P L A N T   B R E E D I N G   A N D   S E E D   S C I E N C E
Volume 73 2016

DOI: 10.1515/plass-2016-0006

Dariusz R. Mańkowski [1*)], Janusz Kozdój [2], Monika Janaszek-Mańkowska [3]

[1] Department of Seed Science and Technology, Plant Breeding and Acclimatization Institute – NRI, Radzików, 05-870 Błonie, Poland; [2] Department of Plant Biotechnology and Cytogenetics, Plant Breeding and Acclimatization Institute – NRI, Radzików, 05-870 Błonie, Poland; [3] Department of Engineering, Warsaw University of Life Science; Nowoursynowska 166, 02-787 Warsaw, Poland;
[*)] Corresponding author: e-mail: d.mankowski@ihar.edu.pl

STRUCTURAL EQUATION MODEL AS A TOOL TO ASSESS THE RELATIONSHIP BETWEEN GRAIN YIELD PER PLANT AND YIELD COMPONENTS IN DOUBLED HAPLOID SPRING BARLEY LINES (*HORDEUM VULGARE* L.)

ABSTRACT

The aim of this study was to describe and characterize the relationships between yielding factors and grain yield per doubled haploid (DH) plant of spring barley as well as relation between yield components and duration of each stage of plant development. To describe these relations structure equation modeling was used. The study included plants of doubled haploid spring barley lines (*Hordeum vulgare* L.) derived from two-rowed form of Scarlett cultivar. The SAS® system was used to analyze the model of relationships between grain yield per plant and yield components. Our results indicate that the number of spikes per plant and grain yield per spike had a direct and decisive influence on the grain yield of the investigated DH plants of spring barley. Based on the path model analysis it was found that the most important factor determining grain yield per DH plants of spring barley was the number of spikes per plant and the duration of tillering and shooting stages.

Key words: doubled haploids, *Hordeum vulgare* L., Structural Equation Modeling (SEM), yield related traits

INTRODUCTION

Homozygous doubled haploid lines (DH) are important in plant breeding research for obtaining varieties with improved handling characteristics. DH lines are

---

Communicated by Grzegorz Żurek

crossed with other varieties within the same species in order to broaden the genetic variability of traits that are important for the breeder. One of the most important criteria in assessing DH lines of spring barley is formation of the yielding factors that determine the grain yield. In terms of cereals physiology, formation of the yielding factors is conditioned by the morphological structure of plants (number of shoots in the plant) and the morphological structure of the spike (spike length, number of spikelets, number of kernels). These structural elements of the plant and spike are formed during the successive stages of plant development (Kirby and Appleyard, 1984, Klepper *et al.*, 1998: Ugarte *et al.*, 2007; Sreenivasulu and Schnurbusch, 2012) in varying thermal and precipitation conditions. According to Klepper *et al.* (1998), the number of plants per area unit is determined by the seeding density and is formed from the sowing stage to the beginning of the shooting stage. The number of spike-bearing tillers per plant of spring cereals, including spring barley, is determined by the duration of the tillering stage, tillering rates during this stage, the total number of tillers generated, and the rate of tiller mortality which occurred between the beginning of shooting (jointing) and the early booting (spike development in the flag leaf sheath) stages (Klepper *et al.*, 1998). The number of kernels per spike is determined by the rate of spikelet formation per spike, the final number of spikelets, and the duration of the spikelet stage, as well as the duration of floret development from the floret primordium stage to the anthesis stage (Klepper *et al.*, 1998). A single kernel weight depends on structural and physiological-biochemical processes occurring both before and shortly after the anthesis stage, as well as the duration and rate of kernel maturation (filling stage). The number of kernels per plant at harvest is determined by the number of spikes per plant, the number of fertile spikelets per spike and the number of kernels per spikelet (Sreenivasulu and Schnurbusch, 2012). The processes of initiation, growth and development of the spike and its components - spikelets, florets and kernels of cereal plants are well documented (Williams, 1975; Kirby and Appleyard, 1984; Naylor and Munro, 1993; Nátrová and Jokes, 1993).

Growth and development of crops, including spring barley, is a non-linear multifactor process (Kahazaei *et al.*, 2008). Grain yield per plant is a quantitative feature that, for utilitarian reasons, determines the most important final effect of complex morpho-physiological processes that occur during its growth and development. Yield depends on the interaction of direct factors (genetic factors, physiological and biochemical factors, structural factors) and indirect factors (habitat factors, cultivation factors, etc.). These two groups of factors are equivalent to quantitative features, which play a significant role in obtaining the grain yield per plant at a given level (Kozak, 2004; Gozdowski *et al.*, 2007). Statistical analysis of yielding factors allows to understand biological mechanisms that determine the plant yield, which may be important for breeding and cultivation (Fraser and Eaton, 1983).

