Field Crops Research 174 (2015) 40–47

Contents lists available at ScienceDirect

Field Crops Research

journal homepage: www.elsevier.com/locate/fcr

Evaluation of variability, heritability and environmental stability of seed quality and yield parameters of *L. angustifolius*

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ARTICLE INFO

Article history:
Received 7 May 2014
Received in revised form
11 December 2014
Accepted 15 December 2014
Available online 4 February 2015

Keywords:
Seed quality
Yield
Narrow-leafed lupin
Heritability
Variability
Environmental stability

ABSTRACT

Lupin seeds are gaining attention as highly palatable and healthy ingredients for human food in the last decades. But, lupin cultivars available today in some respect do not meet the requirement for a broad application in the food industry. Important seed quality traits, e.g. protein and alkaloid content, vary considerably depending on environmental conditions. In this study, advanced German breeding lines of narrow-leafed lupins (*Lupinus angustifolius* L.) were evaluated in 12 environments all over Germany to determine environmental stability and heritability of important agronomic and nutrient traits and to identify environmentally stable genotypes for these traits. Heritability was high for all analysed traits ($h^2 > 50\%$), especially for seed alkaloids ($h^2 = 95.2\%$) and thousand seed weight ($h^2 = 94.5\%$). Out of the genotypes tested one breeding line were identified, which combined constantly low alkaloid content with a high and stable seed protein content and yield. However, results of range and genotypic- and phenotypic coefficient of variation revealed a low genetic diversity within the tested breeding material for most of the traits, which limits the potential for improvement of lupins without broadening the gene pool in German lupin breeding.

1. Introduction

Growing and breeding of narrow-leafed lupins (*Lupinus angustifolius* L.) has a very short history in Germany. Being introduced to Northern Germany in the 18th century from their Mediterranean origin, narrow-leafed lupins were predominantly used as green manure and forage crop. Seeds could be used at that time only to a limited extend because of their high alkaloid content. Systematic breeding of narrow-leafed lupins, as well as of yellow and white lupins (*L. luteus* and *L. albus*), was initiated at the beginning of the 20th century by selection of mutants with reduced alkaloid levels (Sengbusch, 1931). With this achievement lupins became fully accessible as feed and food crop for the first time. Breeding of narrow-leafed lupins in Germany was just started in the 1990s, due to the breakdown of the previous cultivation of yellow lupins because of the anthracnose disease (*Colletotrichum lupini*) (Eickmeyer, 2008). Accordingly, breeding work on *L. angustifolius* in Germany lasts for about 20 years, only.

More recently, increasing evidence is given by several studies that lupin derived products are also very valuable for human nutrition. Especially novel food ingredients, like lupin corn fibre and lupin protein concentrates, are gaining attention in the last years. A number of studies have revealed bioactive properties, like hypoglycaemic and hypocholesterolaemic effects of lupin fibre and protein (Johnson et al., 2003; Magni et al., 2004; Sirtori et al., 2004; Bertoglio et al., 2011). The composition of lupin corn is also quite unique. Being high in protein (30–40%) and dietary fibre (45–50%), low in fat (6%) and with virtually no starch, lupin corn has the potential of a very low glycaemic index human health food (Sipsas, 2008).

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http://dx.doi.org/10.1016/j.fcr.2014.12.009
0378-4290/

For an extended introduction of lupin corn in human food, improvement of lupin seeds is required to meet the higher standards of the food industry. Thus far, considerable environmental impacts on seed quality and yield have been reported in several studies. Seed protein, alkaloids and yield of German narrow-leafed lupins are proved to be significantly influenced by temperature and soil pH (Jansen, 2008; Jansen et al., 2009, 2010, 2012). Bhardwaj et al. (1998) reported also a significant impact of growing conditions on seed composition of *L. albus* in the United States. In general, environmental influences seem to exceed genetic influences in trait variation (Jansen et al., 2005). This was also observed in a multi-environmental trial of Australian narrow-leafed lupin cultivars in which the environment always showed a larger effect on lupin seed quality than the genotype (Cowling and Tarr, 2004). Up to now, no evaluation of German narrow-leafed lupin breeding material regarding genetic variables and environmental stability of important seed quality and yield traits has been conducted. Therefore, the aim of this study was (i) to characterise new German *L. angustifolius* breeding lines with respect to variation in major seed quality and yield traits and to get information on the environmental impact on these traits, (ii) to calculate broad sense heritability and the expected genetic advance based on these data in order to get information on achievable improvement of respective traits by breeding, (iii) to identify environmentally stable genotypes.

2. Materials and methods

2.1. Plant material

50 genotypes of *Lupinus angustifolius* L., provided by Saatzucht Steinach GmbH & Co KG (Bocksee, Germany) were tested. The plant material comprises 42 advanced breeding lines and 8 German cultivars of *L. angustifolius* ('Probor', 'Boregine', 'Borlu', 'Vitabor', 'Haagena', 'Sonate', 'Boruta', 'Haags Blaue'). Two cultivars ('Haags Blaue' and 'Boruta') and 6 breeding lines belong to the restricted branching (determinate) type and the rest belongs to the normal branching (indeterminate) type.

