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## Influence of Sowing, Nitrogen Nutrition and Weather Conditions on Stand Structure and Yield of Spring Barley

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The processes of stand structure and yield formation of spring barley were studied under different weather conditions and crop management. The multifactorial small-plot trials focused on the combined effect of variety, sowing density and nitrogen nutrition were carried out in two years with contrast weather conditions for yield formation (2011 and 2013). Evaluation of the above-ground biomass and the segmentation of tillers into three groups was conducted in four growth stages (BBCH 25, 31, 39 and 75). The performed analyses confirmed that for effective use of inputs and high yield, it is important to create a sufficient number of strong tillers at the beginning of vegetation. In year with low proportion of strong tillers at the end of tillering (2013), the differentiation of tillers is delayed and their productivity decreases. In this year therefore, yield formation is shifted from the number of spikes to the number of grains in a spike. The comparison of barley genotypes revealed that high yield plasticity can be obtained especially in the variety Bojos, which is able to compensate effectively the changes in spike number by increased grain number in a spike. This variety is also able to create a high proportion of strong tillers even under unfavourable conditions. This knowledge could help to improve the breeding and management strategy in spring barley for the expected weather conditions in the near future, especially higher temperatures in early spring.

**Keywords:** spring barley, yield formation, tillers formation and differentiation, genotype plasticity, grain quality

### Introduction

The formation and differentiation of tillers is influenced by environmental conditions and availability of resources of which the most important are nutrients, water and light. The key period for the yield formation of spring barley is tillering when the basis of a sufficient number of productive stems and spikes is formed. Increasing frequency of unfavourable conditions at early spring, especially higher temperatures and drought periods (Trnka et al. 2004), can affect negatively the formation of spring barley yield and grain quality pa-

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rameters. Lobell and Field (2007) demonstrated, based on detrending of past yields, that barley is one of the most vulnerable crops in the context of global change with clearly negative response of global yields to increased temperature. Rötter et al. (2011) showed that also under Nordic conditions can increasing temperature considerably reduce total growth duration and yield of barley, even with adjusted earlier sowing. They also emphasized the negative effects of reduced number of rainy days in combination with increasing temperature. However, elevated temperature affects yield and grain quality of barley more significantly than changes in rainfall.

Barley varieties have been selected for optimal performance under current climatic conditions. With global change, characterized by shifts in weather patterns and increases in frequency and magnitude of extreme weather events, new ideotypes will be required with a different set of physiological traits (Semenov and Halford 2009). Thus, under changing environmental conditions, there is a need to understand yield formation and the possibilities by which barley genotypes are able to compensate the negative impacts of weather conditions and obtain stable yields under increasing variability of the environment (Del Moral and Del Moral 1995).

The ecologists use the size distribution of plants as a suitable indicator of changes taking place in the structure of plant populations. This can be expressed by a histogram showing the change of frequency of the size proportion of structural units (tillers) in time. In previous studies (Křen 1991) we described a two-peak distribution of tiller weight (Fig. 1), which allows separation of three groups of tillers: strong (later productive), intermediate (productivity yet undetermined), weak (later mostly unproductive). Tillers of the intermediate category corresponding with the position of local minimum can thus create productive stems or later die in dependence on the subsequent weather conditions and effectiveness of the management practices.

The objective of this study was to evaluate the effect of weather conditions, genotype, nitrogen nutrition and sowing rate on spring barley tillering and tiller differentiation, and to find phenotypic traits which are a prerequisite for yield stability even under conditions unfavourable for the process of tillering.

