0.92**	–0.41	0.23	0.51	0.95**	0.62*	0.95**	0.69*	–0.91**	0.43	–0.04
Tybalt	60	–0.07	–0.22	–0.62*	0.26	0.89**	0.52	0.78*	0.38	–0.77*	0.22	–0.30
	120	–0.63*	–0.45	–0.13	0.42	0.90**	0.52	0.96**	0.41	–0.87**	0.49	0.08

*α = 0.05; **α = 0.01

In our study, the rates of photosynthesis varied between years depending on weather conditions (Table 4). At growth stages 52–55 BBCH, very low average rainfall (9 mm) was noted in 2007, and there was no rain in 2008; mean daily temperatures in 2007 and 2008 were 2.4°C and 1.8°C higher, respectively, than the long-term average (1981–2010). In the last year of the study (2009), at growth stages 52–55 BBCH, rainfall total was by 16 mm lower and mean daily temperature was by 3.7°C lower than the respective long-term averages. The rates of photosynthesis decreased at growth stages 52–55 BBCH in all years of the study. As shown by weather data, spring wheat plants were cultivated in low water availability compared to multi-year data, which probably caused decreased photosynthetic activity in flag leaves and increased the contribution of ear photosynthesis to grain filling.

Table 4. Weather conditions in years of investigation data from the Meteorogical Station in Bałcyny, Poland

Month	Decade of month	Mean temperature (°C)				Precipitation total (mm)			
		2007	2008	2009	mean 1981–2010	2007	2008	2009	mean 1981–2010
March	1	4.0	3.6	1.5	0.3	13.1	20.0	12.0	12.0
	2	4.9	3.2	1.2	1.4	11.6	14.2	14.4	11.3
	3	7.0	1.9	2.9	3.4	3.2	12.9	41.6	9.4
April	1	4.6	6.0	8.7	5.7	23.8	21.9	3.7	10.4
	2	8.3	7.2	8.1	7.4	3.0	10.9	0.0	14.2
	3	9.0	9.8	12.5	10.2	0.0	1.0	0.0	7.0
May	1	8.5	11.4	11.5	12.1	26.7	41.8	11.5	21.9
	2	12.9	12.0	11.0	12.1	40.4	2.2	1.8	17.8
	3	19.1	13.3	13.9	13.8	12.6	4.4	76.3	30.9
June	1	18.2	17.6	12.1	15.9	9.0	0.0	12.1	28.1
	2	18.6	14.9	13.9	15.7	19.2	11.2	46.4	29.7
	3	15.8	17.0	18.2	16.3	32.6	16.6	46	28.3
July	1	15.7	17.2	18.6	17.8	95.7	10.8	60.0	36.1
	2	19.5	18.4	19.1	18.6	15.8	23.8	10.3	24.3
	3	17.3	19.2	19.1	18.9	65.0	12.4	11.9	29.6
August	1	18.3	18.2	19.7	18.6	11.2	43.0	13.4	24.8
	2	19.6	19.0	17.8	18.4	47.0	26.0	11.2	21.8
	3	17.0	16.2	18.1	16.5	22.8	34.1	1.1	30.5

The period just before anthesis is a very sensitive period in wheat and the photothermal quotient (radiation/temperature) has a major influence on grain number and thereby yield (Fischer 1985). Szwejkowski et al. (2002) describing the climate of Pojezierze Warmińsko-Mazurskie (the place of the current study) emphasizes that the typical climatic features of this region are: dry springs, intensive rains during harvest, big amplitude of temperatures, shortage of warmth and insolation.

In conclusion, our findings suggest that higher nitrogen fertilization levels contributed to an increase in net photosynthesis rates in spring wheat flag leaves and ears. A statistical analysis revealed a positive correlation between the grain yield of spring wheat and the rate of ear photosynthesis.

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