

## Yield and grain quality of spring barley as affected by biomass formation at early growth stages

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### ABSTRACT

Timely and reliable prediction of grain yield and quality of spring barley represents a key prerequisite for effective crop management. Within this study we evaluated the relationships between yield components, grain quality, biomass production and the number of tillers in different growth stages. For this purpose, in three years (2011–2013) multifactorial field trials focused on the combined effects of cultivar, sowing density and nitrogen nutrition were conducted. Based on ANOVA it was found that the formation of grain yield was affected by individual factors in the following order of importance: year, nitrogen, cultivar and sowing rate. The final grain yield significantly correlated both with the number of tillers and dry weight of above-ground biomass per unit area. The best estimation of yield provided both parameters at early growth stage ( $R = 0.83^{**}$  and  $0.81^{**}$  for number of tillers and the above-ground biomass at BBCH 25). The grain protein content was inversely related to early growth parameters ( $R = -0.64^{**}$  and  $-0.41^{**}$  for number of tillers and above-ground biomass at BBCH 25). Based on the comparison of relationships between the years, it can be concluded that the early growth of barley and tiller differentiation is a key parameter for the formation of yield and grain quality.

**Keywords:** *Hordeum vulgare* L.; above-ground biomass; tillering; grain yield formation; grain protein content

Spring barley yield and quality predictions are of major interest to the growers and the malting industry in order to allow effective crop management and convenient organization of barley grain production. To attain higher effectiveness of crop management practices, extensive research on cereal stand structure was conducted in the 1980s and 1990s (e.g. Porter 1984). The assessment of cereal stands and yield formation is usually based on the classical concept as reported by Heuser (1927/28) and later on by a number of other authors who divided grain yield into spike number per unit area, grain number per spike and grain weight. At present, this concept based on the plant number

and numbers of formed and eventually reduced tillers per stand unit area prevails in both applied research and practice. However, the concept was often criticized because it does not provide enough precise quantification of differences in stands and some authors expressed a need of innovated criteria for stand assessment (e.g. Křen 1990).

Recently, methods for the estimation of grain yield or quality, based on spectral characteristics of the barley stand in various growth stages have been developed (Wessteiner and Kühbauch 2005, Zhao et al. 2005). These methods facilitate the evaluation of whole field area in very short time. However, a fundamental problem with the spec-

tral characteristics is the lack of generality. This is primarily due to a lack of understanding of the relationships between the parameters of biomass production or development and reduction of basic modular units (tillers) in different growth stages and yield components or quality.

Therefore, this study is focused on analyzing the relationships between grain yield, its components and quality parameters and: (i) parameters characterizing the increase in biomass per unit area during the growing season; (ii) parameters describing the change in the number of structural units of the canopy (tillers, stems, spikes).

## MATERIAL AND METHODS

Evaluation of the canopy development and yield formation of spring barley was performed in small-plot field experiments established at Kroměříž locations in Central Moravia within the period of three years (2011–2013). The location is characterized by mean annual temperature 8.7°C and precipitation 599 mm. The soil type is Luvic chernozem and texture silt loam. The previous crop was grain maize. During the growing season the standard measures of plant protection were used.

The experimental treatments (Table 1) were established in five replications to ensure contrasting differences in the stand density and nitrogen nutrition. Three replications were used for assessment of grain yield and quality parameters and two were used for plant sampling, which enabled the analyses of the canopy structure. In sampling plots, squares of 0.25 m<sup>2</sup> (0.5 × 0.5 m) were marked out to obtain plants for analyses of stand structure in five developmental stages: (i) mid tillering BBCH 25; (ii) beginning of stem elongation BBCH 31; (iii) end of stem elongation (flag leaf stage/beginning of booting) BBCH 39; (iv) medium milk BBCH 75, and (v) full ripeness BBCH 91.

Analyses of stand structure which were done manually involved assessment of the numbers of plants and tillers and the amount of above-ground dry biomass. The harvest was carried out using a small plot harvester Sampo 2010 equipped with automatic weighing and sampling system (Sampo Rosenlew, Pori, Finland). After harvest the weight of thousand grains was determined using grain counter Contador (Pfeuffer GmbH, Kitzingen, Germany) and for the proportion of grains above 2.5 mm on the Sortimat screeningmaschine (Pfeuffer GmbH, Kitzingen, Germany) was used. The grain samples were then used for analyses of the protein content (N × 6.25) with elemental analyzer Leco (LECO, St. Joseph, USA). The four- and two-way ANOVA (followed by the Tukey's post-hoc test) and correlation analysis (Pearson's correlation coefficient *R*) were completed using the Statistica 12 software (StatSoft Inc., Tulsa, USA).