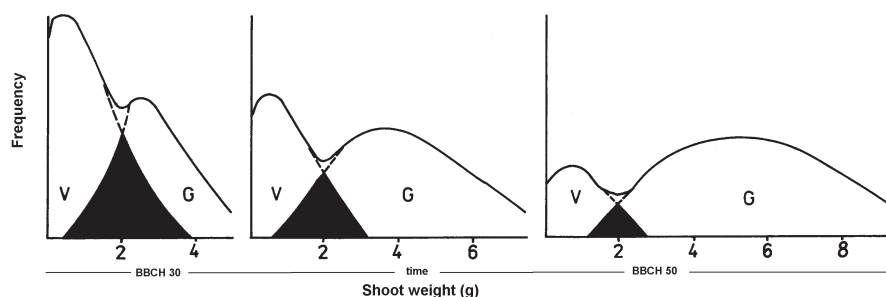


Figure 1. A schematic presentation of changes in the distribution of tiller weights during the period of generative development (BBCH 30–59). V – vegetative tillers, G – generative tillers (stems); the dark area represents tillers that can become, depending on availability of resources, either vegetative or generative (according to Křen 1991)

## Materials and Methods

Evaluation of the stand development and yield formation of spring barley was performed in small-plot field experiments established at locations Kromeriz in Central Moravia in two years 2011 and 2013 differing in weather conditions. The location is characterized by warm, slightly humid climate with mean annual temperature of 9.1 °C and precipitation of 567.7 mm. The soil type is Luvi-haplic Chernozem and the texture is clay-loam. The preceding crop was grain maize. During the growing season the standard crop protection was used.

Experiments were conducted as contrast variants which took into account the differences in stand density and in the nutritional status of plants. Experimental variants represented all combinations of following factors: spring barley variety differing in tillering ability (Prestige, Bojos, Sebastian), nitrogen fertilization with calcium-ammonium-nitrate prior to sowing (0 and 90 kg N ha<sup>-1</sup>) and sowing rate (2.5, 4 and 5.5 millions of germinating seeds – MGS per hectare). Each experimental variant was established in five replications. Plot size was 10 m<sup>2</sup> and the replications were arranged in randomized block design. In each experimental variant three replications were harvested and two were used for sampling, which enabled the analyses of the stand structure. In sampling plots, squares of the size 0.25 m<sup>2</sup> (0.5 × 0.5 m) were harvested for analyses of stand structure in five developmental stages: second part of tillering – BBCH 25, beginning of stem elongation – BBCH 31, end of stem elongation – BBCH 39, medium milk – BBCH 75, ripening – BBCH 91.

Analyses of stand structure and nutritional status, which were done manually, involved: i) assessment of the numbers and weight of three group of tillers according the size and expected productivity (Fig. 1) – strong (productive), intermediate (later productive or dying) and weak (later mostly dying), ii) assessment of dry matter (DM) weight of these three groups of tillers.

The harvest was carried out using a small plot harvester Sampo 2010 equipped with automatic weighing and sampling system (Sampo Rosenlew, FI). After the harvest a thousand grains weight was determined using grain counter Contador (Pfeuffer, DE) and proportion of grains above 2.5 mm on the Sortimat screening machine (Pfeuffer, DE). The grain samples were then used for analyses of the protein content (N × 6.25) using elemental analyzer Leco (LECO, USA).

The basic statistical analyses (ANOVA, Tukey post-hoc test, and regression and correlation analyses) were done using Statistica 12 software (Statsoft, USA). Within individual years, weather data between the evaluated growth stages were taken from a permanent meteorological station (located within 500 m from experimental field; Table S1\*).

## Results

### *Weather conditions and effects on stand development*

In 2013 the tillering period was about two weeks shorter as well as the grain formation period (Table S1). This resulted in shortening of the growing season by 30 days compared to

\* Further details about the Electronic Supplementary Material (ESM) can be found at the end of the article.

2011. However, in the shorter period of tillering the sum of active temperatures was higher by 14 °C and the sum of precipitations was higher by 10 mm. This led to formation of smaller number of tillers and smaller amount of dry weight of the total above-ground biomass in all variants compared to 2011 (Fig. S1).

The length of stem elongation period was nearly the same in both years, but it was colder with more precipitations in 2013. The duration of period from heading until milk ripening was also nearly the same in both years with similar temperatures and precipitations. The rest of the growing season until ripening was by 16 days longer in 2011 with higher temperatures and more precipitations compared to 2013 when the period of grain formation was practically without precipitations (Table S1).