2.2. Experimental design and characterisation of environments

The lupins were grown in a thre years study from 2010 to 2012 at four locations in Germany, with three sites located in Mecklenburg-Western Pomerania (Northern Germany), i.e. Bornhof (53°5′N, 12°9′O), Dratow (53°5′N, 12°8′O) and Groß Lüsewitz (54°1′N, 12°3′O) and one site located in Bavaria (Southern Germany), i.e. Steinach (48°9′N, 12°6′O). Table 1 shows sowing and harvesting dates, climate data and soil characteristics for each site. The experiments were conducted in a randomised complete block design with four replications and different plot sizes. In Steinach and Dratow the plot size was 4.5 m^2 (3.0 m × 1.5 m), in Groß Lüsewitz 4.2 m^2 (2.8 m × 1.5 m) and in Bornhof 10.5 m^2 (7.0 m × 1.5 m). Seed density of normal branching types was 100 seeds per m^2 and of restricted branching types 120 seeds per m^2.

Trial site environments were further characterised by conducting principal component analysis considering weather conditions during the growing season from March to August (Fig. 1). Loadings were represented by the amount of rainfall from sowing to anthesis (pre-RF), from anthesis to harvest (post-RF), total rainfall (total RF) and the average temperature from sowing to anthesis (pre-T) and from anthesis to harvest (post-T). Scores represent 9 environments, because trial sites Bornhof and Dratow were considered as one location as respective weather data where received from the same weather station. The denoting of the scores is as follows: BD (Bornhof & Dratow), GL (Groß Lüsewitz), Steinach (S), 10 (2010 growing season), 11 (2011 growing season), 12 (2012 growing season). In the PCA two-dimensional graph, trial sites where grouped predominantly according to the influence of the respective year. In growing season 2011 the highest total-rainfall was received, whereas in growing season 2012 a low pre-anthesis rainfall was observed. The 2010 season was characterised by high post-anthesis temperature and lowest amounts of total- and post-rainfall. Location Steinach in 2010 was characterised by highest pre-anthesis rainfall and locations Bornhof and Dratow in 2011 by the highest post- and total rainfall.

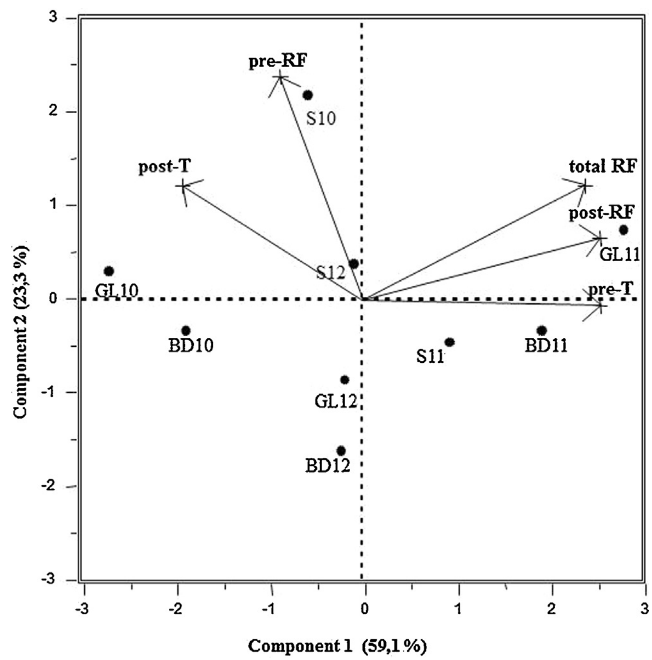

Fig. 1. Principal component analysis of trial site environments. Loadings are represented by total rainfall in mm from sowing to harvest (total RF), rainfall in mm from sowing to anthesis (pre-RF), rainfall in mm from anthesis to harvest (post-RF), average temperature in °C from sowing to anthesis (pre-T) and average temperature in °C from anthesis to harvest (post-T). Location Bornhof and Dratow are considered as one location (BD) because weather data for these sites where received from the same station. Trial sites are labelled as follows: Groß Lüsewitz (GL), Steinach (S) and Bornhof & Dratow (BD) in growing season 2010 (10), 2011 (11) and 2012 (12).

2.3. Agronomic and seed quality traits

The recorded seed quality traits comprise crude protein content, oil content, alkaloid content, raffinose-oligosaccharides (RFO) and non-starch polysaccharides (NSP). Protein-, oil-, RFO- and NSP-contents are expressed as % in flour on a dry matter basis. The alkaloid content is expressed as mg/kg flour on a dry matter basis. The considered agronomic traits are yield (t/ha), thousand seed weight (g) and protein yield (t/ha).

For the analysis of seed quality traits, two samples were prepared per genotype and experiment by mixing subsamples from two replicates each: replicates 1 and 2, and 3 and 4. Yield and thousand seed weight are based on results of all four replications at three locations (without Steinach). Evaluation of seed non-starch polysaccharides was conducted on two year trials (2010, 2011) with two replications at four locations.

2.4. Methods

Seed yield was determined after harvesting and calculated to t/ha dry matter. For examination of thousand seed weight, 30 g of lupin seed were weighed at a corn analysing machine (MARVIN-Universal, GTA Sensorik GmbH Neubrandenburg) via 2D measurement of the seeds.