## RESULTS

**Yield response to experimental treatments and year.** Four-way analysis of variance showed a significant effect of all the assessed factors on yield in the order year, N fertilizer dose, cultivar and sowing rate (Table 2). Two-way interactions were also statistically significant, especially those between year and cultivar, year and nitrogen dose and year and sowing rate. Three- and four-way interactions were statistically insignificant. The effect of nitrogen dose and sowing density on grain yield within individual year and cultivar is shown in Figure 1. The highest yield level was obtained in 2011 across all cultivars, sowing rates and nitrogen fertilization. On the contrary, the lowest yield was observed in 2013. The cultivars Prestige and Sebastian showed higher inter-annual yield variation compare to relatively stable yields in cv. Bojos. The effect of nitrogen fertilization was relatively consistent within individual years

Table 1. Characteristics of experimental treatments of spring barley

Factor	Number	
Cultivar (tillering intensity)	3	Prestige (low), Bojos (middle), Sebastian (high)
Nitrogen fertilisation prior to sowing (kg N/ha)	3	0, 45, 90 (calcium-ammonium-nitrate )
Seeding rate – millions germinating seeds per hectare	3	2.5; 4.0; 5.5
Year/date of sowing/date of harvesting	3	2011/16.3./4.8.; 2012/16.3/28.–31.8.; 2013/17.4/6.8.

Table 2. Analysis of variance of factors influencing spring barley grain yield

Source	Sum of squares	df	Mean squares	F-ratio	P-value
Year (A)	340.1	2	170.1	572.7	< 0.001
Nitrogen dose (B)	281.5	2	140.7	474.0	< 0.001
Seed rate (C)	5.6	2	2.8	9.4	< 0.001
Cultivar (D)	29.0	2	14.5	48.9	< 0.001
A × B	14.1	4	3.5	11.8	< 0.001
A × C	5.0	4	1.2	4.2	0.003
A × D	19.8	4	5.0	16.7	< 0.001
B × C	0.3	4	0.1	0.2	0.931
B × D	2.9	4	0.7	2.5	0.047
C × D	0.2	4	0.0	0.2	0.965
A × B × D	1.4	8	0.2	0.6	0.789
A × C × D	1.6	8	0.2	0.7	0.715
A × B × C	1.1	8	0.1	0.5	0.883
A × B × C × D	1.9	16	0.1	0.4	0.982
Residual	48.1	162	0.3		

across the monitored cultivars and sowing rates. Generally it can be stated that the yield increase from the dose 0 kg N/ha to 45 kg N/ha was greater than the increase from 45 kg N/ha to 90 kg N/ha. Lower yield response to nitrogen fertilization was reported in 2012. The effect of sowing rate on yield was generally very low and only in the year 2013 yield tended to decrease with lower sowing density.

**Correlation analysis between yield and biomass or structural parameters of canopy.** It is evident from Table 3 that highly significant correlations were found between the final grain yield and number of tillers, and also dry weight of the above-ground biomass per m<sup>2</sup>. In the number of tillers, higher values of correlation were found in the first three dates of assessment, on the contrary, the above-ground biomass showed higher values of correlation coefficients with yield also in the second part of vegetation. Detailed analysis of relationships between the number of tillers, dry weight of the above-ground biomass and grain yield showed that these relationships have an asymptotic character (Figures 2a,b). For this reason it was more suitable to use a non-linear regression (sigmoidal function  $y = a/(1 + \exp(-(x - x_0)/b))$ ; where  $a$ ,  $b$  and  $x_0$  – parameters of function;  $y$  – yield;  $x$  – number of tillers or dry weight of the above-ground biomass) for fitting of these relationships. This leads to increased

significance of the correlation coefficients compare to linear regression. At the same time it is apparent that in individual years a shift of the relationships occurs, namely for a more favourable year (2011) to higher yields, and for a less favourable year (2013) to lower yields.

The number of spikes (1/m<sup>2</sup>) at harvest correlated better with the number of tillers (1/m<sup>2</sup>) than with the above-ground biomass (Table 3). The values of the correlation coefficients for relationship to the number of tillers increased during vegetation. On the other hand, correlations with the above-ground biomass stagnated more or less during vegetation period.

Spike productivity (g/spike) correlated highly significantly with the number of tillers and the above-ground biomass at mid tillering (BBCH 25). Later during vegetation, correlations with both parameters were low and at harvest they were significantly inverse. In the number of grains per spike, practically identical trends in the change of correlation coefficients were observed.