#### *Formation and differentiation of tillers*

The highest differences in the number of tillers per 1 m<sup>2</sup> were found between the years and between the variants at the end of tillering (BBCH 25; Fig. S1). In this growth stage, a significant effect of nitrogen nutrition on the number of tillers and above-ground biomass is obvious. These differences decreased at the end of stem elongation (BBCH 39), particularly in 2013.

Time dynamics of tiller differentiation is clearly apparent in Fig. 2 which shows the course of changes in the proportion of strong, intermediate and weak tillers within individual years. The total number of tillers decreased and the number of strong tillers increased during stand growth and differentiation in accordance with the scheme shown in Fig. 1. It is evident that in favourable year 2011 the number of strong tillers at the growth stage BBCH 25 is much closer to the final number of spikes than in 2013. This means that in 2013 there was a higher proportion of productive stems and lately spikes formed from intermediate or weak tillers. Since the end of tillering a gradual reduction of weak tillers has been proceeding as well as the change of strong and partly also intermediate tillers to productive stems (Fig. 2).

As the proportion of strong tillers at the end of tillering on the final number of spikes per area unit seems to be important indicator of tiller differentiation during the growing season, we performed a detailed analysis of this parameter in relation to year, barley genotype and nitrogen nutrition (Fig. 3). These results show an apparent interaction between year and nitrogen nutrition. While in 2013 the influence of nitrogen nutrition on the proportion of strong tillers was small, in 2011 there is a clear evidence of an increase in this proportion due to nitrogen. It is also evident that the variety Bojos creates during tillering a considerably higher proportion of strong tillers to the number of spikes than the varieties Prestige and Sebastian.

#### *The effect of year, barley variety and management on the formation of yield components*

The analysis of relationships between individual yield components and the final grain yield (Fig. 4) shows that significant correlations were obtained each year in a different yield component and were similar in all varieties investigated. In 2011, a significant correlation to yield was demonstrated in the number of spikes per unit area of stand. It is apparent that correlations for the varieties Prestige and Sebastian are shifted downwards

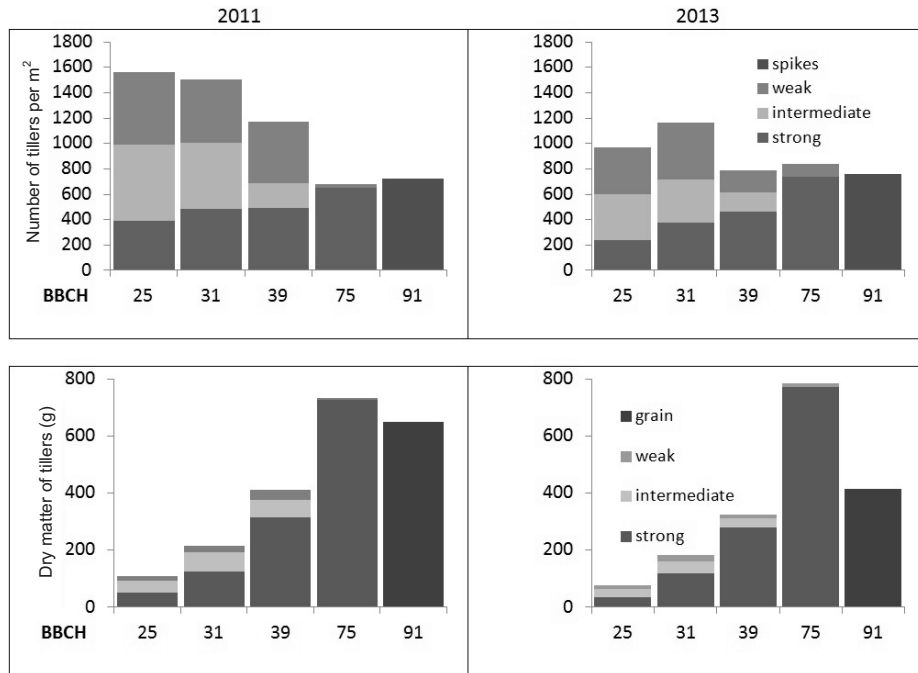


Figure 2. Changes of the numbers and dry weight of above-ground biomass of individual categories of tillers during the growing season in 2011 and 2013 (averages of three varieties, three sowing rates and two doses of N)

