

Designing breeding strategies for organic production of spring barley (*Hordeum vulgare* L.)



Aina Kokare

Propositions

1. Plant breeding for organic farming can be done in a conventionally managed breeding programme by selecting beneficial traits for organic cultivation
(this thesis)
2. The use of an organic ideotype score in a breeding programme aimed to produce cultivars suitable for organic farming also benefits conventional farming.
(this thesis)
3. Science can benefit from climate change.
4. Organic foods contain high levels of biologically active compounds that protect both plant and consumer health.
5. A zero-waste life style requires (the principles of) organic agriculture.
6. To overcome social differences in health, school gardens, cooking lessons and healthy lunches should be part of the school curriculum for children and adolescents.

Propositions belonging to the thesis, entitled

Designing breeding strategies for organic production of spring barley

(*Hordeum vulgare* L.)

Aina Kokare

Wageningen, 29 November 2022

**Designing breeding strategies for
organic production of spring barley
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of spring barley (*Hordeum vulgare* L.)

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Thesis

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Abstract

Organic agriculture is gaining importance in the agricultural sector in Europe. More and more breeders are interested to serve that market by improving varieties adapted to a such low-input farming system. By taking into account that plant breeding requires considerable input of resources and that organic agriculture is a highly diverse production system with yet a limited but growing market, it is essential to elaborate on the most effective selection programs to develop suitable varieties for organic farming. To design breeding programs for organic agriculture, it is essential to analyse appropriate initial breeding material, effective selection environments and selection criteria for field and marker assisted selection. We studied these issues in barley as example crop in three field experiments at two organically managed and two conventionally managed sites in Latvia. Modern barley varieties generally have higher yield potential than older traditional cultivars and generally shorter plant length, but still sufficient variation for traits that have high priority in organic farming such as yield, yield stability and various morphological traits contributing to weed suppressive ability. Regarding the selection environment, selection in organic conditions (direct selection) is recognized as the most effective approach. The results of this study reveal that selection for organic farming could be also integrated in a conventionally managed breeding program (indirect selection). Direct and indirect selection in early breeding stages can be equally suitable for the development of varieties adapted to organic farming, if specific selection criteria are taken care of during parental choice and progeny selection, and if in later stages of the breeding program the most promising candidate varieties are tested under various organic conditions.

This study showed that direct or indirect selection affects yield and traits contributing to weed suppression differently. In direct selection, the selection procedure where mild selection for weed suppressive ability was combined with strong selection for grain yield gave the best response. In an indirect selection approach, the best response was obtained by combining both criteria: high grain yield and high weed suppressive ability. However, indirect selection under conventional conditions proved to be less effective than direct selection under organic conditions.

In the genome-wide association study 35 QTLs were identified for four traits contributing to weed suppressive ability, and 80% of these QTLs were management-specific and only some in both organically and conventionally managed systems, which suggests that it is necessary to continue QTL discovery studies for other important traits under organic farming systems. QTL found in the organic farming systems could be used for marker-assisted selection under organic growing conditions (direct selection) and might also be useful within conventional breeding programmes (indirect selection) to select the genotypes for organic farming.

Key words: barley, breeding for organic farming, genetic resources, selection criteria, yield and yield stability, weed suppressive ability, selection environment

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Chapter 1

General Introduction

1.1 Introduction

Worldwide the production has more than doubled yield in the last century by conventional, high input agronomic practices of synthetic fertilisers and pesticides, and improved varieties (Knapp et al., 2018). Besides increasing supplies of food and other products, modern agriculture has been considered one of the causes of nitrogen surplus, greenhouse gas emissions, and contributes substantially the agrochemical pollution, soil degradation and loss of biodiversity (Kiers et al., 2008). Organic farming is based on traditional farming methods relying on ecological processes (e.g. applying organic stable manure and natural predators for pest management), biodiversity and crop cycles adapted to local conditions, and refrains from chemical-synthetic inputs. The International Federation of Organic Agriculture Movements (IFOAM – Organics International, www.ifoam.bio) has defined organic agriculture as a way of farming that combines best environmental practices, a high level of biodiversity, and sustains the health of soil, ecosystems and people. Organic production ensures not only the sustainability of agricultural production but also promotes ecological, social and ethical challenges (IFOAM, 2005).

There is an ongoing debate among scientists and politicians on the role of organic farming in limiting global climate change, while ensuring a sufficient amount of food for a growing world population (a.o. Tuomisto, 2012). Compared with conventional agriculture, lower yields in organic agriculture are the main discussion point in adopting organic management practices in agricultural production (De Ponti et al., 2012; Kirchmann, 2019). Improvement of the organic management system as well as better adapted varieties for organic farming could reduce the yield gap and enhance yield stability in organic practice (Crespo-Herrera and Ortiz, 2015).

Despite relatively small funding towards organic research, studies and breeding activities for organic systems have increased over the last decades. Still, more knowledge is required to optimise breeding strategies for varieties adapted to organic agricultural systems (see e.g. the recent EU project LIVESEED, www.liveseed.eu). The main basic questions for this comparatively new research area of breeding for organic farming systems are: which selection criteria and which selection environments are the most appropriate to select varieties for organic farming conditions? This thesis will

contribute to these themes and focus on designing breeding strategies for organic barley production in Latvia.

In this chapter more background on organic farming in Europe and Latvia will be provided, including the role of cereals in organic farming systems, and more specifically the role of barley. Then more in detail the problem will be described with respect to the needs for barley variety improvements and ways to achieve that, leading to the research objectives and research questions of this thesis, followed by the research design and methodology, and finally the thesis outline.

Organic Farming in the Europe Union

Organic farming is a fast-growing agricultural sector in the European Union (EU), as consumers' interest in organically produced goods has increased. In the EU, the total organically managed area in 2020 covered 14.7 million hectares of agricultural land (Eurostat, 2022). Organic farmland in the EU shares on average 9.1% percent of the total agricultural land. Latvia took the 6th place with 14.7% organic farming out of the total agricultural land in 2020 and was among the ten countries with the highest growth of organic farmland in the EU (LSM, 2022).

Organic agriculture in Latvia

In Latvia, during the last decade, both the number of organic farms and the area of organic farming has increased. In 2020, there were 4530 organic farming enterprises (Latvian Ministry of Agriculture, 2022). In 2020, the area of organically grown cereals was 60.849 ha. The highest proportion of organic cereal production area (ha) was composed of oats (49%), wheat (19.7%) and buckwheat (13.8%). Spring barley shared 5.6% of the total organic area of cereals in 2020. Average yield of organically grown barley in Latvia was 2.0 t ha⁻¹.

Barley breeding in Latvia

Currently, there are no private, commercial breeding companies in Latvia. Historically barley breeding was conducted by two state-owned breeding institutions (in Priekuli and Stende), which are currently merged into the Institute of Agricultural Resources and Economics (AREI, www.arei.lv).

Barley breeding is financed mainly by the government (program coordinated by Ministry of Agriculture) except for a couple of small initiatives for particular products receiving private financing. Financial resources have always been insufficient. Historically spring barley breeding began at the plant breeding station in Priekuli in 1913, and in 1920-ties started at the plant breeding station in Stende as well (Holms, 1990). The hulled barley has always been bred for both feed, malt and food purposes. The key selection criteria for conventionally grown barley are: yield under moderate agrochemical input, high protein and starch content in the grain, and disease resistances with the emphasis on powdery mildew. In 2000, breeding for hulless barley for healthy food and feed purposes was added and later included also in the organic breeding program.

Barley variety Rubiola, bred in AREI, entered the Latvian Plant Variety Catalogue in 2011 with a remark “after testing approved as suitable for growing in organic farming”. Initially, ‘Rubiola’ was selected from a conventional program and tested as advanced line in organic trials. The assesment of the Value for Cultivation and Use (VCU) for ‘Rubiola’ was performed under both organic and conventional conditions.

With the increased popularity of organic agriculture in Latvia and thanks to the initiative of the Association of the Latvian Organic Agriculture, breeding for organic farming for barley, oats, wheat, pea and potatoes is being financed by the government since 2013.

In Priekuli, the testing of barley breeding material under organic farming conditions started in 2005. Step by step, barley breeding for organic farming developed into a program that is currently more important than breeding for conventional farming. Several research projects on breeding for organic farming were performed at AREI with financial support by the EU funds to develop breeding methods and technologies. Currently, the Priekuli Research Center barley breeding program is performed on 4.5 ha, of which 2.0 ha is organically managed for selection, breeding-related research, and seed production under certified organic growing conditions.

1.2 Problem description

Needs for organic farming, main problems

One of the current EU Green deal targets is to achieve at least 25% of the EU's agricultural land under organic farming by 2030 (European Commission, 2020). Organic farming is considered one of the ways to achieve these Green Course goals. Moreover, turning to organic management practice can allow farmers to become more competitive, reduce expenditures for inputs, be less dependent on imported resources, and increase their autonomy (Bouttes et al., 2019).

Organic farming has been criticized for being lower yielding and less efficient in resource usage than conventional farming (Trewavas, 2004; Tuomisto et al., 2012; Jouzi et al., 2017). By analyzing crop production data based on large-scale organic farming Kirchmann (2019) revealed that organic yields were 35% lower for all crops in organic than conventional farming systems. Based on previous and other scientific studies, the author pointed at insufficient nutrient supply in organic farming as the main reason for the yield gap between organic and conventional farming. Furthermore, organic farming often relies primarily on modern cultivars selected under conventional management systems (Lammerts van Bueren et al., 2011). These varieties are usually adapted to intensive management with a high nutrient supply. They are also lodging resistant provided by the use of dwarf genes in cereal breeding. Several studies have shown that the performance of such varieties declines under organic conditions (Yusuff et al., 2007; Murmu et al., 2013; Chozin et al., 2017). Arable crops have less chance of producing high yields and providing yield stability under organic growing conditions. They may suffer from weed competition, nutrient deficiency, especially nitrogen, due to low mineralization activity in the soil under cold spring conditions by use of organic fertilizers, and diseases due to the lack of chemical-synthetic inputs (Lammerts van Bueren et al., 2002; Lammerts van Bueren et al., 2011; Stagnari et al., 2013; Knapp & Van der Heijden, 2018). To overcome these aspects, a different approach is required in the production practice and the breeding programs for organic farming compared to conventional breeding. To design a breeding program for varieties adapted to organic farming we should take into account not only the main factors affecting productivity in general, such as regional climatic and soil conditions but also the specific

aspects related to low input, such as: yield stability, nutrient use efficiency, and weed suppressive ability. The incorporation of the previously mentioned traits into genotypes for organic farming would help organic agriculture not only to become competitive, but it will also make conventional production more sustainable and environmentally friendly. Thereby contributing to the implementation of the European Green Deal (European Commission, 2020). Below a short overview of state of the art in literature will be discussed with respect to these three main traits to support the research questions in this thesis with respect to organic barley production.

Yield stability

Organic farmers have fewer means to directly compensate or mask negative conditions during crop growth than conventional colleagues with access to chemical-synthetic crop growth and protection inputs. Therefore, crop yields under organic conditions show higher variation over the years and locations than on conventional farms. Thus, organic farming has greater challenges than conventional farming for producing high and stable yields with good quality by adapting to organic soil fertility, weed, pest, and disease management. Therefore, achieving yield stability rather than merely yield as such is one of the cornerstones of organic farming, which ensures the persistence of the farm over the long term (Wolfe et al., 2008; Lammerts van Bueren et al., 2011; Bouttes et al., 2019; Bocci et al., 2020). High crop yield stability is also key for future crop production and breeding to adapt crops to changing climate conditions (Döring and Reckling, 2018). However, despite the essential contribution of scientific and technological developments in breeding, crop management and cropping systems, considerable fluctuation in yield still exists. Moreover, yield stability is not yet a trait included in the official testing protocols for Value of Cultivation and Use (VCU) (Peltonen-Sainio et al., 2013; Osman et al., 2015).

Selection for yield stability under organic, low-input growing conditions is a challenge for barley breeding programs, because of the relatively high interaction between genotype and environment (G×E). Accordingly, a high number of organic test environments is required, which often exceeds the capacity of the work and budget available (Muellner et al., 2014; Mühleisen et al., 2014).

For the farmer, the choice of crop varieties is often balancing between yield stability and high yield potential. Some farmers grow varieties that show reduced yield variation, while others prefer varieties that give high grain yield in good years while accepting considerable losses in poor years. The results of variety performance over different organic and low input environments would give valuable information to find the genotypes that are better adapted to specific conditions and help farmers choose appropriate varieties.