Correlations with the thousand-grain weight were inverse for both the relationship to the number of tillers and the above-ground biomass. The number of tillers showed strong and significant effect compared to the above-ground biomass. The highest values of correlation coefficients for this yield component were found at the end of stem elongation (BBCH 39).

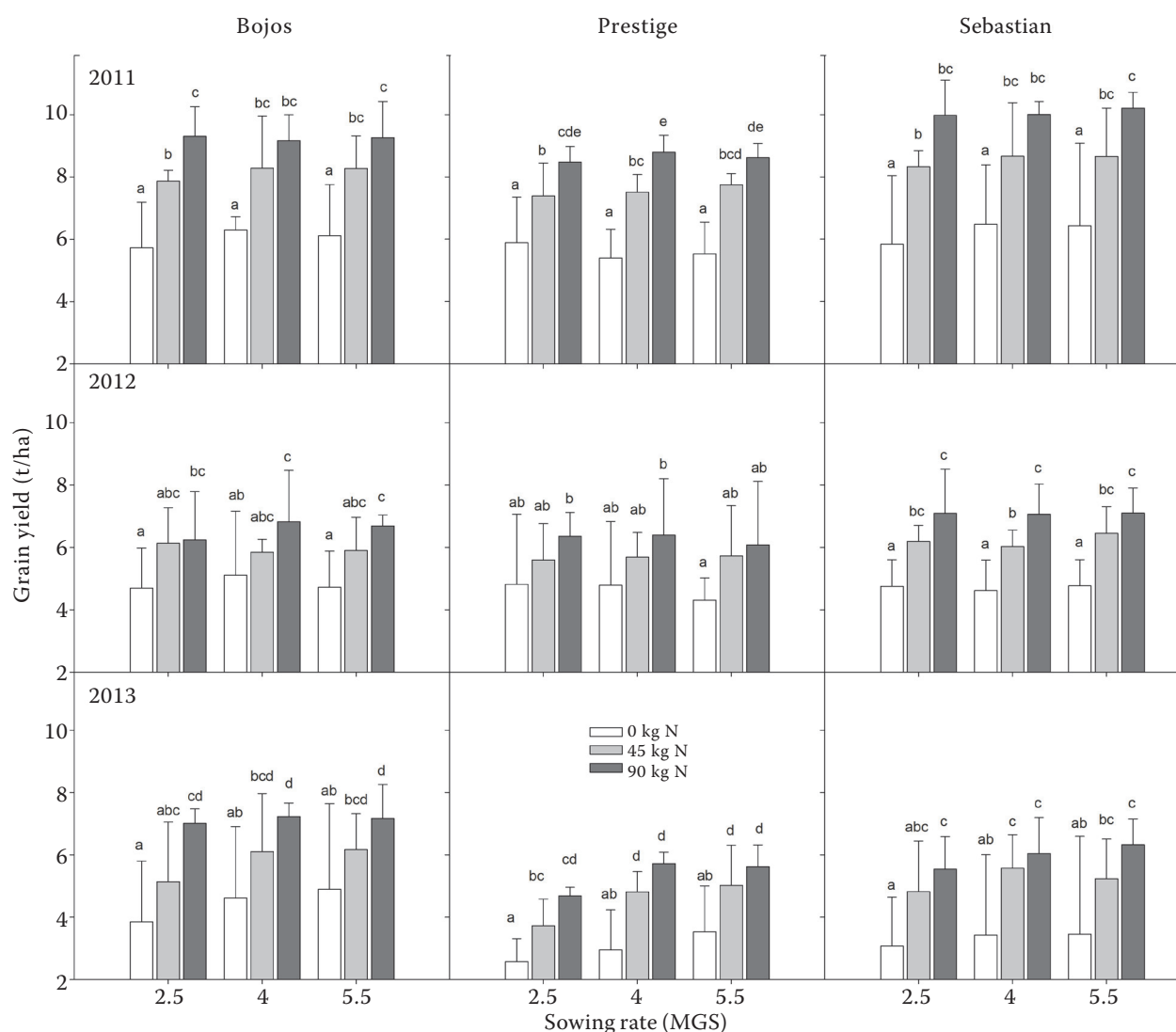


Figure 1. Grain yield response to nitrogen nutrition, sowing rate in three cultivars of spring barley separately within individual years. The means (bars) and 95% confidence intervals are presented ( $n = 3$ ). Different letters denote statistically significant differences between nitrogen nutrition and sowing rate treatments within individual years and cultivars (Tukey's post hoc test following two-way ANOVA;  $P = 0.05$ )