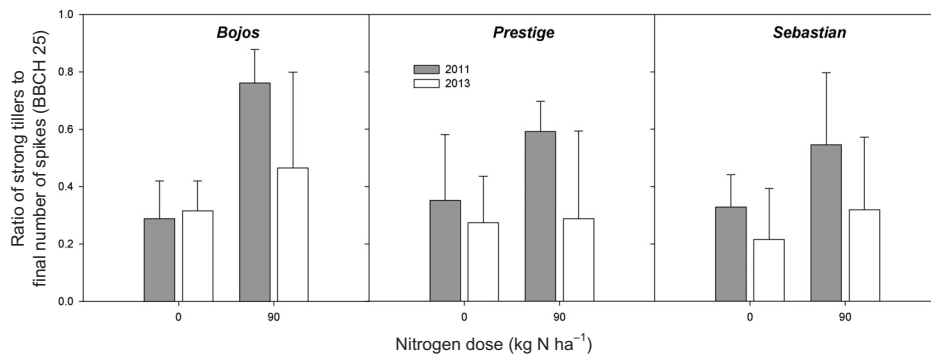


Figure 3. Ratio between the number of strong tillers at BBCH 25 and final number of spikes per area unit for individual spring barley varieties in two contrast years. The means (columns) and 95% confidence intervals (error bars) are presented ( $n = 3$ )

compared with the variety Bojos, which means that at the same number of spikes, the variety Bojos obtained a higher yield. On the other hand, grain number in a spike had a dominant effect on yield in the less favourable year 2013. These relationships for individual varieties follow each other, when the variety Bojos obtains in that year generally higher number of grains in spike and thus a higher yield.

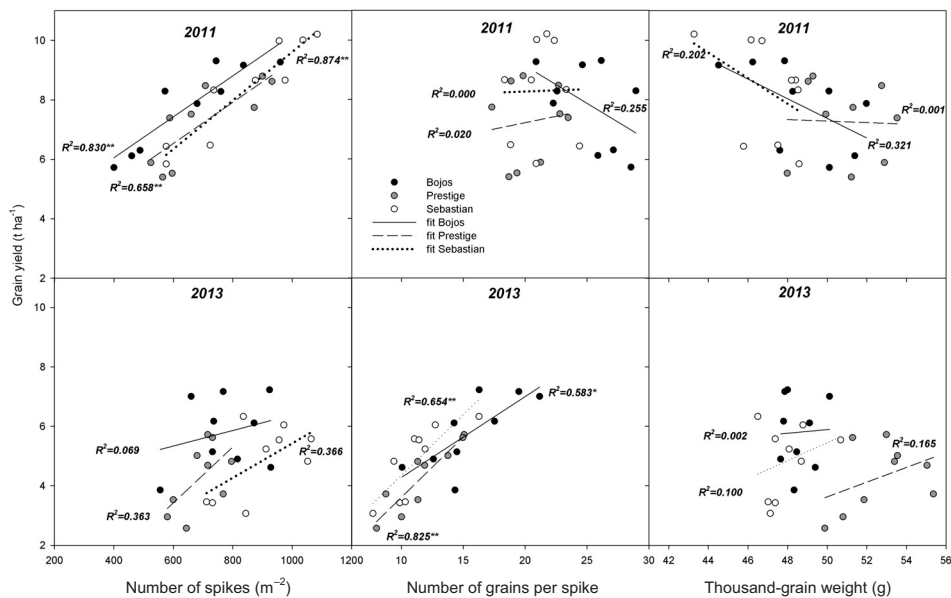


Figure 4. Relationships between individual yield components and final grain yield. The relationships were created separately for individual years studied and spring barley varieties. \* and \*\* indicate significant relationships at  $p = 0.05$  and  $p = 0.01$ , respectively