Several statistical methods are used to describe the relationships between yielding traits and final functional yield, such as grain yield in cereals (Kozak and Azevedo, 2010). Most frequently used methods are linear multiple regression models, in particular, the modification of this method known as a simple path analysis – PA (Wright, 1921; Kenny, 1979). Another method, derived from multiple regression, is sequential yield component analysis – SYCA (Eaton and Kyte, 1978, Eaton and McPherson, 1978), which, in contrast to the multiple regression, takes into account the ontogenetic approach (Kozak, 2007). There is also a method that combines SYCA with variance analysis and it is called two-dimensional partitioning of yield variation – TDP (Eaton 1986; Gołaszewski, 1996; Gołaszewski *et al.*, 1998, Kozak, 2006). The correlation between crop yield and yielding factors can also be analyzed using structural equation models – SEM (Kenny, 1979; Timm, 2002, Kozak *et al.*, 2007), which are regarded as an important statistical tool designed to study and describe cause and effect relationships that characterize different, complex phenomena and biological processes (Kozak and Kang, 2006; Kozak and Azevedo, 2010).

The aim of this study was to describe and characterize the relationships between yielding traits and grain yield per DH plant of spring barley as well as relation between yielding traits and duration of each stage of plant development. To describe these relations SEM analysis was used.

MATERIALS AND METHODS

Field experiment

The study included plants of doubled haploid spring barley lines (*Hordeum vulgare* L.) derived from two-rowed form of Scarlett cultivar. The examined DH lines were derived from androgenic embryos, obtained from an *in vitro* culture of isolated microspores (Kozdój *et al.*, 2010). The experiment duration comprised three years (2004, 2007 and 2008) to reflect the variability of weather conditions.

Plants were grown in the experimental field of Plant Breeding and Acclimatization Institute – National Research Institute in Radzików, near Warsaw in Poland (52°12’43”N; 20°38’12”E). The experiment was carried out in randomized complete block design. In each year of study, at the turn of March and April, the seeds were sown on plots of an area of 1 m^2, 100 seeds per plot. Recommended mineral fertilization was used before sowing, and a chemical weed control was performed on every plot during the tillering stage.

Throughout the growing season (from sowing to harvest), calendar exact dates of entry in the subsequent stages of plant development were recorded. Stages of plant development were determined on the basis of its morphological structure, i.e. the number of leaves in the seedling, the number of tillers per plant, the number of nodes on the main shoot, the emergence of spike above the flag leaf and entering the anthesis stage. The maturation stages of kernels were distinguished on the basis of their consistency, colour and hardness, according to the Zadoks decimal scale (Zadoks *et al.*, 1974). To demonstrate the relationships between the duration of stages of plant development, grain yield and yield components, the duration of the tillering stage (DC 21-29) and shooting stage (DC 30-49) was calculated as well as the duration of spike maturation stage, which included the subsequent stages of spike emergence (DC 51-59), anthesis (DC 61-69), milk developement (DC 71-79), dough development (DC 83-89) and full maturity (DC 91-92). These data are presented in Table 1.

Table 1

Duration of stages of plant development of DH spring barley in vegetation season and characterization of conditional weather in years 2004, 2007 and 2008 (Kozdój *et al.*, 2010)

Stage	Year	Period	Duration [days]	Daily mean temperature [°C]	Sum of temperatures [°C]	Sum of rainfall [mm]
Sowing – emergence (DC 0–1)	2004	IV.14–IV.24	10	10.6	117.0	17.0
	2007	III.29–IV.15	17	8.8	144.8	13.2
	2008	IV.02–IV.17	15	8.5	136.5	16.2
Seedling (DC 11–19)	2004	IV.25–V.4	9	12.0	119.7	15.0
	2007	IV.16–IV.24	8	9.5	85.6	5.4
	2008	IV.18–IV.27	9	9.8	98.4	13.4
Tillering (DC 21–29)	2004	V.05–V.30	25	11.4	295.5	43.2
	2007	IV.25–V.14	19	12.6	251.1	33.4
	2008	IV.28–V.11	13	13.2	184.9	29.4
Shooting (DC 30–49)	2004	V.31–VI.14	14	16.1	241.3	17.2
	2007	V.15–V.28	13	18.4	257.9	30.2
	2008	V.12–VI.03	22	15.1	348.3	27.8
Spike emergence and anthesis	2004	VI.15–VI.24	9	15.6	156.3	22.6
	2007	V.29–VI.07	9	18.7	186.9	19.6
	2008	VI.04–VI.09	5	19.6	117.5	0.0
Milk development (DC 71–79)	2004	VI.25–VII.04	9	15.7	157.3	16.6
	2007	VI.8–VI.28	20	19.3	405.3	53.6
	2008	VI.10–VII.02	22	18.6	428.5	15.0
Dough development (DC 83–89)	2004	VII.05–VII.20	15	17.4	278.9	49.8
	2007	VI.29–VII.03	4	18.3	91.6	14.6
	2008	VII.03–VII.10	7	18.9	151.5	12.6
Maturity (DC 91–92)	2004	VII.21–VII.27	6	19.9	139.4	19.6
	2007	VII.04–VII.07	3	16.1	96.5	26.0
	2008	VII.11–VII.17	6	19.4	136.0	31.5
Total vegetation season	2004	IV.14–VII.27	104	14.5	1505.4	201.0
	2007	III.29–VII.09	102	14.9	1519.6	196.0
	2008	IV.02–VII.17	106	15.1	1601.6	145.9