#### Correlation analysis between grain quality parameters and biomass or canopy structure.

The correlations of the number of tillers and the above-ground biomass with the assessed parameters of grain quality are shown in Table 4 which reveals that the proportion of grain > 2.5 mm was positively affected by tillering and intensive growth at the beginning of vegetation (BBCH 25). The trends in the changes of correlation coefficients during vegetation were similar to those with spike productivity. In contrast, in grain protein content highly significant inverse correlations were found for the number of tillers and the above-ground biomass during tillering (BBCH 25), which gradually decreased during vegetation. The number of

tillers showed stronger effects compared to the above-ground biomass. However, when we analysed in detail the relationship between the protein content in grain and number of tillers or dry weight of the above-ground biomass at BBCH 25, it was evident that this relationship is significantly modified by year (Figure 2c,d). It is apparent that early production of biomass in 2013 had the most significant effect on protein content in barley grain. A negative relationship was also observed in the year with higher productivity (2011), but the relationship is significantly shifted to the lower protein content in grain. Similar character of relationships was also found for the number of tillers at growth stage BBCH 25.

Table 3. Correlations between the number of tillers (A) and above-ground dry biomass (B) per m<sup>2</sup> and grain yield and its components in spring barley (2011–2013)

Yield parameter ( <i>n</i> = 81)		Growth stage BBCH				
		25	31	39	75	91
Grain yield (t/ha)	A	0.833**	0.730**	0.698**	0.261*	0.307**
	B	0.810**	0.698**	0.791**	0.581**	0.444**
Number of spikes per m <sup>2</sup>	A	0.361**	0.541**	0.563**	0.710**	—
	B	0.300**	0.543**	0.437**	0.402**	0.653**
Spike productivity (g)	A	0.351**	0.134	0.110	−0.369**	−0.550**
	B	0.330**	0.094	0.247*	0.094	−0.244*
Number of grains per spike	A	0.420**	0.205	0.196	−0.316**	−0.496**
	B	0.382**	0.131	0.286**	0.112	−0.233*
Thousand grain weight (g)	A	−0.441**	−0.451**	−0.577**	−0.391**	−0.400**
	B	−0.268*	−0.212	−0.219*	−0.052	0.001

## DISCUSSION

Estimation of individual yield components and the grain yield as a whole at early stages of barley growth is one of the most important prerequisites

for optimisation of crop management. The classical methods for the assessment of stand structure, based on counting plants and stems (spikes) per unit area are labour consuming and interpretation of the results is often difficult. These methods