Adaption to low nitrogen availability in organic farming systems

Insufficient supply of nutrients, especially nitrogen (N), in organic farming is a major factor influencing yields (Kirchmann, 2019). Organic barley yields less compared to a conventional crop because of lower levels of nitrogen (N) fertilizations and irregular N availability due to slow nutrient mineralization from organic fertilisers, which depends on weather conditions and soil microbiological activity due to factors influencing mineralization in the soil (Osman et al., 2015; Lammerts van Bueren and Struik, 2017). The low nutrient availability, especially N can affect the reproductive development of the crop and such yield determining components as: spikelets per spike and kernels per spike, thus the improved N use efficiency is closely related to improvements of those components as well as thousand grain weight and harvest index (Tian et al., 2016). Enhancing the nitrogen availability in organic farming, includes adequate crop rotation with incorporating legumes and catch crops and timing of organic fertilisers. Besides increasing N availability with agronomics measures, specially adapted varieties to organic farming conditions are needed. Therefore, developing barley varieties with improved adaptation to low and irregular nitrogen availability is an important approach to enhancing yield in organic farming conditions. In addition, these varieties can help to improve nutrient management in conventional farming and reduce the use of mineral fertilizers as envisaged in the EU Green deal objectives.

Traits contributing to weed suppressive ability

Weed infestation is one of the most relevant issues of organic farming systems. Weeds can cause yield loss and decrease quality through competition with the crop for light, water, and nutrients (Andrew et al., 2015). In an organic management system, an adequate crop rotation, soil treatment and harrowing

at early development stages are the main possibilities to control weeds ecologically (without herbicides) diminishing the negative impact of weeds on crop yields as well as decreasing the weed seed bank in the soil for following crops (Lutman et al., 2013; Harker and O'donovan, 2013).

Besides improving the weed management practice, a sustainable and economically viable option is the choice of cultivars with superior competitive ability against weeds (Worthington and Reberg-Horton, 2013; Mahajan et al., 2020). Such varieties can suppress weeds and reduce the additional costs on mechanical weed control measures and contribute to a long-term weed management strategy (O'Donovan et al., 2007; Bastiaans et al., 2008; Andrew et al., 2015).

Competition is based on the ability of a crop cultivar to access light, nutrients, and water resources in a limited space, thus suppressing the growth and reproduction of nearby weed species (Worthington and Reberg-Horton, 2013). Varieties differ in their ability to cope with the negative influence of weeds on crop growth. Plant competitiveness against weeds is associated with a wide range of plant morphological and physiological traits and their interactions (Hoad et al., 2005; Hoad et al., 2008). The straw length has been shown to be the most important trait for weed suppressive ability and also indirectly for minimising yield loss in the presence of weeds (Lemerle et al., 1996; Mahajan et al., 2020). For example, Murphy et al. (2008) have found a negative association between weed weight and plant height in wheat. Tall barley varieties were found to be less susceptible to creeping weeds pressure than short ones (Østergård et al., 2008). Increased plant height and early maturity were associated with reduced weed biomass, while strong early season vigour was related to increased yield, increased spikes per m², and reduced weed biomass in wheat (Watson et al., 2006). Mason et al. (2007) proposed that earlier flowering and maturity could help the plants outcompete weeds and produce better yields in organic systems. Mahajan and Chauhan (2013) found that traits such as high and early seedling vigour with rapid leaf area development during the early vegetative stage are likely to be most helpful for weed suppression in rice. Early vigour, plant height at early growing stage, and leaf index were found to contribute to weed competitive ability in wheat (Kissing Kucek et al., 2021). A higher ground cover, presumably induced by planophile leaf inclination, in wheat reduced weed

ground cover, was observed by Drews et al. (2009). A study of Christensen (1995) showed significant correlation between weed dry matter and rate of canopy height development for barley. Mahajan et al. (2020) indicated a greater height and high panicle production are desirable attributes for weed competitiveness in barley genotypes. Also Piliksere et al. (2013) studied weed suppressive traits in more or less weedy plots that were organically managed. In that study with 20 barley genotypes, traits such as erectophile crop growth habit at tillering (at growth stage (GS) 29) and at booting (GS 39) stages, as well as crop canopy height at the beginning of stem elongations stage, correlated negatively with dry weight of total of the annual weeds, but no correlation with the weight of perennial weeds was found. Piliksere et al. (2013) also found that a higher crop plant height at maturity stage (GS 92) resulted in lower dry weight of total weeds, and that a longer growth period from sowing to heading and to ripening resulted in higher total weed dry weight.

Knowledge about the trait association with competitive ability against weeds, provides the possibility to select indirectly for weed suppressive genotypes in weed free selection fields (Hansen et al., 2008; Bertholdsson, 2011; Mahajan et al., 2020). The ability to suppress weeds should be a selection criterion with high priority in breeding programmes for organic farming.

Farmers' barley variety ideotype for organic farming in Latvia

With an increase of the interest of Latvian farmers in organic farming, the question arose of suitability of conventional varieties to organic growing conditions. The first barley variety trials under organic conditions in Latvia have shown that modern varieties performed better than older varieties but are nevertheless not optimal (Strazdina and Bleidere, 2004; Kokare and Legzdina, 2006). During this thesis research, we carried out field seminars with farmers' participation, to find out what they considered as the main problems and what were their desires in terms of variety properties and choice. The aim of this investigation was to get a good overview of the characteristics of spring barley cultivars required by Latvian organic barley growers to arrive at better adapted cultivars for organic farming systems.

The results show that grain yield is of the highest priority for organic farmers, and 2 to 3 t ha⁻¹ is considered an acceptable level for Latvian conditions, but

3-4 t ha⁻¹ is considered a more optimal level (Table 1.1). Refraining from herbicides makes competitive ability of varieties against weeds important, especially in the relatively low-profit barley crop, for which Latvian farmers cannot afford to invest much labour such as mechanical weeding.

In order to improve the barley competitive ability against weeds at the beginning of the crop growth period, the farmers recognized that it is important to reach rapidly a high density of the crop canopy. That will help the soil shading and prevent weeds to prevail. Harrowing plays an important role in weed control in organic farming, but most of the farmers do not use it due to lack of investment into modern equipment. One of the participants did not consider mechanical weeding out of fear of damaging the crop, ultimately decreasing the yield. Also Osman et al. (2016) recognised that the ability to tolerate harrowing or recover after harrowing is an important trait for organic farmers to include in variety evaluation. Characteristics such as plant growth habit at the tillering stage and early vigour were also mentioned to help the variety to compete with weeds at early stages of the crop development.

Farmers indicated that also in the second half of the growing period, there is a risk of large amount of weeds that will make the crop harvest difficult and cause additional costs in the drying process. The length of the plant could help to cope with that problem, but the plant length cannot be too large as it is associated with lodging, to which farmers gave great importance. Although under organic conditions, the risk of lodging is not as high as under conventional conditions, where high levels of synthetic fertilisers are used, still, low lodging resistance can negatively affect yield and quality.

Where disease resistances were concerned, loose smut was pointed out as the most serious pathogen in organic barley production due to the practice of farm saved seeds and the lack of effective seed treatments also in organic commercial seed production.

The length of the growing period was considered by the farmers important with respect to the management of labour on the farm. Early and medium early varieties were more desirable than late varieties. Earliness allows harvesting of crops in time before the probability of rain becomes high, which can affect grain quality. An early crop also allows to distribute labour more evenly over the harvest season.

Table 1.1 Importance of the barley traits for Latvian organic farming, recognised by farmers giving scores from 1 (low importance) to 4 (high importance), 2010-2011

Traits	View of organic farmers	
	Priority scores	Trait expression
Grain yield	4	3 - 4 t ha ⁻¹ is an optimal level
Plant growth habit	1	Preference was given to more prostrate growth habit
Resistance to harrowing	2	Resistant, plants recover well after damages from harrowing
Tillering capacity	3	The better tillering capacity, the denser the canopy, which can compete well with weeds; trait is directly related to the yield
Early vigour	3	Good early vigour
Canopy height at stem elongation stage	2	The higher the better
Leaves	2	Broad, declining
Plant height at maturity	2	Medium to tall, 85 – 105 cm
Lodging resistance	3	Good lodging resistance
Loose smut <i>Ustilago nuda</i> (Jens) Rostr.	3	Resistant
Net bloch <i>Pyrenophora teres</i>	2	Resistant
Powdery mildew <i>Blumeria graminis</i>	2	Resistant
Length of growth period	3	Short to medium, preferably mature at the beginning of August
Nutrient use efficiency	3	Efficient uptake of nutrients and water from the soil under conditions of low N as well as under periodic drought stress.
Protein content, %	2	High, above 14%
Volume weight, g l ⁻¹	2	High, above 700 g l ⁻¹
Thousand grain weight, g	2	High, above 40 g
Starch content, %	1	Above 62%

Choosing appropriate conditions to select varieties for organic farming

As more and more breeders involved in conventional breeding programs also aim to serve the organic market, not only questions concerning selection criteria of relevance for varieties suitable for organic farming, but also questions around the choice of the most efficient selection environment appear (Wolfe et al., 2008). The question is whether organic or conventionally managed selection fields can or should be used. Organic farming is a highly diverse production system and it produces crops without the use of any synthetic fertilisers or pesticides. Since plant breeding requires considerable input of resources and the market for organic varieties is limited, it is essential to find the most appropriate and cost effective selection conditions that will provide acceptable to good varieties for organic farms.

The views on an appropriate selection environment for organic farming are contrasting. Some authors argue that conventional plant breeding cannot always provide suitable cultivars for organic farming (Murphy et al., 2007; Dawson and Goldringer, 2009; Lammerts van Bueren et al., 2011; Chable et al., 2020). The authors note that the conventionally bred varieties aim to maximize the responsiveness to input and the economic efficiency of agriculture, and they cannot cope well with organic management practice. They also argue that essential traits such as competition with weeds and the ability to use nutrients efficiently from organic nutrient resources perform differently between organic and conventional farming systems and cannot be selected under conventional conditions. For an example, Kamran et al. (2014) found that cultivars grown under the organic management system were earlier flowering, lower yielding, and lower test weight than their performance under the conventional management system. However, Mahajan et al. (2020) suggest that the selection of tall plants and high tillering capacity for weed competitiveness could be carried out in a weed-free environment, while weed tolerant genotypes should be selected in weedy conditions.

The genotype by environment interaction ($G \times E$) is a highly important aspect in breeding. The selection environment has great effects on the performance of cereal varieties under organic conditions (Miko et al., 2014). Some authors consider that indirect selection under conventional conditions is less effective than direct selection under organic conditions (Brancourt-Hulmel et al., 2005;

Murphy et al., 2008; Reid et al., 2009; Ceccarelli et al., 2000; Ceccarelli, 2015). They support the selection under conditions close to the target environment where the varieties will be grown in the future. Studies conducted with comparing already registered wheat varieties under conventional and organic conditions reveal that for organic environments, direct selection is more efficient than indirect selection (Kitchen et al., 2003; Przystalski et al., 2008; Annicchiarico et al., 2010; Kamran et al., 2014). Kirk et al. (2012) used spring wheat crosses, initially made for conventional purposes, for selection under organic and conventional growing conditions. Authors have found that the populations selected in organic environments had higher yield than those conventionally selected if grown at organically managed sites. Grain quality parameters such as protein content and thousand grain weight were higher in both management environments for the populations selected under organic conditions. From this experiment, it was concluded that selection in organically managed field conditions offered advantages over indirect selection in conventionally managed conditions. In regard to this, some (small) plant breeding programs fully managed under organic conditions have been initiated in Austria, France, Germany, the Netherlands and Switzerland for different crops (Crespo-Herrera & Ortiz, 2015).

However, the breeder's choice of conditions to select varieties suitable for organic farming often depends on economic aspects too. In regard to this, there are examples of commercial wheat breeding schemes for organic farming where selection at early rounds of selection is conducted under conventional conditions, and where in the later generations, the programme is split into an organic and a conventional part (Löschenberger et al., 2008; Miko et al., 2014). That breeding strategy allows first to select for highly heritable traits under more controlled conventional conditions, and in later generations, select for the traits that are more influenced by the environment, under organic conditions.

Crespo-Herrera & Ortiz (2015) argue that a conventional breeding program also has to deal with the genotype-by-environment interaction because of different input levels in conventional farming systems, which can vary to great extent. To this aspect, Dawson and Goldringer (2009) pointed out earlier that the efficiency of indirect selection under conventional conditions can depend on the degree of differences between the systems. Crespo-Herrera & Ortiz

(2015) propose the implementation of shuttle breeding between organic and conventional farming to open the possibility of developing cultivars adapted to both conditions.

Mapping of the traits relevant for organic farming

Molecular marker technology has grown rapidly since the eighties of last century, and is now increasingly used by many breeding companies to enhance selection efficiency (Dwivedi et al., 2007; Sivolap, 2013). Marker assisted selection (MAS) has greater efficiency and reliability if traits are very hard to score making phenotyping expensive or traits are highly affected by environmental variation. Molecular markers can both be used for the choice of the parents for crossings and at different stages of the selection process by selecting for certain genes or gene alleles (Desta and Ortiz, 2014). The main advantage of the use of molecular markers in early generations is the ability to identify loci or combinations of gene alleles in plants in a short period of time. This saves field area and time that would otherwise be used for the phenotyping and multiplication of rejected accessions not containing the desired alleles (Dubcovsky, 2004; Miedaner and Korzun, 2012).