In the full maturity stage 15 plants were randomly sampled from each plot. The length of stem (longest shoot), the number of spikes per plant, the length of spikes per plant, the total number of sterile and fertile spikelets per spike was determined for each of these plants. The number of fertile spikelets per spike was equal to the number of kernels per spike. Grain yield per plant was equal to the mass of kernels contained in all spikes of this plant. The mean weight of kernels per spike was then equal to the ratio of kernels weight per plant and number of spikes per plant. The mass of a single kernel was defined as the ratio of the mass of kernels from the plant to the number of kernels per plant. A total number of 79 lines was tested in the experiment lasting three years. Weather during the growing season was described based on the daily mean temperature, sum of temperatures and sum of rainfall for each phenophase, from sowing to harvest (Table 1). In 2004, the meteorological data developed for the town of Błonie (7 km from Radzików) were obtained from the Institute of Meteorology and Water Management, and, in the years 2007 and 2008, data were obtained from a meteorological station of PBAI-NRI in Radzików.

Statistical analysis

Table 2

Means values and coefficients of variance (CV) of analyzed exogenous and endogenous variables

Variable description	Mean	CV
Exogenous variables		
Duration of tillering and shooting (DC 21–49)	38.5 days	7.25%
Duration of spike maturation (DC 51–92)	43.1 days	1.59%
Endogenous variables		
Number of spikes per plant	7.7	46.22%
Length of the longest shoot	68.3 cm	19.11%
Length of spike	8.2 cm	14.32%
Number of sterile spikelets per spike	3.9	54.03%
Number of fertile spikelets per spike (grain number per spike)	21.4	16.28%
Grain yield per spike	1.1 g	26.17%
Grain yield per plant	8.9 g	56.58%

The correlation between grain yield per plant and the yielding traits was examined using Pearson correlation coefficients analysis. To analyze the model of relationships between grain yield per plant and yield components structural equation models were performed using the SAS® System version 9.2 (Hatcher, 1994; Yung, 2008; SAS Institute Inc., 2009). Data set containing analyzed traits was divided into two subsets (Table 2) in accordance with the SEM methodology (Bollen, 1989; Hatcher, 1994; Timm, 2002, Armitage and Colton, 2005). The first subset consisted of causative variables (exogenous variables), i.e. variables which do not affect any of the other analyzed traits. The second subset consisted of effect variables (endogenous variables), i.e. variables which affect other analyzed traits. All analyzed traits were considered as manifest variables and the calculations were performed for deviations of the analyzed traits from their mean values, in order to eliminate intercepts from structural models. Searching for the proper cause and effect model was based on the methodology proposed by Shipley (2002) and Mańkowski (2013). Several alternative relationship models were analyzed. Every model was analyzed for the data obtained in each year of experiment, then the model that best described the observed relationships was selected. The appropriate model was chosen on the basis of fit statistics. For this model, an analysis of the data took from all three years of experiment was carry out.

RESULTS

The duration of subsequent stages of growth and development of spring barley varied between years and depended on the changes of thermal and precipitation conditions (Table 1). Growth and development of plant proceeded in weather conditions, which can be considered quite dry (2004 and 2007) and dry (2008).

Tillering and shooting stages (DC 21-49) as well as spike maturation stage (DC 51-92) had a similar duration in the years of study and commonly lasted 38.5 and 43.1 days, respectively (Table 2). The highest variability was observed for grain yield per plant (56.6%), the number of sterile spikelets per spike (54.0%) and the number of spikes per plant (46.2%). The lowest variability was observed for the length of spike (14.3%) and the number of fertile spikelets per spike (16.3%).

Table 3

Pearson linear correlation coefficients for analyzed variables

Variables	1	2	3	4	5	6	7	8	9
1 — Duration of tillering and shooting	1.00								
2 — Duration of spike maturation	0.22 **	1.00							
3 — Number of spikes per plant	0.69 **	0.25 **	1.00						
4 — Length of stem	0.50 **	0.52 **	0.32 **	1.00					
5 — Length of spike	0.52 **	0.19 **	0.47 **	0.62 **	1.00				
6 — Number of sterile spikelets per spike	–0.48 **	–0.43 **	–0.30 **	–0.54 **	–0.31 **	1.00			
7 — Number of fertile spikelets per spike 1	0.55 **	0.42 **	0.41 **	0.71 **	0.77 **	–0.76 **	1.00		
8 — Grain yield per spike	0.67 **	0.13 *	0.41 **	0.66 **	0.79 **	–0.58 **	0.86 **	1.00	
9 — Grain yield per plant	0.76 **	0.18 **	0.93 **	0.46 **	0.65 **	–0.44 **	0.62 **	0.70 **	1.00

The analysis of Pearson correlation coefficients (Table 3) revealed the presence of strong correlation among all analyzed traits, which indicated a complex relationships occurring within the analyzed data set.

In the light of observed interdependencies and merits of physiology, growth, development and yielding of cereal plants (Klepper *et al.* 1998; Hay and Porter, 2006), a set of seven models model describing relationships between the analyzed traits were developed.

The postulated models were analyzed according to the SEM methodology. During those analyzes the necessary adjustments were introduced. These adjustments resulted from the statistics for the individual relationships. Model that best fit to the empirical data obtained in all three years of experiment was selected on the basis of its fit statistics (Table 4).