#### *Relationships between tiller differentiation, yield components and grain quality parameters*

Generally, the most significant effect on the yield and individual yield components was observed in a year and nitrogen nutrition (Fig. S2). The effect of sowing rate was generally very small and statistically insignificant. From the perspective of the individual yield components, the most important interaction was found for the effects of nitrogen nutrition and year. While the number of spikes in 2011 was significantly affected by nitrogen nutrition, in 2013 this effect was very small and insignificant. On the contrary, the number of grains in spike was affected by nitrogen nutrition only in 2013. The protein content in grain was mainly influenced by year, with significantly higher protein content in the less favourable year 2013.

The consequences of time differences in the formation and differentiation of strong tillers (proportion of strong tillers on final number of spikes) on yield components are shown

in Table 1. The importance of formation of strong tillers at the beginning of vegetation (BBCH 25) was confirmed in both the years by significant correlations between the proportion of strong tillers and grain yield. Since the beginning of stem elongation the correlation between proportion of strong tillers and yield was insignificant in 2011. In contrast, in 2013 the significant correlations were found for this relationship till the growth stage BBCH 75. In 2013 the number of tillers formed in BBCH 25 reached only 60% of the year 2011 (see Figs 2 and S1). Delayed formation resulted also in later differentiation of tillers and increase of differences in spike productivity. Correlations with thousand-grain weight were insignificant and mostly inverse. Higher differences in the development and productivity of stems led to a strongly inverse relationship with grain protein content in all growth stages, unlike of 2011, when this relationship was only observed in BBCH 25.

Table 1. Comparison of correlations between yield components and grain quality traits and the proportion of strong tillers formed at important growth stages (BBCH) from productive spikes in BBCH 91

Characteristic	% of strong tillers at specific BBCH from productive spikes (BBCH 91)							
	BBCH 25		BBCH 31		BBCH 39		BBCH 75	
	2011	2013	2011	2013	2011	2013	2011	2013
Grain yield (t.ha <sup>-1</sup> )	0.752**	0.673**	0.151	0.680**	0.300	0.804**	0.200	0.588**
Thousand-grain weight (g)	-0.120	-0.203	-0.293	-0.192	0.164	-0.027	0.204	-0.040
Protein content (%)	-0.599**	-0.618**	0.059	-0.679**	0.007	-0.697**	0.111	-0.543*
Spike productivity (g)	-0.155	0.594**	0.143	0.717**	0.390*	0.771**	0.293	0.664*
Number of grains per spike	-0.126	0.642**	0.267	0.753**	0.351	0.790**	0.237	0.669**
Proportion of grains above 2.5 mm (%)	-0.016	0.199	-0.263	0.349	0.224	0.431	0.331	0.285

\* and \*\* indicate significant relationships at  $p = 0.05$  and  $p = 0.01$ , respectively.

## Discussion

The stand structure is a result of a simultaneous growth of individual plants within the framework of a population and changes in dependence on the dynamics of growth processes, which are limited both spatially and temporally, in the course of the growing season. It is widely known that stand structure, especially proportion of strong tillers in the total number of spikes at harvest can influence yield components (Křen et al. 1992).

The combination of high N dose with favourable weather conditions in 2011 had a positive effect on tiller formation, resulting in higher proportion of strong tillers at the end of tillering and maintaining for longer time high number of intermediate tillers. Tiller differentiation occurred in these stands relatively early, which led to the decrease of total number of tillers per area unit and equalization of the survived ones.

An unfavourable year (2013) was characterized with later sowing and higher temperatures during tillering. These resulted in long-day conditions and shortening of the tillering



period. Del Moral and Del Moral (1995) showed that the maximum number of tillers was inversely related to the temperature during tillering. This means that higher temperatures are shortening the tillering period resulting in a lower number of tillers formed. Peltonen-Sainio et al. (2009) found, that under long-day conditions, the tillers are dominated by main shoot and tiller yield potential remains clearly underutilised even under conditions favouring growth. The combination of both, higher temperatures and long-day conditions during tillering was probably the main driver of lower tillering and delayed differentiation of tillers.