Until now, genetic mapping has been carried out mainly for traits important under conventional farming, by phenotyping them under conventional conditions. With the increased availability of technological methods, the development of high-throughput facilities and the decrease of costs of the markers, the possibility to use molecular markers in breeding varieties for organic farming systems will increase (see e.g. Lazzaro et al., 2019). The mapping of traits important for organic farming, phenotyped under both organic and conventional conditions can be important for organic breeding to find out whether other alleles appear for certain traits under organic conditions than under conventional conditions (Lammerts van Bueren et al., 2010). There are not many studies using GWAS that describe the effect of organic and conventional conditions on the quantitative traits. In a study on wheat, Zou et al. (2017) pointed out that not all identified QTLs are common in both conventional and organic management systems. Therefore, it is of interest to further study whether all QTLs detected in conventional management systems can be used directly in the selection of the genotypes for organic farming. To our knowledge, that has not yet been done for barley. It is necessary first to

increase the knowledge about the degree to which the traits contribute to performance under organic conditions before traditional field selection methods can be complemented by selection with molecular markers.

1.3 Research objectives and main research questions

The overall objective of the research reported in this thesis is to design of a barley breeding approach for organic farming. The breeding strategy includes a research-based choice of selection criteria as well as the best selection environment to obtain cultivars adapted to organic farming conditions.

The research questions are:

1. How do varieties differ in yield and yield stability under conventional and organic conditions?
2. How do different morphological physiological characteristics related to the weed suppressive ability perform under organic and conventional farming growing conditions?
3. Is selection for barley varieties adapted to organic farming systems more effective under organic conditions than under conventional conditions?
4. How does direct selection under organic and indirect selection under conventional conditions of barley genotypes affect traits relevant for organic farming systems?
5. Can specific genomic regions associated with traits favourable for organic farming be identified in an association mapping population?

1.4 Methodology

This thesis is based on experimental field work, laboratory research, statistical design and data analyses. For the purpose of the study, spring barley (*Hordeum vulgare* L.) was chosen, which plays an important role mainly as a feed and also food crop in Latvia, under both conventional and organic farming systems.

Three field trials were performed to answer the five research questions. The experiments were carried out at the Institute of Agricultural Resources and Economics, Priekuli Research Centre (AREI Priekuli RC) (latitude 57.3148 °N, longitude 25.3388 °E). Field trials were carried out during the years 2006 – 2012. To compare selection results and performance of barley genotypes between conventional and organic environments, two organic and two

conventional sites were selected for all project activities. Organic 1 (O1) – situated at the Priekuli Plant Breeding Institute in the organic crop rotation and Organic 2 (O2) – at an organic commercial farm, to add a possible target environment. The other two sites were conventionally managed fields of the Priekuli Plant Breeding Institute: Conventional 1 (C1) with medium input located in a crop rotation for breeding purposes and Conventional 2 (C2) with high/medium input management practice situated in the institute's seed production crop rotation fields.

The part of the project that included molecular analyses was carried out in cooperation with the Faculty of Biology of the University of Latvia.

The overall methodology applied for the 5 experiments was as follows:

For **Experiment 1** a set of 10 two-row contrasting old and modern spring barley accessions was evaluated at the two organic and two conventional growing sites during three growing seasons (2006-2008). The experimental design was a randomized complete block design, with four replications. Yield and yield components were analysed.

For **Experiment 2** the same field trials with the 10 barley accessions under organic and conventional conditions as in Experiment 1 were used, and traits contributing to weed suppressive ability were evaluated and analysed.

For **Experiment 3** two cross combinations aimed at organic agriculture were chosen, derived from parents e.g. contrasting in maturity date, plant length, development rate, yield potential. The parents for both cross combinations were also included in the previous experiment. The selection procedures with similar selection pressure under naturally occurring disease infection were carried out parallel in each of these breeding populations at the four growing sites (two organically and two conventionally managed) from 2006 until 2009 (F3-F6). The five best lines out of each of the two populations selected in each separate environment (in total 40 lines) were then compared at all four sites during two subsequent growing seasons, in 2010-2011. An organic ideotype score was developed and applied in the selection process, which served to consistently rate the phenotypic performance of the lines according a weighted rate for each trait of importance for adaptation to organic farming.

For **Experiment 4** a large set of genotypes (134) was grown at four locations (two organic and two conventional) during three seasons (2010-2012). Varieties of Latvian and foreign origin and breeding lines from selection

nurseries were included in the study. Based on the dataset of the three years' performance of the 134 genotypes in four environments, we mimicked a selection procedure in the first two years (2010 and 2011) and then used the third year's data to compare the performances of the selected genotypes derived from all four sites under the organic O1 and O2 sites.

For **Experiment 5** a genome-wide association study (GWAS) of barley traits important for environmentally friendly and organic farming was conducted. For this analysis, the phenotyping results of Experiment 4 were used with the difference that the set of genotypes was supplemented by 19 genotypes (in total 153).

1.5 Thesis outline

The outline of this thesis is shown in Figure 1.1. The thesis consists of seven chapters which comprise crop selection material, selection criteria, environment and schemes and provide body for designing a barley breeding strategy for organic farming.

Chapter 1 provides the background information on the conditions of crop growth in organic farming and how these conditions differ from those of conventional agriculture. It considers the need for effective selection methods and selection criteria to be applied to create barley varieties which perform well in organic farming. This chapter contains key problems addressed, overall objective and research questions addressed in this thesis.

Chapter 2 is based on Experiment 1 and comprises the three main stages of the breeding process: the choice of selection material, selection criteria, and selection environment. It investigates how the adaptation of barley varieties of different origin and time of release differs under various organic and conventional conditions. The highly relevant traits for organic farming such as yield and yield stability of old and modern varieties under organic and conventional conditions were compared. The heritability for yield and yield components under organic and conventional conditions were analysed. Also the question was addressed how the ranking of the old and modern (conventionally bred) varieties changed under organic conditions compared to conventional conditions? Results led to suggestions for the suitability of the old and modern conventionally bred varieties for organic farming as well as

for the most appropriate growing conditions for selection of genotypes adapted to organic farming.

Chapter 3 is based on Experiment 2 and is connected to the three main stages of the breeding process: the choice of selection material, selection criteria, and selection environment. It analyses how different morphological and physiological characteristics related to the weed suppressive ability of ten barley varieties, released in different periods of time, perform under organic and conventional farming growing conditions. The results led to recommendations for traits to prioritise to ensure the establishment of good crop ground cover at early growth stages and to provide good weed suppressive ability.

Chapter 4 is based on the results of Experiment 3, and deals with question how effective selection in breeding populations carried out under conventional conditions is in comparison to the selection under organic conditions to obtain genotypes suitable for organic growing conditions. An organic ideotype score (OIS) was developed and applied with the aim to combine several desired traits into one genotype. The differences in yield, weed suppression ability between the lines selected under conventional conditions compared with those selected under organic conditions were analysed. On the basis of the results recommendations for the best selection environment are given, taking into account the differences of the two populations.

Chapter 5 is based on the results of Experiment 4 and analyses how effective is direct (under organic conditions) and indirect (under conventional conditions) selection on grain yield and weed suppressive ability for organic farming, using a dataset of a three year field trial with a large set of genotypes in multiple year combinations (order). Based on the outcome, recommendations are given for the best combination of the selection criteria when performing selection either under organic conditions or under conventional conditions.

Chapter 6 is based on the results of a three year field trial (see Experiment 4) using the phenotypic evaluations of 153 genotypes for a molecular-genetic study to establish the chromosome regions associated with specific traits that are beneficial to organic farming. Identification of such chromosome regions will provide an opportunity to broaden the knowledge about genetic control of the traits relevant for organic farming. It also provides an opportunity to

complement traditional selection methods with molecular marker-aided selection.

Chapter 7 summarizes and discusses the main findings of research chapters 2-6 and describes an optimal breeding scheme for organic barley breeding as well as giving the recommendations for improvement of existing conventional breeding programs aimed at organically grown barley varieties.

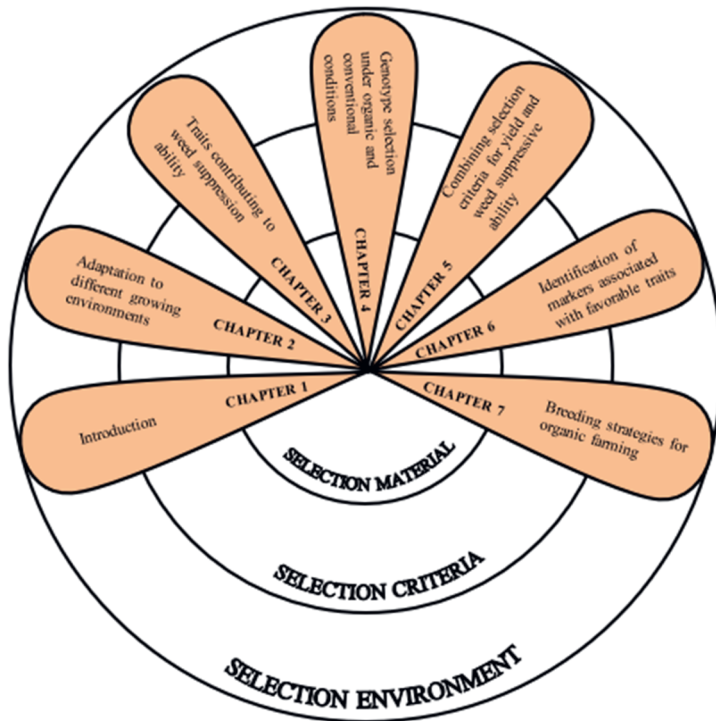


Figure 1.1. Schematic overview of the research design

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Chapter 2

Performance of spring barley (*Hordeum vulgare*) varieties under organic and conventional conditions

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Abstract

Organic agriculture needs spring barley varieties that are adapted to organic growing conditions and have good and stable grain yield across years, even under less favourable growing conditions. The aim of this study was to compare how varieties differ in yield and yield stability under conventional and organic management conditions. The results help to decide under which growing conditions selection of genotypes for organic farming is most effective. Grain yield and yield components of 10 varieties were estimated in field trials for three years at four sites: two conventionally and two organically managed sites. Varieties differed in stability: some varieties had high yield under conventional conditions and relatively high and stable yield under organic conditions. Heritabilities for yield and yield components were lower under organic (especially in the field with low weed control) than under conventional conditions. Heritabilities for yield components were lower than those for yield itself. Selection for yield components, therefore, may be less effective than selection directly for grain yield. Our data showed that generally the top performing cultivar under conventional conditions also performed as the best under organic conditions, but there were also exceptions. Therefore, we conclude that selection of genotypes for organic farming may take place under conventional conditions, but that a final testing should be conducted under organic conditions to confirm the suitability of the selected varieties for cultivation on organic farms.

Keywords:

barley, heritability, organic farming, yield components, yield stability

2.1 Introduction

Organic agriculture is defined by the International Federation for Organic Agricultural Movements (IFOAM, 2013) as a production system that sustains the health of soils, ecosystems and people. It relies on ecological processes, biodiversity and cycles adapted to local conditions. Organic agriculture searches continuously for optimisation of the farming systems through agronomic improvements and it needs adequate varieties to realise its potential. In Latvia, the area under organic agriculture is increasing rapidly and accounted for about 10 % (approximately 184,120 ha) of the total crop production area in 2011 (Agricultural Report, 2012).

Barley is the main cereal crop for feed in Latvia. The current yields under organic conditions range from approximately 1.5 to 4.5 t ha⁻¹, compared to 4.0 to 6.0 t ha⁻¹ in conventional farming (unpublished data summarized from 18 farms over two years in the project “Technological solutions in cereals production in Latvia” financed by the Latvian Ministry of Agriculture). Those differences in yield range are mostly due to differences in the crop management system, especially with respect to weed control and the level of fertilisation. In conventional barley production the average amount of applied nitrogen (N) ranges from 60 to 150 kg ha⁻¹ whereas in organic barley production the amount of nitrogen applied through organic fertilisers is approx. 30-40 kg ha⁻¹. The main sources of nitrogen in organic farming are stable manure, green manure or crop residues. Mechanical weed control during crop growth establishment is often insufficient as knowledge and equipment for adequate mechanical weed management is often lacking. Also there are in organic cereal production no other means available to control pathogens causing e.g. netblotch and leaf stripe than choosing resistant varieties and apply adequate crop rotation. For some seed-borne diseases there are more options, such as hot air treatment for common bunt (*Tilletia caries*) for wheat and loose smut (*Ustilago nuda*) for barley (Forsberg et al. 2003), and seed treatments based on mustard powder for bunt. Latvian organic farmers still depend on varieties developed for (high-input) conventional agriculture, as plant breeding in Latvia has only recently included a focus on organic farming. ‘Rubiola’ is the first Latvian spring barley variety, which was registered specifically for organic farming; the selection for this variety was conducted under conventional conditions but testing in the final breeding stage

was carried out under organic growing conditions (Legzdina et al., 2008). Until recently, most organic variety research in Latvia concentrated on the evaluation of conventionally bred varieties grown under organic conditions (Strazdina and Bleidere, 2004; Legzdina et al., 2005; Kokare and Legzdina, 2006). From the results, it has been argued that organic growers urgently need varieties that are better adapted, as levels of important traits such as weed competitiveness and adaptation to low nitrogen availability in conventional varieties are inadequate.