Table 4

Fit summary for analyzed SEM model of DH spring barley

Goodness-of-fit statistics	Years			
	2004	2007	2008	All three years (2004,
Fit function	0.5415	0.7037	0.4201	0.7543
χ^2 statistic	88.805	41.517	37.388	236.883
Degrees of freedom	19	19	19	19
P-value	<0.0001	<0.0001	<0.0001	<0.0001
Root Mean Square Residual (RMSR)	0.8824	0.3731	0.9140	0.8103
Standardized Root Mean Square Residual (SRMSR)	0.0793	0.0118	0.0613	0.0781
Goodness of Fit Index (GFI)	0.9933	0.9739	0.9929	0.9731
Adjusted Goodness of Fit Index (AGFI)	0.9673	0.9515	0.9729	0.9695
Bentler-Bonett Normed Fit Index (NFI)	0.9550	0.9621	0.9891	0.9648
Bentler-Bonett Non-Normed Fit Index (NNFI)	0.9505	0.9541	0.9701	0.9574

The postulated model was stored as a path diagram (Fig. 1). Exogenous variables in the postulated model constituted the duration of tillering and shooting stages (DC 21-49) and the duration of spike maturation (DC 51–92). The model also included the relationships between exogenous variables (mutual relations between the duration of development stages mentioned above). These relationships were expressed in the path model as covariances of exogenous variables, marked in the path diagram with a recursive rounded arrow and a symbol f_1 (Fig. 1).

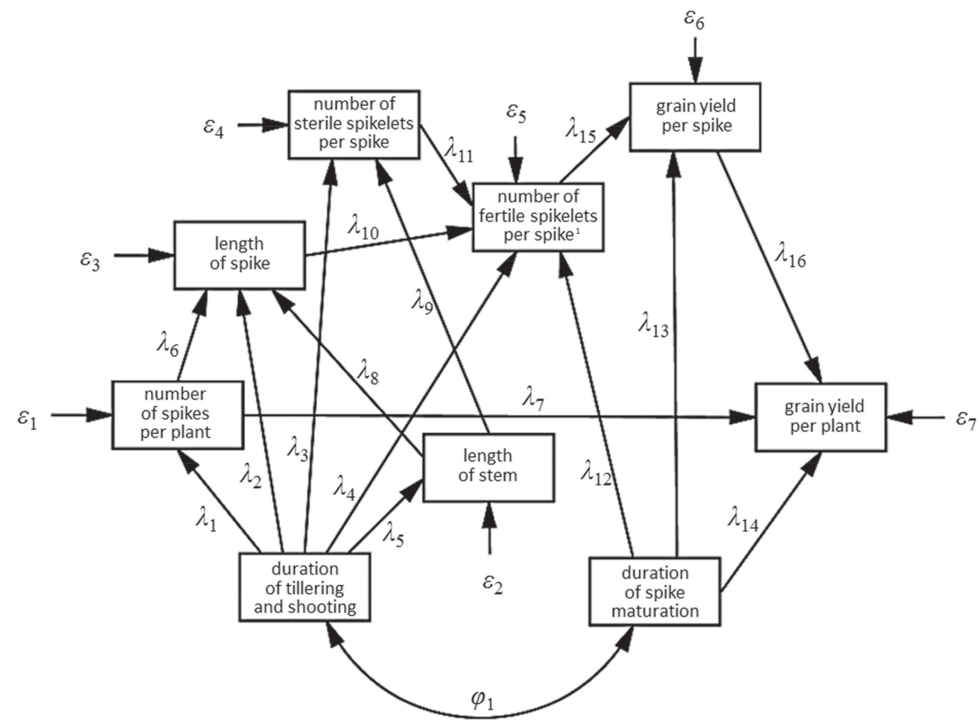

Fig. 1. Path diagram describing the relationships between DH spring barley grain yield and yield components. Abbreviations: $\varepsilon_1, \varepsilon_2, \ldots, \varepsilon_7$ – errors (residuals) of endogenous variables; $\lambda_1, \lambda_2, \ldots, \lambda_{16}$ – path coefficients; φ_1 – covariance between exogenous variables; [1] – equivalent to grain number per spike

The next level of the model was a group of endogenous variables, conditioned, *inter alia*, by the exogenous variables and defined in the literature as yield components, i.e. number of spikes per plant, length of stem, mean length of spike, average number of sterile spikelets per spike, average number of fertile spikelets per spike (grain number per spike) and grain yield per spike. The last endogenous variable was grain yield per plant. The model also took into account the impact of uncontrollable factors on the formation of endogenous variables. This effect was included on the diagram as residuals, marked from e_1 to e_7 (Fig. 1). The relationships between endogenous and exogenous variables were marked in the diagram with non-recursive arrows and symbols from λ_1 to λ_{16} (Fig. 1).