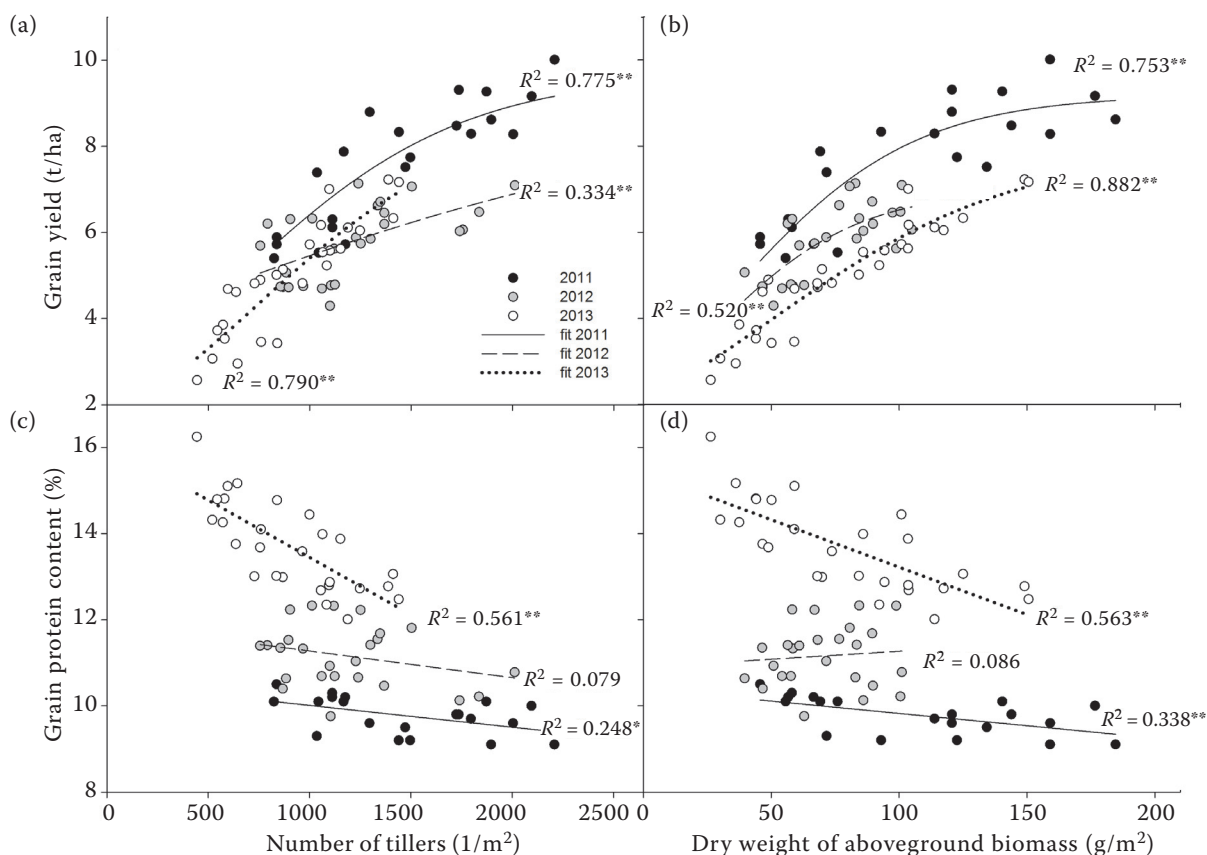


Figure 2. Relationships between dry weight of above-ground biomass (b, d), number of tillers (a, c) determined at tillering (BBCH 25) and total grain yield (a, b) and grain protein content (c, d). The relationships were fitted separately for individual years. \**P* = 0.05; \*\**P* = 0.01



Table 4. Correlations between the number of tillers (A) and above-ground dry biomass (B) per m<sup>2</sup> and selected characteristics of spring barley grain quality (2011–2013)

Grain quality parameter ( <i>n</i> = 81)		Growth stage BBCH				
		25	31	39	75	91
Protein content in grain (%)	A	–0.640**	–0.552**	–0.551**	0.064	–0.026
	B	–0.409**	–0.345**	–0.405**	–0.005	0.148
Proportion of grains above 2.5 mm (%)	A	0.266*	0.102	0.023	–0.360**	–0.420**
	B	0.343**	0.083	0.205	0.187	0.020

do not allow assessing the relationships within the stand (inter- and intra-plant competition). These relationships are more reflected in the values of the total above-ground biomass (Křen et al. 2007). The reason for it is that the size structure of tillers and particularly the proportion of strong tillers play a crucial role in determining the final number of ears and yield (Křen 1990). Recently, there has been rapid development of the remote sensing methods which serve to facilitation of agronomic decisions (reviewed by Hatfield et al. 2008). However, remote sensing assessments are usually closely related to the above-ground biomass or leaf area (Alvaro et al. 2007). All these facts point to the need for understanding the relationships between the dynamics of above-ground biomass and formation of yield components. Therefore, we focused in this study on the correlation analysis of relationships between grain yield or quality and biomass or structural parameters of the canopy, where sufficient range of yield components was achieved by combined effects of cultivar, sowing density, nitrogen nutrition and year.

The most important factors affecting yield formation were year, nitrogen nutrition and cultivar. The yield results show differences in inter-annual yield variation between cultivars with the highest yield stability in cv. Bojos. From our results, it is obvious that the cultivar with high plasticity should provide, in years unfavorable for achieving a high spike number, sufficient compensation by increasing the spike productivity. Similarly García del Moral et al. (2003) showed that yield stability of barley cultivars under different environments is closely associated with the number of grains per spike ensuring the achievement of a high number of grains per area unit also under the conditions of lower spike number.

From the results of correlation analysis, it is evident that the crucial period for the formation of

spring barley yield and grain quality is determined by the growth stages of tillering and beginning of stem elongation. The highest values of correlation coefficients for the relationships to grain yield were generally achieved during the growth stage of tillering. García del Moral and García del Moral (1995) showed that the maximum number of tillers is inversely related to the temperature during tillering. This means that cold temperature is prolonging the tillering period resulting in a higher number of tillers formed. As the tillering dynamics is affected by the length of photoperiod (Miralles and Richards 2000) the sowing date may also substantially influence the final number of tillers. In addition to temperature and sowing date, sufficient water availability is the critical factor for adequate tillering in spring barley (Svobodová and Míša 2004). Higher formation of tillers and their lower mortality may be supported also by timely nitrogen nutrition (Baethgen et al. 1995).

Although slightly higher correlation coefficients for the relationships to grain yield were achieved with the number of tillers, it is possible to conclude that the yield estimation by the above-ground biomass provides high reliability, particularly in early growth stages. This represents a significant potential for involvement of remote sensing methods in early estimation of grain yield and agronomic decision-making processes.

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