Organic farmers have fewer tools at their disposal than conventional farmers to influence the growing environment to accommodate their crops. Therefore, organic breeding programs should aim for varieties that can cope with varying levels of abiotic and biotic stress factors without excessive fluctuations in performance level. Successful barley varieties for organic conditions should not only show a good yield under favourable growing conditions, but also a good yield stability across different years and farms under less favourable growing conditions. For economic reasons conventional breeders involved in breeding for organic farming also want to know which is the most appropriate selection environment to obtain varieties that are adapted to organic management conditions. Testing and selection under organic management then could be an option, but the question is whether the heritability for relevant traits may be too low under low-input and heterogeneous conditions (e.g. Ceccarelli, 1996).

To get more insight into the prerequisites for an efficient breeding program to improve adaptation of two-row spring barley for organic, low-input growing conditions in Latvia, a three year field trial was conducted. The performance of a set of contrasting varieties grown in two organically and two conventionally managed fields with different input levels was compared. The research focused on the following questions: (a) do the varieties differ in yield and yield stability under organic conditions and conventional management conditions? (b) does the ranking of the varieties change under organic conditions compared to conventional conditions? and (c) is heritability for yield and yield components under organic growing conditions lower than under conventional conditions? These questions are necessary to decide under which growing conditions selection for barley genotypes for organic farming is most effective.

2.2 Materials and methods

Experimental design

A diverse set of 10 two-row old and modern spring barley varieties (Table 2.1) was evaluated during three growing seasons (2006-2008) in four replicates of 12.3 m² plots in a randomised complete block design at four sites: two conventionally and two organically managed sites (Table 2.2).

Table 2.1 Description of the varieties included in the variety trials in Priekuli 2006-2008, based on a priori experience at the State Priekuli Breeding Institute

Variety	Country of origin	Year of registration or market release	Characteristics
Rubiola	Latvia	2011	tall plants, bred for organic farming
Idumeja	Latvia	2000	medium-tall plants, early plant growth, early maturing
Annabell	Germany	1999	short plants, high-input type,
Ansis	Latvia	1995	short plants, high-input type
Inari	Finland	1994	medium-tall plants, medium early maturing
Anni	Estonia	1993	short plants, good yield under low-input conditions, good stress resistance
Abava	Latvia	1978	tall plants, good yield under poor growing conditions
Dziugiai	Lithuania	1947	tall plants, very rapid early development, resistant to acid soil conditions
Primus	Sweden	1901	very tall plants, late maturing, high TGW, test weight
Latvijas vīetējie	Latvia	landrace ~ 1800	very tall plants, very late maturing, high TGW, test weight

Plant material

All varieties originate from Baltic or Nordic regions (Table 2.1), are generally adapted to local climate conditions and have been grown in Latvia except for 'Inari', 'Anni' (still grown in Estonia), 'Primus' originated from Sweden and 'Dziugiai' (planned to be reintroduced as a heritage variety in Lithuania).

Currently 'Abava', 'Annabell', 'Ansis', 'Idumeja' and 'Rubiola' are in the Latvian Plant Variety Catalogue. 'Annabell' was the variety most widely grown in Latvia during 2005-2011. 'Abava' and 'Rubiola' are currently recommended for organic farming in the catalogue; organic farmers grow also 'Idumeja' and 'Annabell'. Our choice for this set of varieties was based on their different times of release, some specific traits which might be beneficial for organic farming, e.g. early maturing, rapid development, tall plants, good stress tolerance. Varieties that were contrasting for traits important for conventional and organic farming were included e.g. short-tall, early-late maturing etc. (see Table S 2.1).

Sites

For the comparison between conventional and organic conditions two different sites within each farming system were included (Table 2.2). Two conventional (C1 and C2) and organic O1 sites were situated within approximately 1 km distance from each other. The conventional sites were part of the Priekuli Plant Breeding Institute conventionally managed fields. C1 was part of the breeding and experimental fields, and C2 was situated in the commercial seed production field. Organic site O1 was part of the Priekuli Plant Breeding Institute's organically managed trial fields. The organic site O2 was within 5 km distance from institute fields and was located at an organic, commercial farm. The organic fields at both sites have been officially certified for organic agriculture for more than 5 years. All four sites had a similar sod- podzolic soil type with light loamy soil texture. The largest differences are based on the management differences, see Table 2.2.

The nitrogen availability (N fertilisation + N in soil) in both organic sites was similar over the three years, viz. approx. 60 kg N ha⁻¹. However, the input for weed control differed to a large extent. At the organic farmer's field (O2) no weed control was applied, as is the case in most of the current organic barley production fields in Latvia (see introduction), whereas at the institute's organic field (O1) adequate mechanical weed control was applied (Table 2.2). The data from site O2 in 2006 were not included in the statistical analysis due to an extremely low yield level of 0.31 t ha⁻¹ and an average of three kernels/tiller. This crop failure was due to early drought, poor establishment and subsequent high weed pressure.

Both conventional fields were treated according to standard agricultural practices in Latvia, including the use of synthetic fertilisers, herbicides and insecticides, but no fungicides (Table 2.2). The target seed rate at all four sites was 400 germinating seeds per m². The main difference between the two conventionally managed trial fields was the level of nitrogen application; a medium-high nitrogen application level (ca. 80 kg N ha⁻¹) at C1 and a high nitrogen application level (120 kg N ha⁻¹) at C2.

Table 2.2 Description of the soil characteristics and crop management systems of the trials under medium-input organic (OI), low-input organic (O2), medium-input conventional (C1) and high-input conventional (C2) in Priekuli, 2006-2008

Experiment Site	Year	Sowing date	pH	P ₂ O ₅ (mg kg ⁻¹)	K ₂ O (mg kg ⁻¹)	Humus content (g kg ⁻¹)	Available N in soil (kg ha ⁻¹)	Fertility management			Management of	
								Precrop	Type	Amount	Diseases and pests	Weeds
Organic 1 (OI)	2006	28.04	5.9	159	120	23	48	Rape as green manure	Rape as green manure	20 tonnes ha ⁻¹ (ca. 26.5 kg N/ha)	No	1 x harrowing at tillering stage
	2007	19.04	5.1	92	95	17	33					
	2008	23.04	6.0	133	126	19	16	After potatoes				
Organic 2 (O2)	2006	8.05	6.0	242	157	35	33	Perennial grassland	Manure (cows)	20 tonnes ha ⁻¹ (ca. 41 kg N/ha) for precrop	No	No
	2007	2.05	6.1	236	114	35	38	Perennial grassland				
Conventional (C1)	2008	30.04	6.0	183	123	23	12	Oats + pea				
	2006	3.05	5.9	155	124	20	31	Potato	Inorganic (Kemira 18-9-15)	N 80 kg ha ⁻¹ . 40 kg ha ⁻¹ of P and 68 kg ha ⁻¹ of K	Insecticides: Fastak 50 (alfa-permethrin), Sumi-alfa (esfen valerate)	Granstar Premia 50 SX (tribenuron-methyl) 50 % + Primus (florasulam)
	2007	27.04	5.4	161	137	25	24				Fungicides: not applied	
	2008	26.04	6.4	400	153	27	8				No	Secator 19 d.g. (NA-methyl- iodosulfuron)
Conventional (C2)	2006	8.05	6.5	129	202	25	48	Potato	Inorganic (Kemira 18-9-15) and NH ₄ NO ₃	120 N kg ha ⁻¹ (100 kg ha ⁻¹ N before sowing and 20 kg ha ⁻¹ at tillering stage), 50 kg ha ⁻¹ P and 83 kg ha ⁻¹ K	Insecticides: Fastak 50 (alfa-permethrin) Sumi-alfa (esfen valerate) Fungicides: not applied	Granstar Premia 50 SX (tribenuron-methyl) 50 % + Primus (florasulam)
	2007	26.04	6.1	199	130	19	30				No	
	2008	30.04	6.3	294	145	23	9				No	Secator 19 d.g. (NA-methyl- iodosulfuron)

Parameters

During the experiment the following parameters were evaluated: grain yield (GY, t ha⁻¹), number of tillers per m² (NT, measured after harvest on the basis of the stubble; two counts per plot in 0.25 m² frame), and thousand grain weight (TGW, g). The number of kernels per tiller (NK) was calculated from the following formula:

$$NK = GY/NT/TGW*100,000.$$

Statistical analysis

Analysis of variance (Anova) and the calculation of phenotypic correlation coefficients were carried out using GENSTAT 14.0 (2011). Anova was used to determine the impact of the organic versus conventional management systems and other factors such as year and site effects on the yield and yield components. The statistical model used was a split plot structure with environment/block/plot for the experimental design factors (block statement in Genstat; environment is the combination of a year and a site within a farming system) and year × (farming system/ site) × variety for the treatment factors (sites nested within farming systems and crossed with variety and year). This model allows to split up the variance among sites into a part that is due to the effect of farming systems and another part that is due to site effects within each farming system (local conditions of the site plus management on that site), while still being able to contrast site effects within and across different management systems. Significance of pairwise differences between levels of each treatment factor were assessed using the least significant difference (LSD) tables for each treatment factor from Anova ($\alpha = 0.05$). An additional analysis was done to calculate and compare variance components: the REML procedure for analysis of linear mixed models of Genstat was used, using the same structure as was used in the Anova, this time with all terms designated as random factors.

For comparison of the ordered yields of the varieties in the different environments, Spearman's rank correlation coefficient (R_s) between sites and treatments was calculated using the following formula:

$$R_s = 1 - \frac{6 \sum d^2}{n(n^2 - 1)}$$

where $\sum d^2$ is the difference in rank change of each variety squared and summed over the 10 varieties ($n = 10$). Significance was assessed at the 95 % confidence level.

Heritability was estimated from the variance components, for yield and yield components, for organically and both conventionally managed environments separately. The heritability (h^2) was estimated per site and was expressed as a percentage, using:

$$h^2 = (V_g)/(V_g + V_{gy}/y + V_e/ry).$$

where (V_g) denotes genotypic variance (between varieties), V_{gy} genotype x year interaction variance, V_e error variance and r denotes the number of replications and y the number of years. V_{gy}/V_g , the ratio of $G \times Y$ to genotype variance components was used to show relative size of these effects on variation of traits. The value of V_{gy}/V_g ratio > 1.0 indicates a larger contribution of the $G \times Y$ interactions to the variance than that of genotypic differences per se for a trait.

Finlay-Wilkinson regression

The joint regression analysis method proposed by Finlay and Wilkinson (1963), Eberhart and Russell (1966) and Perkins and Jinks (1968) was used to calculate, per variety, the regression coefficient for the slope (b) of the Finlay-Wilkinson regression line, and variance due to deviation from regression (s^2d) as parameters of adaptability and stability, respectively. These parameters were estimated using GENSTAT 14.0 (2011). According to Finlay and Wilkinson (1963), genotypes with a slope larger than 1 are responsive to favourable environments, but the genotypes with a high yield over environments and a slope close to 1 would be stable and have wide adaptation. Genotypes with relatively high average yield values and a slope lower than 1 perform relatively well under unfavourable growing conditions. Eberhart and Russell (1966) proposed the deviation from regression (s^2d) as an

alternative parameter of stability. A variety with a low (s^2d) value is presumed to be highly stable. Each genotype was defined by three values: (1) mean yield over 11 environments, (2) the slope in the Finlay-Wilkinson regression (b), and (3) the deviation from the regression line (s^2d value).

2.3 Results

Comparison of grain yield and yield components over the environments

The mean grain yield (GY) of all varieties over three years did not differ significantly between the conventional medium input C1 (3.87 t ha⁻¹) and high input C2 sites (3.73 t ha⁻¹) (Table 2.3). The mean GY under the organic conditions of O1 (3.12 t ha⁻¹) and O2 (2.69 t ha⁻¹) was significantly lower than under the conventional growing conditions. The mean GY of the organic farmer's field O2 was significantly lower than that of organic O1.

In all three years there was a high correlation for GY over the 10 varieties between C1 and C2 ($r = 0.88-0.95$; Table S 1). High correlations were also found between O1-C1 and O2-C1, (ranging from 0.74 to 0.89) and between O1-C2 and O2-C2 (0.64-0.91). If old varieties 'Latvijas vīetējie' and 'Primus' were excluded from the analysis, correlations between growing sites for GY, NT and NK were weaker, but stronger for TGW, in comparison to the correlations of the whole set of varieties; however the trend remains the same (data not shown).

The mean yield in O1 was 48 % (in 2007) lower than the average value of the two conventional conditions whereas in 2008 no significant differences were found. The performance of O2 was poorer with a yield that was 57 % (2007) and 12 % (2008) lower than at the conventional conditions. GY under O2 conditions was 29 % (in 2007) and 17 % (in 2008) lower compared to O1.