The form of the model described above was implemented in the SAS® System using the CALIS procedure and subjected to statistical analysis. An estimation of SEM parameters is an iterative process, requiring specification of some initial values for the estimated parameters, which was performed using the McDonald method (McDonald and Hartmann, 1992). Then the Levenberg-Marquardt method was used to optimize the parameters of the model (Levenberg, 1944; Marquardt, 1963, Madsen *et al.*, 2004). The convergence criterion was met after 11 iterations. Significant χ^2 statistics (Table 4) resulting from the analysis indicated that the postulated model has not been satisfactorily confirmed by empirical data. Nevertheless, the fit statistics for this model were very high (Table 4) and proved that the postulated model adequately reflected the relationships occurring in the data set. A similar problem occurs quite often in the SEM analysis performed on data derived from natural experiments (Vargas *et al.*, 2007), where the random effect is quite significant. This may be caused by a tendency of χ^2 statistics to give a false significant test result if the postulated model is not an accurate representation of all the empirical relationships occurring in the analyzed process or phenomenon (MacCallum *et al.*, 1996).

Table 5

Path coefficients, variances and covariances for SEM analysis of DH spring barley

Parameter	Estimate	Standard error	Test statistic	Standardized estimate
		Path coefficients		
λ_1	0.874	0.052	16.740**	0.687
λ_2	0.055	0.026	2.141*	0.130
λ_3	–0.208	0.039	–5.332**	–0.279
λ_4	–0.074	0.029	–2.549*	–0.059
λ_5	2.330	0.229	10.188**	0.498
λ_6	0.073	0.019	3.897**	0.219
λ_7	1.119	0.012	96.379**	0.778
λ_8	0.044	0.004	10.300**	0.485
λ_9	–0.064	0.008	–7.670**	–0.402
λ_{10}	1.811	0.065	27.834**	0.602
λ_{11}	–0.953	0.036	–26.582**	–0.559
λ_{12}	0.334	0.096	3.479**	0.065
λ_{13}	–0.114	0.011	–10.439**	–0.252
λ_{14}	–0.482	0.056	–8.558**	–0.065
λ_{15}	0.080	0.002	37.982**	0.918
λ_{16}	6.584	0.132	49.773**	0.398
		Variances		
ε_1	6.658	0.531	12.530**	
ε_2	127.664	10.187	12.530**	
ε_3	0.726	0.058	12.530**	
ε_4	2.789	0.223	12.530**	
ε_5	1.286	0.103	12.530**	
ε_6	0.017	0.001	12.530**	
ε_7	0.444	0.035	12.530**	
		Covariances		
φ_1	0.426	0.110	3.869**	

** – significant at P=0.01; * – significant at P=0.05

Table 6

Direct, indirect and total effects for SEM analysis of DH spring barley

Effect			Direct effect		Indirect effect		Total effect	
			Estimate	Standardized estimate	Estimate	Standardized estimate	Estimate	Standardized estimate
Number of spikes per plant (R^2=0.472)								
X_1	→	Y_1	0.874	0.687	—	—	0.874	0.687
Length of stem (R^2=0.248)								
X_1	→	Y_2	2.330	0.498	—	—	2.330	0.498
Length of spike (R^2=0.476)								
X_1	→	Y_3	0.055	0.130	0.166	0.392	0.221	0.523
Y_1	→	Y_3	0.073	0.219	—	—	0.073	0.219
Y_2	→	Y_3	0.044	0.485	—	—	0.044	0.485
Number of sterile spikelets per spike (R^2=0.352)								
X_1	→	Y_4	–0.208	–0.279	–0.149	–0.200	–0.357	–0.480
Y_2	→	Y_4	–0.064	–0.402	—	—	–0.064	–0.402
Number of fertile spikelets per spike (grain number per spike) (R^2=0.897)								
X_1	→	Y_5	–0.074	–0.059	0.740	0.583	0.666	0.524
X_2	→	Y_5	0.334	0.065	—	—	0.334	0.065
Y_1	→	Y_5	—	—	0.132	0.132	0.132	0.132
Y_2	→	Y_5	—	—	0.140	0.517	0.140	0.517
Y_3	→	Y_5	1.811	0.602	—	—	1.811	0.602
Y_4	→	Y_5	–0.953	–0.559	—	—	–0.953	–0.559
Grain yield per spike (R^2=0.823)								
X_1	→	Y_6	—	—	0.053	0.482	0.053	0.482
X_2	→	Y_6	–0.114	–0.252	0.027	0.059	–0.087	–0.141
Y_1	→	Y_6	—	—	0.011	0.121	0.011	0.121
Y_2	→	Y_6	—	—	0.011	0.475	0.011	0.475
Y_3	→	Y_6	—	—	0.145	0.553	0.145	0.553
Y_4	→	Y_6	—	—	–0.076	–0.513	–0.076	–0.513
Y_5	→	Y_6	0.080	0.918	—	—	0.080	0.918
Grain yield per plant (R^2=0.983)								
X_1	→	Y_7	—	—	1.330	0.726	1.330	0.726
X_2	→	Y_7	–0.482	–0.065	–0.575	–0.077	–1.057	–0.141
Y_1	→	Y_7	1.119	0.778	0.069	0.048	1.188	0.826
Y_2	→	Y_7	—	—	0.074	0.189	0.074	0.189
Y_3	→	Y_7	—	—	0.956	0.220	0.956	0.220
Y_4	→	Y_7	—	—	–0.503	–0.204	–0.503	–0.204
Y_5	→	Y_7	—	—	0.528	0.366	0.528	0.366
Y_6	→	Y_7	6.584	0.398	—	—	6.584	0.398