In barley, decreases in total above-ground biomass and grain yield have been observed under elevated temperatures (Clausen et al. 2011). Högy et al. (2013) reported decrease of grain yield, grain number per ear and thousand-grain weight under elevated temperature. Reduction in grain yields can be attributed to temperature induced metabolic changes, to the shorter duration of crop growth and development, and to perturbation of processes related to carbon assimilation (Stone 2001).

The conclusions drawn by Muravjev (1973) were also confirmed that under favourable conditions the uniformity of stems and ears can be increased by competition induced in the period of stem elongation. Such a competition results in the selection of the strongest and the most vigorous tillers.

In the favourable year 2011 a sufficient number of strong tillers were formed since BBCH 25 which enabled: i) indirect increase of the carrying capacity of the environment through greater formation of secondary roots, thus increasing the utilization of water and nutrients, ii) selection of strong tillers from a sufficient number of tillers formed at the beginning of stem elongation providing higher number and productivity of spikes at harvest. Under unfavourable conditions in 2013, smaller numbers of tillers were formed and their differentiation was delayed. This resulted in: i) lower grain yields due to smaller number of spikes per m<sup>2</sup> and lower spike productivity, ii) greater importance of the previously formed strong tillers for yield formation and components of spike productivity, particularly grain number in a spike.

The results also show differences in yield stability between different varieties. While the variety Sebastian (and to a lesser extent also variety Prestige) showed the highest difference in yield between favourable (2011) and unfavourable year (2013), in variety Bojos the yield differences were much smaller, indicating a higher plasticity of this variety to environmental conditions. From the analysis of yield components role in yield formation, it is obvious that the variety with high plasticity should provide a sufficient compensation by increasing the spike productivity, particularly by increasing the number of grains per spike. Similarly Del Moral et al. (2003) showed that yield stability of barley varieties 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. Another important feature of the variety Bojos, that determines its plasticity under the conditions unfavourable for tillering, is the ability to generate a large proportion of strong tillers already during tillering, and this capability is supported by nitrogen nutrition. This ability is then crucial for the achievement of high productivity of spikes as the productive stems and spikes are mostly created from strong tillers and only to



a limited degree from intermediate ones. Tamm (2003) reported a relatively low genetic variability in tillering among the European spring barley varieties, however, information about differences in the relative proportion of individual tiller categories are not available. The critical period for grain number determination is generally between the end of tillering and end of stem elongation (Chmielewski and Köhn 1999), so that a sufficient number of strong tillers at the beginning of this period are crucial for achieving a high number of grains in the spike. However, Arisnabarreta and Miralles (2008) showed that the critical period for grain number determination differs among barley genotypes and is influenced by light conditions during stem elongation.

It can be concluded that under the changing weather conditions, the strategy of the balanced level of yield parameters and growing of varieties with high ability to compensate among yield components is most beneficial, which enables optimisation of inter- and intra-plant competition during vegetation and effective utilisation of the carrying capacity of the environment, the level of which can be changed by fertilisation.

#### Acknowledgement

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### Electronic Supplementary Material (ESM)

Electronic Supplementary Material (ESM) associated with this article can be found at the website of CRC at <http://www.akademai.com/content/120427/>

Electronic Supplementary *Table S1*. Differences of 2011 and 2013 in the number of days between the evaluated growth stages (BBCH) in the sum of active daily temperatures (°C) and in the sum of precipitation (mm)

Electronic Supplementary *Figure S1*. Effect of year, sowing rate (Millions of Germinating Seeds – MGS per hectare) and nitrogen nutrition on total number of tillers and dry weight of above-ground biomass at two growth stages (BBCH 25 and 39). The means (columns) and 95% confidence intervals (error bars) are presented ( $n = 3$ )

Electronic Supplementary *Figure S2*. Effect of year, sowing rate (Million of Germinating Seeds – MGS per hectare) and nitrogen nutrition on grain yield, individual yield components and protein content. The means (columns) and 95% confidence intervals (error bars) are presented ( $n = 3$ )