On average over all years, the low-input management level of O2 also affected the yield components (NT, TGW) and resulted in significantly lower values compared to O1 and to both conventional sites (Table 2.3). The least affected component was the NK. However, the TGW was extremely low in 2007 due to incidence of cockchafer (*Melolontha melolontha*) in O2 which damaged plant roots and caused premature senescence.

Table 2.3 The average grain yield (GY, t ha⁻¹), number of tillers (NT, number per (TGW, g) and number of kernels per tiller (NK) of ten varieties grown at two organic sites (O1 and O2) and at two conventional sites (C1 and C2), 2006-2008

Traits	Site	Years of observations			Mean
		2006	2007	2008	
GY (t/ha)	C1	2.85 b ¹	4.62 a	4.15 a	3.87 a
	C2	3.44 a	3.79 b	3.96 ab	3.73 a
	O1	2.55 b	2.54 c	4.28 a	3.12 b
	O2	-	1.80 d	3.57 b	2.69 c
	Mean over the sites	2.95	3.19	3.99	
NT (number/m ²)	C1	577 a	553 b	640 a	590 a
	C2	498 b	604 a	531 b	545 b
	O1	417 c	446 c	458 c	441 c
	O2	-	392 d	453 c	423 d
	Mean over the sites	498	499.6	521.3	
TGW(g)	C1	39.7 c	48.0 a	46.6 c	44.8 b
	C2	42.3 b	46.1 b	47.1 c	45.1 ab
	O1	45.7 a	46.1 b	48.7 b	46.8 a
	O2	-	34.5 c	50.9 a	42.70 c
	Mean over the sites	42.61	43.69	48.32	
NK (number/tiller)	C1	12.6 b	17.6 a	14.2 b	14.9 a
	C2	16.5 a	13.7 b	15.8 b	15.3 a
	O1	13.7 b	12.5 b	19.2 a	15.1 a
	O2	-	13.3 b	15.7 b	14.5 a
	Mean over the sites	14.24	14.27	16.28	

¹ Mean values in each comparison between sites within a year with no letter in common are significantly different at $p < 0.05$ according to the least significant difference (LSD) for sites from Anova

Variance components

The partitioning of variance components for GY and yield components indicated that environmental components (year, farming system, site within each farming system and their interactions) were the largest sources of variation for grain yield and yield components (Table 2.4, and for the significances of the terms in analysis of variance see Table S2).

Variance components for genotype and interactions with genotype were relatively small compared to those caused by environmental effects (year, site). Comparatively larger variation due to genotype and interactions (Genotype \times Year, Genotype \times Year \times Site) were found for TGW compared to other yield components. Site effects within farming systems were not very consistent across years so that variability among the four sites was attributed mostly to Year \times Farming system and Year \times Site interactions, while the variance components associated with site effects per se (over years and varieties, within farming systems) was small (or even estimated as negative), relative to effects involving the year differences, differences and interactions with farming system, the overall variety differences, and, especially for number of kernels per tiller, the residual variance.

Table 2.4 Partitioning (%) of variance components for grain yield (GY), number of tillers (NT), thousand grain weight (TGW) and number of kernels per tiller (NK) in two organic (O1 and O2) and two conventional (C1 and C2) sites in 2006-2008. 'Site' refers to Farming system/site, so in the statistical model sites were nested within farming systems. Full replicates (the complete blocks) were nested within years and sites

Component	GY	NT	TGW	NK
Year	4.7	0.0	5.1	1.1
Farming system (conventional versus organic)	16.4	50.2	0.0	0.0
Site	0.0	0.2	0.0	0.0
Year \times Farming system	29.9	0.0	18.6	1.6
Year \times Site	12.0	8.5	43.6	24.4
Genotype	10.4	11.5	11.9	11.6
Genotype \times Year	4.2	1.0	5.8	1.2
Genotype \times Farming system	0.4	0.1	0.0	0.2
Genotype \times Farming system \times Year	0.0	4.4	0.1	2.8
Genotype \times Site	0.0	0.0	0.0	0.0
Genotype \times Year \times Site	1.0	2.2	6.4	2.6
(Year \times Site)/Replication	9.9	2.6	0.4	13.6
Residual	11.1	19.2	8.0	40.9

Heritability

The estimates of the variance components for GY and yield components of each site indicated that in most of the cases the heritability was lower under

organic than under conventional conditions (Table 2.5). Heritability was always the lowest under the conditions of the organic farmer's field O2, due to the large sizes of $G \times Y$ interaction and residual variance under organic conditions. For most environments the heritability of yield components was lower than for grain yield with exception of TGW in O1 and C2. The lack of consistent differences between varieties across years in the O2 field caused a negative value for genetic variance and lead to an estimate of the heritability of 0, see Table 2.5.

Table 2.5 Estimates of broad sense heritability (h^2 , %) and ratio of the genotypic variance and the variance of genotype \times year interaction (V_{gy}/V_g) for grain yield (GY), number of tillers (NT), number of kernels per tiller (NK) and thousand grain weight (TGW) of ten varieties grown at two organic sites (O1 and O2) and at two conventional sites (C1 and C2), 2006-2008

Traits	Growing conditions							
	C1 h^2 (%)	C2	O1	O2	C1 V_{gy}/V_g	C2	O1	O2
GY	70	61	45	24	0.5	0.4	0.4	1.5
NT	40	50	28	18	0.6	0.1	2.0	3.0
TGW	46	73	80	0	1.5	0.4	0.3	n.e. ¹
NK	27	37	31	12	1.1	0.3	0.4	1.4

¹ n.e. = not estimated

Comparison of grain yield and yield components per variety

Under conventional conditions there was group of six varieties: 'Anni', 'Abava', 'Annabell', 'Inari', 'Ansis' and 'Rubiola' that had significantly higher yields (GY) than the other varieties (Table 2.6). These varieties were also the highest yielding under organic conditions with exception of 'Annabell'. Under organic conditions all varieties yielded lower than under conventional conditions. 'Annabell' had the largest yield differences compared to other varieties between both farming systems. Under both conventional and organic growing conditions 'Latvijas vietejie' was the lowest yielding variety.

For all varieties, NT was lower under organic growing conditions than under conventional conditions. Under conventional and organic conditions 'Annabell' had the highest NT but this was at the cost of the TGW. The highest TGW was observed for 'Latvijas vietejie' but that variety had low NT and NK under conventional and organic conditions. Under conventional conditions

NK was highest for 'Inari' but under organic conditions it was highest for 'Ansis'.

Table 2.6 Average grain yield (GY t ha⁻¹), number of tillers (NT, number per m²), thousand grain weight (TGW, g) and number of kernels per tillers (NK) of ten varieties grown under organic (O) and conventional sites (C), 2006-2008

Variety	GY		NT		TGW		NK	
	C	O	C	O	C	O	C	O
Anni	4.28a ¹	3.29a	637b	461bc	43.5d	44.4cf	16abcd	16bc
Abava	4.24a	3.28a	551de	447bc	46.9b	46.9ab	16a	16bc
Annabell	4.14a	2.97bcd	687a	527a	40.4e	41.2f	15cd	14d
Inari	4.12a	3.20ab	541e	442bc	46.7b	47.5a	17a	15bcd
Ansis	4.10a	3.27a	592c	427bcd	43.3d	43.8de	16abcd	17a
Rubiola	4.00a	3.07abc	558cde	424bcd	45.7bc	45.9bc	16abcd	16bc
Dziugiai	3.63b	2.89cd	543e	420cd	43.5d	42.8e	16abcd	16ab
Idumeja	3.35c	2.78de	584cd	411cd	45.2c	46.6ab	13e	14bcd
Primus	3.25c	2.55e	492f	391d	46.1bc	46.0b	14d	14cd
Latvijas	2.90d	2.19f	502f	388d	48.6a	46.9a	12e	12e

¹ Mean values with no letter in common within each organic or conventional site are significantly different at $p < 0.05$

To get more insight in how yield components influence the yield under conventional and organic conditions, we analysed the phenotypic correlations between GY and yield components within both conventional and within both organic sites across all varieties over the years. Differences between the sets of varieties included in the correlation were obtained. Analysing the correlative relationships of the whole set of varieties, NT and NK positively correlated with GY, but TGW had a positive correlation with GY at O2 only in 2007. If old varieties 'Latvijas vietējie' and 'Primus' were excluded from the analysis NT had a positive correlation with GY in 2006 and 2007 in all growing sites, while in 2008, TGW and NK had a positive correlation with GY.

Variety ranking

Spearman's ranks correlation coefficients were calculated to determine if the differences in yield between different conventional and organic growing sites resulted in changes in rank for varieties. All sites showed a significant and

positive rank correlation for yield indicating that generally ranks between sites are retained, with $R_s = 0.53$ to $R_s = 0.90$ (Figure 2.2). 'Anni' and 'Abava' showed high top position ranks according to the average GY in both conventional C1 and C2 and organic O1 sites (Figure 2.2 A, B, D). However, the ranking of some individual varieties for GY under conventional sites differed from that under the organic ones. 'Ansis' took higher ranking position in conventional medium input C1 and also in organic O1 and O2 sites compared to conventional high input C2 site (Figure 2.2 A, B, C). 'Annabell' showed an opposite tendency with a lower rank under organic than under conventional conditions. The smallest changes in varieties ranking were from medium input conventional site C1 to organic O1 site (Figure 2.2 D). The lowest correlation was obtained between both organic sites ($R_s = 0.53$). The ranking of some individual varieties changed considerably between both organic sites (Figure 2.2 F). 'Ansis' and 'Rubiola' took a higher ranking position in organic O2 than in O1, whereas 'Abava', 'Inari' and 'Idumeja' ranked lower in O2 than in O1. The overall rank correlation between conventional and organic conditions was high ($R_s = 0.90$). Here 'Anni' and 'Abava' showed the highest ranks according to the average GY under conventional and organic conditions (Figure 2.2 G). Landrace 'Latvijas vietejie' ranked lowest at all sites.

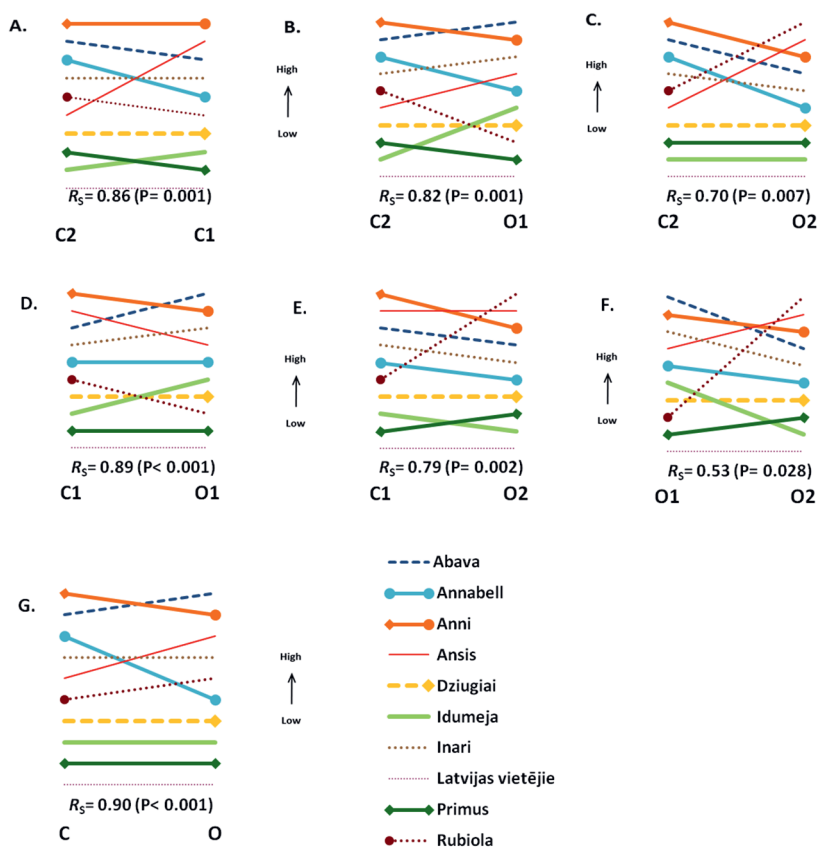


Figure 2.2 The ranking order of varieties with respect to their mean yield (averaged over 2006-2008) between the all site combinations: C2-high input conventional, C1-medium high-input conventional; O1-organic institute site; O2-farmer's field site

Stability of genotype performance

Stability parameters of grain yield of the 10 tested spring barley genotypes evaluated in six conventional and five organic environments (year and site combination) (Table 2.7) are presented.