Abbreviations: X_1 – duration of tillering and shooting; X_2 – duration of spike maturation; Y_1 – number of spikes per plant; Y_2 – length of stem; Y_3 – length of spike; Y_4 – number of sterile spikelets per spike; Y_5 – number of fertile spikelets per spike (grain number per spike); Y_6 – grain yield per spike; Y_7 – grain yield per plant

The values of the model fit statistics proved a good fit to the relationships observed in a set of empirical data. This was confirmed by the low value of root mean square residual (RMSR) and standardized root mean square residual (SRMSR). The SRMSR was lower than 0.08, which shall be considered as the limit for this parameter (Hu and Bentler, 1998, 1999). The high values of fit statistics, such as GFI, AGFI, NFI and NNFI were also obtained Given the importance and interpretation of GFI and AGFI statistics, they can be compared to the coefficient of determination and the adjusted coefficient of determination in multiple regression model (Mulaik *et al.*, 1989). Obtained value of fit statistics (Table 4) were higher than the 0.95, which Hu and Bentler (1998, 1999) reported as the limit for a well-fitting structural models to the empirical data.

The analysis showed that all the path coefficients (Table 5) as well as the estimates of the random errors variation for endogenous variables and the covariance between exogenous variables proved to be statistically significant. The assessment of the coefficients of determination for the endogenous variables (Table 6) shows that the most accurate representation of empirical data in the postulated model was obtained for grain yield per plant (R^2 = 0.98), the number of fertile spikelets per spike (R^2 = 0.90) and for the yield of kernels per spike (R^2 = 0.82). The lowest coefficients of determination were obtained for the length of stem (R^2 = 0.25) and the number of sterile spikelets per spike (R^2 = 0.35).

The high coefficients of determination obtained for the endogenous variables indicate that their variability is largely explained by the analyzed model. Low coefficients of determination for the endogenous variables indicate that they were formed by other factors not tested in this experiment, or relationships not included in the postulated model.

The analysis of direct and indirect effects (Table 6) enabled a detailed characterization of the relationships described by the postulated model. The number of spikes per plant as well as the length of stem depended directly proportional on the duration of tillering and shooting stages (direct effects). The length of spike depended directly proportional on the duration of tillering and shooting stages (both direct and an indirect effects), the number of spikes per plant and length of stem (direct effects). The number of fertile spikelets per spike depended directly proportional on: the duration of tillering and stem elongation stages (proportional indirect effect and inversely proportional direct effect), the number of spikes per plant and the length of stem (indirect effects), spike length and the duration of spike maturation stage (direct effects). The same trait depended inversely proportional on the number of sterile spikelets per spike (direct effect). The number of sterile spikelets per spike depended significantly and inversely proportional on the duration of tillering and shooting stages (both direct and indirect effect) and on the length of stem (direct effect). The grain yield per spike depended significantly and directly proportional on the duration of tillering and shooting stages, the number of spikes per plant, the length of stem, the length of spike (indirect effects), the number of fertile spikelets per spike (direct effect) and inversely proportional on the duration of spike maturation (inversely proportional direct effect, directly proportional indirect effect) and the number of sterile spikelets per spike (indirect effect). The grain yield per plant depended directly proportional on the duration of tillering and shooting stages, the length of stem, the length of spike, the number of sterile spikelets per spike (indirect effect), the number of grains per spike (direct effect), the number of spikes per plant (both direct and indirect effect) and inversely proportional on the duration of the spike maturation stage (directly proportional direct effect and inversely proportional indirect effect) and the number of sterile spikelets per spike (indirect effect).

DISCUSSION AND CONCLUSIONS

Within the available publications which concern the analysis of yield structure and plant development of spring barley, only few lay great emphasis on the contribution of spike development in the formation of grain yield per plant. There are also few works that relate to spring barley DH plants. Nevertheless, the available literature pertaining to the analyzed problem, contains results which confirm the observed and described relationships.

Peltonen-Sainio *et al.* (2009) identified significant and strong correlation between the height of two-rowed forms of spring barley and harvest index ($r = -0.80$). They also identified a significant but moderately strong correlation between the grain filling period and grain yield ($r = 0.58$), the number of tillers per plant ($r = 0.75$) and the number of spike-bearing tillers ($r = 0.64$). The number of tillers per plant and the number of spike-bearing tillers were also significantly correlated with the duration of the tillering stage ($r = 0.59$ and $r = 0.68$, respectively). Del Moral and del Moral (1995) indicated a nonlinear relationship between the increased temperature during the tillering stage and the number of tillers per square meter of field and a very strong significant linear relationship between the number of spikes per area unit and grain yield. Naylor and Munro (1993) indicated that grain yield depends significantly on the height of plants ($r = 0.79$), whereas the number of kernels per spike during seed setting stage is strictly correlated with the number of kernels per spike during harvest time ($r = 0.74$). Based on the analysis carried out for two cultivars of spring barley, Gozdowski *et al.* (2007) found that the number of kernels per spike depended primarily on the length of the spike, and, to a lesser extent, on the height of stem (cumulative $R^2 = 0.67$), while the weight of kernels per spike depended most on the number of kernels per spike, mean kernel weight and length of the spike, and, to a lesser extent, on the height of stem (cumulative $R^2 = 0.97$).