We are interested in the varieties which can provide a yield level higher than the mean of all tested varieties and are stable according to the regression coefficient (b close to 1) and with the smallest possible deviation from the regression (s^2d close to 0). Our trials showed that variety 'Anni' was high yielding and stable according to the regression coefficient ($b = 1$) and according to the deviation from the regression (s^2d) under both organic and conventional growing conditions (Table 2.7). This variety is therefore suggested to have a general adaptation under both conditions. 'Abava', 'Inari' and 'Ansis' displayed above average yield under conventional and organic conditions, but according to the regression coefficient, 'Ansis' was responsive to more favourable conditions under conventional management ($b = 1.41$), while 'Abava' ($b = 1.21$) and 'Inari' ($b = 1.33$) were responsive to more favourable conditions under organic management. 'Rubiola' had an average mean performance under organic and conventional environments and was stable according to the b -value. 'Idumeja' and 'Dziugiai' also had a regression coefficient (b) close to 1 under both conditions, but exhibited below average yield under conventional, while under organic conditions were close to the average yield level, and therefore suited better organic environments. The landrace 'Latvijas vietejie' and the old variety 'Primus' had a slope (b) close to 1, but low mean performance under organic and conventional conditions and they are considered to be poorly suited to both conditions. Under conventional conditions 'Annabell' exhibited above average yield and had a regression coefficient (b) close to 1, but the deviation from the regression (s^2d) was the largest and coefficient of determination was the lowest ($R^2 = 0.49$, Table 2.7). Under organic conditions 'Annabell' yielded not in the top group with a low value for the deviation from the regression ($s^2d = 0.06$, $R^2 = 0.84$, Table 2.7). The regression coefficient for the slope of the regression line was the lowest of all varieties ($b = 0.54$), indicating that it does not seem to be an adaptable variety under organic conditions.

Table 2.7 Comparison of yield stability parameters: the regression coefficient for the slope of the regression line (b), and variance due to deviation from regression (s^2d) and determination coefficient (R^2) in Finlay-Wilkinson regression of ten spring barley varieties under organic and conventional conditions in Priekuļi 2006-2008

Variety	Conventional				Organic				All environments			
	Mean yield (t ha ⁻¹)	b	s ² d	R ²	Mean yield (t ha ⁻¹)	b	s ² d	R ²	Mean yield (t ha ⁻¹)	b	s ² d	R ²
Anni	4.28* ¹	1.07	0.04	0.93	3.29*	1.12	0.08	0.95	3.82*	1.12	0.04	0.96
Abava	4.24*	1.18	0.06	0.92	3.28*	1.21	0.04	0.98	3.80*	1.18	0.04	0.97
Ansis	4.10*	1.41	0.01	0.99	3.27*	1.01	0.14	0.90	3.72*	1.10	0.08	0.93
Inari	4.12*	0.71	0.08	0.74	3.20*	1.33* ²	0.01	0.99	3.70*	1.12	0.09	0.92
Annabel	4.14*	0.85	0.34	0.49	2.97	0.54*	0.06	0.84	3.60*	0.83	0.27	0.68
Rubiola	4.00	1.24	0.06	0.92	3.07	0.92	0.12	0.90	3.58*	1.04	0.08	0.92
Dziugia	3.63	0.76	0.03	0.90	2.89	0.87	0.04	0.96	3.29	0.85	0.03	0.68
Idumeja	3.35*	0.80	0.10	0.75	2.78	1.00	0.14	0.90	3.09*	0.86	0.11	0.86
Primus	3.25*	1.11	0.10	0.85	2.55*	0.99	0.13	0.90	2.93*	0.98	0.10	0.89
Latvijas vietejie	2.90*	0.88	0.05	0.89	2.19*	1.00	0.01	0.99	2.58*	0.93	0.03	0.96
Environmental	1				2.95	1			3.41	1		

*¹ Mean values of varieties within this column are significantly different from environmental mean at $p < 0.05$

*² values within this column are significantly different from 1 at $p < 0.05$

2.4 Discussion and conclusions

Do varieties and their ranking differ in yield under organic versus conventional conditions?

In our trials grain yield of all tested varieties under organic conditions was generally lower than under conventional conditions. The main factor for this was most likely the lower level of fertilisation in both organic fields compared to conventional management, and in addition the high weed pressure in the organic farmer's field, as is often the case under organic management (e.g. Wolfe et al., 2008). In 2008 when the weather conditions in the second half of the vegetation period promoted high yield formation, the organic sites O1 and O2 yielded as well as the conventional ones. This has also been reported in other studies for wheat (Ryan et al., 2004; Lueck et al., 2006; Murphy et al., 2007) and for lentil (Vlachostergios and Roupakias, 2008). The largest changes of the variety ranking were between both organic conditions, which

could be partially explained by differences in the weed management. In organic farms growing conditions may be very variable. Practical experience showed that it is not always favourable to perform harrowing, due to metrological conditions; the crop may be damaged considerably if the soil is too wet or when harrowing is followed by heavy rain. There are differences not only in use of harrowing, but also in type of fertiliser (green manure, stable manure), crop rotation, etc. For that reason and because the organic production area is comparatively small, the choice for resilient varieties appropriate for most of organic farms, which might be in the range from O1 (fairly stable and comparatively high yield level) till O2 with an unstable yield performance between the years, is needed.

Higher positive correlation between conventional medium input and both organic GY, compared to conventional high input or between both organic GY, suggests that results of GY under conventional medium input conditions provide a better prediction for the average variety performance under organic conditions (see Table 2.1). However there were some notable exceptions on the level of individual varieties. The results showed that not all varieties took the same ranking position between conventional C1 and organic O1 and O2 conditions and particularly between the two organic sites, which could be explained by differences in management practices. For example, 'Annabell', which is the shortest variety, may have suffered from high weed pressure and the low nutrient availability under both organic conditions. In 2008, a high infection level of netblotch (*Pyrenophora teres*) particularly affected the yield of 'Annabell' (data not shown). Overall, 'Annabell' ranked lower under organic than under conventional conditions. This suggests that some high-input varieties could be more sensitive to abiotic and biotic stress than others, making them less suitable for organic farming systems. These differences in ranking make additional testing under organic conditions for traits such as weed competitiveness and disease resistance necessary to identify varieties with relatively high performance under low-input management. This is consistent with results obtained by Przystalowski et al. (2008) who analysed datasets of cereals under organic and non-organic sites in six European countries. They concluded that despite an overall high genetic correlation for yield, and other traits such as plant height, there were exceptions on individual variety ranking level in both directions that could be relevant for the selection

process. A variety that had a medium yield under conventional conditions could perform among the top under organic conditions or those that perform best under conventional condition might be moderate under organic conditions. In order not to miss potentially valuable genotypes for organic farming systems these authors advised combining information from both organic and non-organic trials.

Do varieties differ in yield stability under organic conditions compared to conventional conditions?

Yield stability across years is one of the most important breeding objectives for organic, low input conditions where pesticides and (high levels of) fertilisers are not available to stabilize yield (Ceccarelli, 1994; Lammerts van Bueren et al., 2008; Østergård et al., 2005; Przystalski et al., 2008). In our trials the grain yields (GY) between farming systems and sites within farming systems were more variable across years than between genotypes due to a larger Year \times Farming system and Year \times Site effect on GY as is often reported by other authors (e.g. Wolfe et al., 2008; Przystalski et al., 2008). To improve yield stability Bernardo (2002) suggested that breeders must select their lines on the basis of the mean yield performance and the slopes of the Finlay-Wilkinson regression (b) of varieties across all environments. Ozgen (1994) cited by Ulker et al. (2006) considered that a stable genotype should have above average grain yield and a regression coefficient (b) close to 1.0. Becker et al. (1982) regarded small deviations from regression to be the most appropriate criterion for measuring stability in an agronomic sense because this parameter measures the predictability of the genotypic reaction across varying environments.

Our trials showed that landraces and old varieties were the lowest yielding over organic and conventional environments; they did not meet our expectations for good yield under organic conditions. This corresponds with Bernardo (2002), who also pointed out that genotypes which exhibit stability across environments tend to have a low performance. As organic farmers are interested not only in stability, but also in high yield, such varieties with low adaptability should be less suitable for organic conditions (Pswarayi et al., 2008). Most of the other varieties that were included in our study responded better to favourable organic conditions, and could be suitable for organic

management. The high input type variety 'Annabell' had the largest decrease in yield under organic compared to its performance under conventional conditions, and showed to be sensitive to irregular growing conditions. For example, in 2008 there was a dry spell from the end of May until the beginning of June for a period of two weeks, which was followed by much cooler and rainy weather until the end of June and into July causing additional late tillering, especially for 'Annabell'. The effect of this late tillering in the middle of the growing period caused small grains on the secondary shoots, which resulted in low TGW and finally in a low yield of 'Annabell' under organic growing conditions in 2008 (data not shown). The situation was similar under both conventional conditions in 2008 and might explain the large deviation from the regression (s2d) under conventional conditions. The low value for the slope of the regression line under organic conditions indicates that in the years with a higher overall yield level Annabell did not profit as much as other cultivars (possible reasons might be infection with netblotch and drought) and its adaptability was lower than for other cultivars, therefore we doubt its suitability for organic farming. This indicates that some modern varieties were more unpredictable to changes in the environment than the old landraces and varieties developed before the 1980s. Also Ceccarelli (1996) and Pswarayi et al. (2008) argued from their research that modern genotypes are more adapted to stress-free, high yielding environments, and will not always give good results under unfavourable conditions.

Within this set of varieties, suitability to organic farming seems to be associated mainly with time of release (see Table 2.1). The landrace 'Latvijas vietejie' and the old variety 'Primus' which were grown more than 100 years ago and currently are not in production, are very tall, with a good and rapid soil cover, resulting in good weed competitiveness. But the consistently low yield level make these varieties not suitable for direct growing under organic farming. However, the old genotypes can be useful in breeding for organic farming if yield potential can be improved by crossing with newer material. Stable and high yielding varieties differed in time of release (see Table 2.1) and in plant height at the beginning of stem elongation stage and at maturity as well as days to heading and days to maturity. For example 'Anni', 'Inari', 'Idumeja' are short to medium short straw varieties, while 'Rubiola' and 'Dziugiai' are tall. Analysing the correlative relationships between yield and

previously mentioned morphological and biological traits showed that grain yield was mainly negatively associated with plant height at maturity and days to heading and days to maturity in conventional and also in both organic growing sites, which is in contrast to the results found by Murphy et al. (2008) and Reid et al. (2009). This negative correlation between plant height and yield could be explained by the way the present set of varieties was composed including old varieties ('Latvijas vietejie' and 'Primus') that were very tall and low yielding. In an analysis without the two old varieties then was no significant correlation between yield and plant height at maturity time.

The current study suggests that the varieties differ in yield potential. Modern varieties developed after the 1990s have higher yield potential compared to varieties released in the first half of the 20th century and before. Mason et al., (2008) and Calderini and Slafer (1999) reported that modern varieties may outperform older ones in poor environments even despite their limited stability. Our trials suggest that the high input type variety 'Annabell', which has a high tillering capacity can produce a good yield in low yielding environments (e.g. in organic farmer's field O2 in 2007), but only if during the first part of the vegetation period the conditions are favourable for tillering. One can conclude that modern barley breeding can in principle provide high-yielding varieties for organic growing conditions, but one cannot state that those varieties will always be the most stable under variable organic conditions. Their suitability for organic farming should be verified in tests under organic growing conditions, as discussed in the previous paragraph.

Is the heritability lower under organic growing conditions than under conventional conditions?

Low heritability of yield traits in poor or stressful environments is one of the arguments for conducting selection in environments at optimal plant growth conditions (Rajaram et al., 2006). Our data confirm that heritability for barley grain yield in organic, low-yielding environments was indeed lower than in more optimally controlled conventionally managed environments (see also Atlin and Frey 1990; Ceccarelli, 1994, 1996). This suggests that selection for yield for organic conditions could be successfully carried out under conventionally managed conditions. In our trials the heritability for yield components was very low in the organic farmer's field O2 compared to the

other sites; this was mainly due to a high proportion of V_{gy} interaction in the total phenotypic variance. In the better managed organic site O1 the heritability for yield and its components was higher than in the farmer's field O2 but still lower than under conventional conditions (with exception of TGW and NK when compared to C1).

Banzinger and Cooper (2001) and also Löschenberger et al. (2008) suggested that optimally managed on-station experimental trials may be used for assessing qualitative traits which are highly heritable, but that these would not be useful for quantitative traits (yield and yield components) which are more affected by genotype x environment interaction. Our results indicated that yield components had a lower heritability than GY itself, which is consistent with the conclusions drawn by Alexander et al. (1984), Aycicek and Yildirim (2006) and Zecevic et al. (2010) for wheat experiments, and by Bezant et al. (1997) and Yin et al. (2002) for barley QTL studies. Yin and Struik (2008) suggested that this is because yield depends on various interactions and compensating mechanisms from its components. Effect of a QTL can be small on individual components but can altogether result in a significant impact on grain yield itself. In our experiments on yield components, results showed that genotypes can have different combinations of traits to ensure a good yield level. For example, for 'Anni' the high and stable yield performance under conventional and organic conditions was based on a combination of high NT and NK, while for 'Abava' the high yield was based on a combination of high NT and TGW. For practical breeding, harvesting and measuring the yield is easier and less laborious than determining the TKW and counting tillers per plant or kernels per tiller, so that a higher heritability for yield is a favourable outcome for the breeder in terms of labour.

Perspectives for selection strategies for barley adapted to organic farming systems

The question of what is the more suitable selection environment for varieties adapted to organic farming systems is raised not only for technical breeding reasons but also for economic reasons due to the costs incurred due to extra selection fields.

Varieties in organic farming should have an adaptability to variable, organically managed and mostly low-input conditions and direct selection in

organic conditions is recommended by many breeders (e.g. Wolfe et al., 2008; Przystalski et al., 2008; Murphy et al., 2008; Reid et al., 2009). Reid et al. (2009) demonstrated for spring wheat, that selection of genotypes for organic farming under conventional conditions does not result in the same genotypes being selected for each system for all traits. They believed that selection of genotypes for organic production systems should be done under organic conditions. Following that reasoning, one could argue that in spite of relatively low heritability the organic O1 site with a fairly stable and comparatively high yield level could be an appropriate environment for selection of genotypes for organic farming. Unstable farm conditions as in O2 are not suitable to select barley for organic farming; more replicates and repetitions across multiple organic farms could be useful and increase heritability, but it would also significantly increase costs.

Another strategy departing from a focus on how conventional breeding can also serve organic farming could be to choose the most suitable conventional conditions. In our trials, the correlations for GY between sites showed that O1 and O2 had comparatively higher correlations with conventional medium input C1 conditions than with the high input site C2 and between both organic sites. Also similar ranking of varieties between these sites for GY, as well as the higher heritability for yield in the medium-input site C1, can lead to the conclusion that it could be possible to conduct a sufficiently effective selection for GY for organic farming purposes under the conventional medium input C1 conditions. However, our trials also showed that to ensure yield stability under organic conditions additional testing of genotypes under various organic conditions is necessary. Due to different management practices and different levels of soil fertility among organic farms, such tests will help to make decisions which genotype is stable for GY and the most appropriate for cultivation under organic conditions.

Acknowledgments

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Supplementary material

Table S1 Correlation (r) of grain yield (GY), number of productive tillers (NT), thousand grain weight (TGW) and number of kernels per productive tiller (NK) between two organic locations (O1 and O2) and two conventional locations (C1 and C2)

Year	GY			NT			TGW			NK		
	C1	C2	O1	C1	C2	O1	C1	C2	O1	C1	C2	O1
2006	C2	0.95**1		0.89**			0.57			0.81**		
	O1	0.81**	0.78**	0.55	0.52		0.60	0.74*		0.72*	0.63	
2007	C2	0.91**		0.83**			0.91**			0.37		
	O1	0.88**	0.81**	0.57	0.56		0.45	0.73*		-0.03	-0.13	
	O2	0.88**	0.81**	0.84**	0.46	0.81**	0.23	-0.11	-0.51	0.33	0.06	0.75*
2008	C2	0.88**		0.28			0.95**			0.56		
	O1	0.74*	0.64*	0.08	0.73*		0.83**	0.87**		0.42	0.91**	
	O2	0.89**	0.91**	0.56	0.50	0.59	0.89**	0.89**	0.98**	0.57	0.66*	0.33

1** = P<0.01; * = P<0.05

The highest values for one trait and year marked in bold

Table S2 Analysis of variance *p*-values for the effects of main factors: conventional versus organic management systems, year location and variety, and their interactions for grain yield (GY), number of tillers (NT), thousand grain weight (TGW) and number of kernels per tiller (NK) between two organic locations (O1 and O2) and two conventional locations (C1 and C2), 2006 -2008 randomised complete block design per location in every year. The Anova table for GY is shown below as an example of the analysis.

Source of variation	GY	NT	TGW	NK
Year	<0.001	<0.001	<0.001	<0.001
Farming system (conv vrs org)	<.001	<.001	0.200	0.308
Location	0.007	<0.001	<0.001	0.288
Year × Farming system	<0.001	0.149	<.001	<0.001
Year × Location	0.004	<0.001	<.001	<0.001
Genotype	<0.001	<0.001	<0.001	<0.001
Genotype × Year	<0.001	<0.001	<0.001	0.002
Genotype × Farming system	0.006	<0.001	<0.001	0.041
Genotype × Farming system × Year	0.048	<.001	<0.001	0.034
Genotype × Location	0.080	0.096	<0.001	0.190
Genotype × Year × Location	0.032	0.043	<0.001	0.097

Analyses of variance for grain yield (GY)

Source of variation	df	SS	MS	F	Sig.
Year	2	115.4425	57.7213	37.45	<0.001
Farming system (conv vrs org)	1	106.4106	106.4106	69.03	<0.001
Year × Farming system	2	77.5537	38.7768	25.16	<0.001
Location	2	17.8821	8.9411	5.8	0.007
Year × Location	3	24.6377	8.2126	5.33	0.004
Residual environment, replication, *Units* stratum	33	50.8678	1.5414	9.95	
Genotype	9	70.7182	7.8576	50.73	<.001
Genotype × Year	18	19.833	1.1018	7.11	<.001
Genotype × Farming system	9	3.6458	0.4051	2.62	0.006
Genotype × Farming system × Year	18	4.5987	0.2555	1.65	0.048
Genotype × Location	18	4.2492	0.2361	1.52	0.08
Genotype × Year × Location	27	6.7155	0.2487	1.61	0.032
Residual	297	45.9992	0.1549		
Total	439	508.7813			

Chapter 3

Performance of various spring barley varieties for traits contributing to weed suppressive ability under organic and conventional growing conditions

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Abstract

Plant traits contributing to weed suppressive ability, especially at the beginning of the vegetation period, are important in cereal breeding for organic agriculture to reduce labour in managing weeds without herbicides. We analysed different characteristics contributing to weed suppressive ability in ten different spring barley (*Hordeum vulgare* L.) varieties under two organic and two conventional farming locations. The varieties were divided into four groups based on time of release and adaptation to growing environments: old, low input varieties and landraces, medium old varieties with good adaptation to unfavorable conditions, and modern varieties for low-input conditions, and modern varieties for high-input conditions. We found that traits such as productive tillering ability, rapid development and tall plants at the beginning of stem elongation provided high crop ground cover. Therefore, the genotypes combining the planophile growth habit at tillering with fast early development could be considered for organic farming. The selection of genotypes for traits contributing to weed suppressive ability may take place in conventional or organic conditions.

Key words:

conventional agriculture, organic agriculture, weed suppression ability, crop ground cover.

3.1 Introduction

In organic agriculture, weeds are considered one of the main problems for farmers to deal with. Because in organic farming systems no herbicides are allowed, it is important to prevent and control weeds by measures at different levels: farm level (crop rotation, intercropping, plant arrangement allowing mechanical weeding), field crop level (optimizing growing conditions, thereby promoting crop competitiveness against weeds), and variety level (competitive plant architecture, rapid juvenile growth, deep rooting system) (Wilson, 1988; Lemerle et al., 1996; Hoad et al., 2005; Bertholdsson, 2011). Crops differ in their competitiveness against weeds, but also within a crop species there is genetic variation in competitiveness (Lammerts van Bueren et al., 2002).

One aspect of competitiveness against weeds- is weed suppressive ability. Weed suppressive ability is the ability of the crop to suppress the growth of weeds, and it contributes to a long-term strategy to reduce the weed seed bank in the soil. The another aspect is the weed tolerance - an ability to tolerate weed competition which can be measured by the ability of the crop to maintain high yields under weedy conditions (Coleman et al.

2001; Worthington and Reberg-Horton, 2013; Andrew et al., 2015; Korres et al., 2016; Mahajan et al., 2020). Weed suppressive ability in cereals is connected with various plant morphological and physiological traits and interactions (Kruepl et al., 2007). This will include strengths in some characteristics compensating for weaknesses in others, e.g, a variety that changes the plant growth habit from planophile to erectophile over the growing season will cover the soil better than a taller variety later on (Hoad et al., 2008).

Certain characteristics are indicated as desirable for organic varieties to improve weed suppression: good tillering ability, rapid early growth, taller plants. These are especially important under organic conditions, where the seed is not chemically treated and emergence can be lower compared to conventional crops. Flag leaf inclination angle and high leaf area index are features that allow the plants to compete with weeds during the second half of the growing season (Hoad et al., 2005).

This study aims to identify how different morphological physiological characteristics of ten contrasting barley varieties related to the weed

suppressive ability perform under organic and conventional farming growing conditions. Which morphological traits related to weed suppressive ability could be used as selection criteria for organic farming? Which environment would be most suitable to carry out selection for these characteristics?

3.2 Materials and methods

This study on the 10 varieties was performed as a preparation for a breeding program for organic farming. We wanted to find out how these varieties perform for the traits related to weed suppressive ability under different organic and conventional growing conditions. The overall goal was to include these varieties for hybridization to obtain breeding material for organic farming.

The data for this research question were collected from the same trial as in Chapter 2, so the set of varieties, growing locations, meteorological conditions, management practice, and experimental design were the same as described in Chapter 2. Here we provide a short summary, see Table 3.1 with the ten varieties.

During the vegetation period, the following measurements and scorings were done: growth habit in the tillering stage (Zadoks scale 25 to 29): 1 - erectophile, 9 - planophile; development speed in tillering stage (GS 29-30): scores (1 - slow, 9 - rapid); plant height at the beginning of stem elongation (GS 30-31); crop ground cover (GS 30-31) (visually estimated plant covered area in the plot, %); length of flag leaf (GS 47-50), cm; width of flag leaf (GS 47-50), cm; plant height at harvest (GS 90), cm; number of productive tillers per m² (GS 80-90), productive tillering capacity was calculated by dividing the number of productive tillers per m² and the number of emerged plants per m².

Data processing was done using GENSTAT 14.0 (2011). The data were analysed using descriptive statistics and Pearson correlation analysis. Analysis of variance (ANOVA) was carried out to analyze the main effect of genotype, year, location, and their interactions on the traits. The following model was used for ANOVA:

$$y = \mu + G + Y + L + Y \times L + G \times Y + G \times L + G \times Y \times L + e, \text{ where}$$

the genotype (G), year (Y), and locations (L) were fixed factors in the model, and error (e) were the random term.

Table 3.1 Description of the ten varieties included in the trials during 2006-2008

Variety	Intensity group		Year of registration or use	Remarks
Primus	old, low-input	OL	1901	very tall plants, medium-planophile early growth habit, late maturing
Latvijas vietējie			landrace ~1800	very tall plants, very late maturing
Dziugiai			1947	very rapid, early development, resistant to acid soil conditions
Idumeja	modern, low input	ML	2000	medium-tall plants, erectophile early plant growth habit, early maturing
Rubiola			2007	bred for organic farming
Inari	modern, high input	MH	1994	medium plant height, medium early maturing
Annabell			1999	currently the most popular variety in conventional farming
Ansis			1995	short plants, high-input type
Abava	medium old, with good adaptability	MA	1978	low-input type: good yield under poor growing conditions
Anni			1993	stable yield under low-input conditions, good stress resistance

3.3 Results and Discussion

Traits related to the weed suppressive ability were influenced significantly ($p < 0.01$) by all main factors (genotype, year, growing locations) and two-way (genotype, year), as well as three-way (genotype, year, location), interactions. (Additional file 1). In the growing locations the differences in tillering capacity, crop ground cover, plant height at the stem elongation stage, and height at maturity were observed (Table 3.2). In the organic location O2 (farmer's field) the number of productive tillers, crop ground cover, and plant height at the stem elongation stage and at harvest were significantly lower ($p < 0.001$) than in other growing locations. Early planophile growth habit during tillering stage was observed for the modern, high-input group varieties Ansis and Annabell (Addition file 2).

Table 3.2 Mean values of traits associated with weed suppressive ability at two organic locations (O1 and O2) and two conventional locations (C1 and C2), from 2006 to 2008, among ten spring barley varieties.

Traits		Environment			
		O1	O2	C1	C2
Emergence, plants per m ²	mean value	343.1	276.8	319.8	331.9
	differences **	O2	O1, C1, C2	O2	O2
Growth habit in tillering stage (1 - erect, 9 - planophile)	mean value	4.4	4.3	4.6	4.5
	differences **	-	-	-	-
Productive tillering capacity	mean value	1.3	1.6	1.8	1.7
	differences **	O2, C1, C2	O1, C1	O1	O1
Development speed at tillering stage (1 - slow, 9 - rapid)	mean value	5.3	5.0	5.2	5.2
	differences **	-	-	-	-
Plant height at the beginning of stem elongation (cm)	mean value	26.8	21.7	29.2	28.5
	differences **	O2	O1, C1, C2	O2	O2
Crop ground cover (%)	mean value	65.1	60.0	62.2	57.3
	differences **	O2, C2	O1, C1	O2	O1
Length of flag leaf (cm)	mean value	10.4	9.8	10.4	10.3
	differences **	-	-	-	-
Width of flag leaf (cm)	mean value	0.75	0.75	0.79	0.78
	differences **	-	-	-	-
Number of productive tillers per m ²	mean value	440.6	365.1	587.9	549.8
	differences **	O2, C1, C2	O1, C1, C2	O1, O2	O1, O2
Plant height at harvest (cm)	mean value	74.7	64.4	74.7	76.5
	differences **	O2	O1, C1, C2	O2	O2

** differences are significant at the 0.01 level

This plant growth habit is considered as an important feature affecting weed suppressive ability (Hoad et al., 2008). Because, the early planophile growth habit has several advantages in comparison with the erectophile form and it results in a higher light interception, and in more effective shading of weeds. Hoad et al. (2008) suggested that, to a certain extent, the planophile growth habit in combination with large leaves and good crop establishment allows the plant to compensate for its length and therefore it is more suitable for short straw varieties in the first part of the vegetation period in the circumstances

with a large proportion of weeds. However, our findings showed a different tendency. There was a negative correlation between the growth habit at tillering stage and crop ground cover (Table 3.3) in all growing locations, indicating that the early planophile or expanded plant growth habit at tillering stage was associated with low crop ground cover. We observed that the varieties in our trial with an early planophile growth habit developed slowly during the tillering stage. The correlation analysis showed that between the growth habit at tillering stage and the development speed at tillering stage, there was a moderately close negative correlation ($r = -0.682$, $p < 0.01$). In our experiment, the modern, high input varieties Ansis and Annabell, and Anni from the medium old group had a planophile plant growth habit, and had developed more slowly than other varieties at the early tillering stage. An erectophile growth habit and fast development speed at the tillering stage were observed for the old, low-input variety Dziugiai and the modern, low-input varieties Idumeja and Rubiola (Additional file 3.2). We therefore suppose that 'Ansis', 'Annabell', and 'Anni' may compete less successfully with weeds than varieties with an erectophile growth habit combined with a fast development speed.