Postulated SEM model comprised relationships between analyzed traits supported by the knowledge of growth physiology, development and yielding of cereal crops. In addition, it allowed to evaluate the internal recursive relationships (covariances) between the analyzed exogenous variables. This approach had already been successfully applied to this type of analysis in different species of crops like pearl millet, sorghum grain, winter wheat, wild oat, lowland rice and grass pea (Maman *et al.*, 2004; Guillen-Portal *et al.*, 2006, Kozak *et al.*, 2007, 2008).

Our results indicate that the number of spikes per plant and grain yield per spike had a direct and decisive influence on the grain yield of the investigated DH plants of spring barley. Other examined yielding traits affected the yield indirectly and their impact, except for the duration of tillering and shooting stages, was low. Yield of grains per spike resulted from the direct effect of the number of fertile spikelets per spike, and thus, the number of grains per spike, as well as partly direct and partly indirect influence of the duration of spike maturation, from early booting to maturity stages. Other traits like length of spike, duration of tillering and shooting stages, length of stem and number of spikes per plant also had significant but indirect impact on yield of grains per spike. The number of fertile spikelets per spike depended on the length of spike, average length of stem, duration of tillering and shooting stages (proportional effect) and on the number of sterile spikelets (inversely proportional effect). The duration of stages of plant development significantly influenced the formation of the analyzed structural traits of spike and yield components. It was also concluded that there is a significant covariance between the duration of subsequent stages of plant development. A positive value of the estimated covariance leads to the conclusion that there is a linear and directly proportional relationship between these stages.

Based on the analysis of the path model shown in Figure 1, it was found that the most important factor determining grain yield per DH plants of spring barley was the number of spikes per plant and the duration of tillering and shooting stages.

ACKNOWLEDGEMENTS

We thank Mrs. S. Oleszczuk of the Department of Plant Biotechnology and Cytogenetics of Plant Breeding and acclimatization Institute - National Research Institute for sharing barley DH lines for research.

REFERENCES

Armitage, P., Colton, T. (eds.), 2005. Encyclopedia of Biostatistics, 2nd edition. John Wiley & Sons Inc., Hoboken, USA.

Bentler, P.M., Yuan, K.-H., 1999. Structural equation modeling with small samples: Test statistics. Multivariate Behavioral Research, 34(2), 181–197.

Bollen, K.A., 1989. Structural Equations with Latent Variables. Wiley & Sons Inc., NY, USA.

Eaton, G.W., 1986. Two-dimensional partitioning of yield variation. Hort. Sci., 21(4): 1052–1053.

Eaton, G.W., Kyte, T.R., 1978. Yield component analysis in strawberry. Journal of the American Society for Horticultural Science 103: 578–583.

Eaton, G.W., McPherson, E.A., 1978. Morphological component of yield in cranberry. Horticultural Research 17: 73–82.

Freaser, J., Eaton, G.W., 1983. Applications of yield component analysis to crop research. Field Crop Abstracts 36, 787–796.

del Moral, G.M.B., del Moral, G.L.F., 1995. Tiller production and survival in relation to grain yield in winter and spring barley. Fields Crops Research, 44: 85–93.

Gołaszewski, J., 1996. A method of yield component analysis. Biometrical Letters, 33(2): 79–88.

Gołaszewski, J., Idźkowska, M., Milewska, J., 1998. The TDP method of seed yield component analysis in grain legume breeding. J. Appl. Genet., 39(4): 299–308.

Gozdowski, D., Kozak, M., Kang, M.S., Wyszyński, Z., 2007. Dependence of grain weight of spring barley genotypes on trials of individual stems. Journal of Crop Improvement 20(1/2), 223–233.

Guillen-Portal, F.R., Stougaard, R.N., Xue, Q., Eskridge, K.M., 2006. Compensatory mechanisms associated with the effect of spring wheat seed size on wild oat competition. Crop Science, 46, 935–945.

Hatcher, H., 1994. A Step-by-Step Approach to Using SAS® for Factor Analysis and Structural Equation Modeling. Carry, NC: SAS Institute Inc.

Hay, R., Porter, J., 2006. The physiology of crop yield. Balckwell Publishing, Oxford, UK.

Hu L.-T., Bentler P.M. 1998. Fit indices in covariance structure modeling: Sensitivity to underparameterized model misspecification. Psychological Methods, 3(4), 424—453.

Hu L.-T., Bentler P.M. 1999. Cutoff criteria for fit indices in covariance structure analysis: Conventional criteria versus new alternatives. Structural Equation Modeling, 6(1), 1—55.

Kenny, D.A., 1979. Correlation and causality. John Wiley & Sons Inc., Hoboken, USA.

Khazaei, J., Naghavi, M.R., Jahansouz, M.R., Salimi-Khorshidi, G., 2008. Yield estimation and clustering of chickpea genotypes using soft computing techniques. Agronomy Journal 100, 1077–1087.

Kirby, E.J.M., Appleyard, M., 1984. Cereal plant development and its relation to crop management. In: Cereal production. Proceedings of the Second International Summer School in Agriculture held by the Rogal Dublin Society in cooperation with WK Kellog Foundation. Gallagher E. J. (Ed.) Dublin. 161–173.

Klepper, B., Rickman, R.W., Waldman, S., Chevalier, P., 1998. The physiological life cycle of wheat: Its use in breeding and crop management. Euphytica 100, 341–347.

Kozak, M., 2004. New concept of yield component analysis. Biometrical Letters 41(2), 59–69.

Kozak, M., 2006. Two-dimensional partitioning of yield variation: A critical note. Plant Breeding and Seed Science, 53: 37–42.

Kozak, M., 2007. Ontogenetic chain and Markov condition in crop science. Nature and Science, 3(5): 5–8.

Kozak, M., Azevedo, R.A., 2010. Does using stepwise variable selection to build sequential path analysis models make sense? Physiologia Plantarum, DOI: 10.1111/j.1399-3054.2010.014341.x.

Kozak, M., Bocianowski, J., Rybiński, W., 2008. Selection of promising genotypes based on path and cluster analyses. Journal of Agricultural Science, 146: 85–92.

Kozak, M., Kang, M.S., 2006. Note on modern path analysis in application to crop science. Communications in Biometry and Crop Science Vol. 1, No. 1: 32–34.

Kozak, M., Singh, P.K., Verma, R.M., Hore, D.K., 2007. Causal mechanism for determination of grain yield and milling quality of lowland rice. Field Crops Research 102, 178–184.

Kozdój, J., Mańkowski, D.R., Oleszczuk, S., 2010. Analysis of the potential for yield formation in doubled haploids of spring barley (*Hordeum vulgare* L.) obtained via androgenesis (*in Polish*). Biull. PBAI, 256: 97–116.

Levenberg, K., 1944. A method for the solution of certain non-linear problems in least squares. The Quarterly of Applied Mathematics, 2, 164–168.

MacCallum, R.C., Browne, M.W., Sugawara, H.M., 1996. Power analysis and determination of sample size for covariance structure modeling. Multivariate Behavioral Research, 1(2), 130–149.

Madsen, K., Nielsen, H.B., Tingleff, O., 2004. Methods for non-linear least squares problems. 2nd edition. Informatics and Mathematical Modeling, Technical University of Denmark.

Maman, N., Mason, S.C., Lyon, D.J., Dhungana, P., 2004. Yield components of pearl millet and grain sorgum across environments in the central great plains. Crop Science, 44, 2138–2145.

Mańkowski D.R., 2013. Structural equation models SEM in agricultural research. PBAI-NRI Monographs and Dissertations No. 42, Radzików, Poland. (*in polish*)

Marquardt, D., 1963. An algorithm for least-squares estimation of nonlinear parameters. SIAM Journal of Applied Mathematics, 11, 431–441.

McDonald, R.P., Hartmann, W., 1992. A procedure for obtaining initial values of parameters in the RAM model. Multivariate Behavioral Research, 27, 57–176.

Mulaik S.A., van Alstine J., James L.R., Bennett N., Lind S., Stilwell C.D. 1989. An evaluation of goodness-of-fit indices for structural equation models. Psychological Bulletin, 105, 430—445.

Nátrová, Z., Jokeš, M., 1993. A proposal for a decimal scale of the inflorescence development of wheat. Rostl. Vyr. 4: 315–328.
Naylor, R.E.L., Munro, L.M. 1993. Apical development in barley and wheat varieties. Aspects of Applied Biology. 34: 105–110.
Peltonen-Sainio,P., Jauhiainen L., Rajala, A., Muurinen, S., 2009. Tiller traits of spring cereals under tiller-depressing long day conditions. Field Crops Research, 113: 82–89.
SAS Institute Inc., 2009. SAS/STAT 9.2 User's Guide, Second Edition. SAS Publishing, SAS Institute Inc., Cary, NC, USA.
Shipley B. 2002. Cause and correlation in biology. A user's guide to path analysis, structural equations and causal inference. Cambridge University Press, Cambridge.
Sreenivasulu, N., Schnurbusch, T., 2012. A genetic playground for enhancing grain number in cereals. Trends in Plant Science. 17, 2: 91–101.
Timm, N.H., 2002. Applied multivariate analysis. Springer-Verlag Inc., New York, USA.
Ugarte, C., Calderini, D.F., Slafer, G.A., 2007. Grain weight and grain number responsiveness to pre-anthesis temperature in wheat, barley and triticale. Field Crops Res. 100: 240–248
Vargas, M., Crossa, J., Reynolds, M.P., Ghungana, P., Eskridge, K.M., 2007. Structural equation modeling for studying genotype × environment interactions for physiological traits affecting yield in wheat. Journal of Agricultural Science 145, 151–161.
Williams, R.F., 1975. The growth o fan inflorescence. In: The shoot apex and Lear growth. A study in quantitative biology. Cambridge Univ. Press: 183–198.
Wright, S., 1921. Correlation and causation. Journal of Agricultural Research 20, 557–585.
Yung, Y.-F., 2008. Structural Equation Modeling and Path Analysis Using PROC TCALIS in SAS® 9.2. SAS Global Forum 2008 Proceedings; paper 384-2008.
Zadoks, J.C., Chang, T.T., Konzak, G.F., 1974. A decimal code for growth stages of cereals. Weed Research 14, 415–421.