Table 3.3 Pearson correlation coefficients between crop ground cover and traits associated with among of ten spring barley varieties, in two organic (O1 and O2) and two conventional (C1 and C2) cultivation sites, from 2006-2008.

Trait	Crop ground cover			
	O1	O2	C1	C2
Emergence	0.003	-0.142	0.057	-0.081
Productive tillering capacity	-0.127	0.021	-0.139	-0.106
Growth habit in tillering stage	-0.359**	-0.241	-0.289	-0.320**
Development speed in tillering stage	0.667**	0.408**	0.545**	0.618**
Plant height at the beginning of stem elongation stage	0.602**	0.435**	0.577**	0.792**

* correlation is significant at 0.05 level; ** correlation is significant at the 0.01 level

A positive correlation between growth habit at tillering, plant height at the beginning of stem elongation, and the crop ground cover was observed (Table 3.3). Therefore, we assume that the varieties with rapid early development and those with taller plants at the stem elongation stage could achieve higher crop ground cover at this stage and finally could compete with weeds better than those that develop slowly and have short plants at early

development stages. However, to confirm this assumption, the evaluation of the genotypes for weed suppressive ability should be done in several environments with variable weed infestation levels.

Lower field emergence and shorter plants (averaged across three years) resulted in lower barley crop ground cover at early growing stages in farmer's location O2 than in O1. It shows that depending on the crop establishment, the average canopy height at the stem elongation stage may vary between the locations. The lowest plant height at the beginning of stem elongation was in location O2 (Table 3.2). The tallest plants in this environment were found for varieties with the fastest rate of development: 'Dziugiai' from the old, low-input group, 'Abava' from medium old group, and 'Idumeja' and 'Rubiola', representing the modern low-input group (Additional file 3.2). The shortest ones were the modern, high-input varieties Annabell and Ansis which also had the slowest development at the tillering stage. The plant height at the beginning of stem elongation related positively ($p < 0.001$) to the yield in location O2, unlike in the other growing conditions (Table 3.4).

Table 3.4 Pearson correlations between yield and traits associated with weed suppressive ability among ten spring barley varieties in two organic (O1 and O2) and two conventional (C1 and C2) locations, from 2006-2008.

Traits	Environment			
	O1	O2	C1	C2
Productive tillering capacity	0.406**	0.653**	-0.021	0.432**
Growth habit in tillering stage	-0.093	0.260	0.289	0.241
Development speed in tillering stage	-0.198*	-0.089	-0.077	-0.200
Plant height at the beginning of stem elongation stage	-0.381**	0.418**	-0.341**	0.164
Crop ground cover	0.113	0.086	0.259	0.201
Length of flag leaf	-0.463**	0.218*	-0.171	-0.224*
Width of flag leaf	-0.470**	0.362**	-0.005	-0.179
Plant height before harvest	-0.008	0.687**	-0.064	-0.306**
Number of productive tillers per m ²	0.428**	0.765**	0.147	0.453**

* significant at 0.05 level; ** correlation is significant at the 0.01 level

In addition, assessing the correlation of plant height at the beginning of stem elongation between the environments, it was found that the values obtained in O1 and O2 did not strongly correlate with each other (Table 3.5). The reason may be the differences between both organic locations in the presence of weeds and nutrient availability. It indicates that different organic

environments require different approaches to the choice of varieties. In the environments with high weed incidence, such as the O2 environment, where perennial weeds predominate, priority should be given to the genotypes with fast early development speed and tall plant height at the stem elongation stage. A close positive correlation was observed between values of plant height at the stem elongation stage in the O1 and conventional C1 locations. Therefore, the plant height at the stem elongation stage may be a reliable trait to select for in a conventional location with a medium input level when aiming at varieties for optimally managed organic conditions

Table 3.5 Pearson correlation coefficients between traits associated with competitiveness against weeds among ten spring barley varieties in two organic (O1, O2) and two conventional (C1, C2) environments, from 2006-2008.

Growing environment	O1			O2	
	O2	C1	C2	C1	C2
Tillering capacity	0.398**	0.219	0.09	0.237**	-0.001
Growth habit in tillering stage	0.825**	0.847**	0.815**	0.798**	0.883**
Development speed in tillering stage	0.821**	0.889**	0.908**	0.861**	0.812**
Plant height at the beginning of stem elongation	0.116	0.739**	0.574**	0.047	0.213
Length of flag leaf	0.304**	0.737**	0.451**	0.285**	0.377**
Width of flag leaf	0.089	0.665**	0.533**	0.131	0.251**
Crop ground cover	0.367**	0.660**	0.285	0.468**	0.236**
Plant height before harvest	0.614**	0.772**	0.892**	0.347**	0.581**
Number of productive tillers	0.348**	0.227*	0.327**	0.266**	0.359**

** significant at 0.01 level

However, the correlation between organic location O2 and all other locations for the values of the plant height at the stem elongations stage was weak, indicating that specifically adapted varieties to this environment should be chosen.

For O2 environment, the varieties with the tallest plant height at stem elongations, such as Dzuigiai and Idumeja could be the most suitable (Additional file 3.2).

According to Hoad et al. (2008) the crop ground cover can be used as a good indicator of shading characteristics. At the beginning of the vegetation period, better crop ground cover was observed for old, low-input varieties: Dzuigiai, Primus, and Latvijas vietējie (Additional file 3.2). Modern, low-input and medium old, with good adaptability varieties better covered the soil than the new modern, high-input short-stem varieties, which covered about half of the research plot at the beginning of stem elongation. Although the crop ground cover had no significant correlation with yield in our trials, it could considerably contribute to the weed suppressive ability at the early growing stages (Table 3.4). A moderate correlation between the values of crop ground cover was observed between organic and conventional locations. The highest crop ground cover in both organic locations was reached by 'Dzuigiai' and 'Abava'. In the weedy location O2 the most weed suppressive variety seems to be Latvijas vietējie because the long and wide leaves at early growing stages ensured high soil coverage in this environment. However, the yield level of 'Latvijas vietējie' was the lowest (Chapter 2).

The length of the flag leaf, the width of the flag leaf, and plant height before harvest play a significant role in shading during the second half of the growing season to increase the suppressiveness of the varieties against late summer weeds. Late summer weeds may hinder the barley harvest. When parts of the weeds get into the crop harvest, they increase their total mass and moisture, creating additional cleaning and drying costs. During ripening, weed seeds spill out and thus contaminate the soil, creating a seed bank in the soil for coming years. Our analysis showed that the length and width of the flag leaf were mainly associated with the growing location and genotype by year interaction ($G \times Y$) ($p < 0.01$). These two leaf parameters positively correlated with each other ($r = 0.763$, $p < 0.01$) and also with the height of the plant ($r = 0.370$ Length of flag leaf, $r = 0.381$ Width of flag leaf, $p < 0.01$). The longest and widest leaves were found for two the old, low-input varieties Latvijas vietējie and Dzuigiai (Additional file 3.2). The plant height of these old, low-input varieties, when they reached the full maturity stage, was the highest, which refers to good weed suppressiveness throughout the vegetation season of those

varieties. A significant, positive correlation between both traits characterizing leaf area and yield was found only in the location O2, whereas in O1 the correlation was negative (Table 3.4).

The plants were shorter under organic than under conventional conditions, especially in location O2, which may create the additional risks in this environment that late summer weeds may become dominant. Therefore, there may be an advantage under these organic farming conditions for taller varieties such as the ones from old, low-input group. However, there are indications in the literature that the old varieties and landraces are not always acceptable for production in organic conditions. As demonstrated by Kokare et al. (2014), landraces have low-yield potential and poor lodging resistance. Nevertheless, the inclusion of these old varieties in the breeding program for organic farming could help to improve weed suppression ability. Our selected modern, low-input and medium-old varieties with good adaptability have a medium tall straw, resulting in better lodging resistance. They also have a higher tillering rate than old, low-input varieties in our trial. The modern low input varieties included in this trial developed faster in the spring and reached the same early plant height as old low input. During the tillering stage, the crop ground cover of modern low input varieties was within the level of the old low input varieties. Bertholdsson et al. (2005) indicate that the morphological and physiological traits, such as flag leaf inclination angle, rapid growth in the early development stage, tillering ability, plant height, etc., negatively correlated with yield. Other studies have also shown that plant height does not sufficiently explain the differences in yield under conventional and organic conditions (Østergård & Jensen, 2004). Our study confirms that the length and width of the flag leaf and plant height before harvest within conventional environments C1 and C2 are negatively related to yield (Table 4). In the organic location O1 there was also a negative correlation between leaf parameters and yield. It could be partly explained by the old low input intensity group varieties with relatively tall plants and low yield potential. They produced lower yields under favorable growing conditions than other varieties (Kokare et al., 2009). Contrastingly, in O2 a positive correlation between yield and plant height was observed (Table 3.4), indicating that tall varieties may improve not only competitive ability with weeds but yield as well.

For the length and the width of flag leaf, and the plant height before harvest, correlations between O1 and O2 were poor (Table 3.5). For organic location O2 there was no clear advantage found to perform selection in organically managed O1 location or conventional location C1 with medium input. The selection result could be less predictable because of the differences in crop rotations and on farm crop management conditions in organic O2 location. Therefore additional testing of candidate varieties can be recommended in various organic farms such as location O2. Higher correlations were observed between organic (O1) and conventional (C1) conditions. This may suggest that the selection of genotypes for organic farms with optimal weed management practice may also be implemented in conventional conditions with medium input level.

In order to confirm our findings, it is necessary to increase the number of varieties in a trial considerably (see Chapter 5). In addition, the evaluation of the genotypes for competitive ability against weeds should be done in several environments with variable weed infestation levels.

3.4 Conclusions

From this trial with a limited set of ten varieties, we learned that in breeding for organic farming, priority should be given to the traits that allow to establish good crop ground cover at early growth stages, especially in environments with low weed management practice. The genotypes with an erectophile plant growth habit, fast early development speed at the tillering stage and tall plants at the beginning of stem elongation stage could be suitable for organic environments.

Our results show that despite a high tillering ability, the genotypes with a planophile growth habit at the tillering stage had a slow development speed and therefore low crop ground cover. Therefore, in the selection of genotypes the planophile growth habit at tillering should be combined with fast early development.

With respect to the question what selection environment is suitable for the selection of genotypes for traits such as growth habit at tillering, fast development at tillering, tall plant height at the beginning of stem elongation, high values for length and width of the flag leaf, and tall plant height at harvest, our trials show that selection for such traits may just as well take place in conventional or organic conditions.

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Supplementary material

Additional file 1 *P* -values (from ANOVA) for barley genotype, year and location main effects, and their interaction (significant effects at a 95% confidence level are marked in bold).

Traits	Genotype (G)	Year(Y)	Location (L)	Genotype × year (G × Y)	Genotype × location (G × L)	Genotype × year × location G × Y × L)
Growth habit in tillering stage (1 - erect, 9 – planophile)	<.001	<.001	<.001	<.001	<.001	<.001
Development speed at tillering stage (1 - slow, 9 – rapid)	<.001	<.001	<.001	<.001	0.010	0.701
Plant height at the beginning of stem elongation (cm)	<.001	<.001	<.001	<.001	<.001	0.070
Crop ground cover (%)	0.292	<.001	<.001	0.001	0.004	0.504
Length of flag leaf (cm)	<.001	0.004	<.001	<.001	0.187	0.367
Width of flag leaf (cm)	<.001	<.001	<.001	<.001	0.448	0.437
Number of productive tillers per m ²	<.001	<.001	<.001	<.001	0.012	<.001
Plant height at harvest (cm)	<.001	<.001	<.001	<.001	0.011	0.169

Additional file 2 Mean values for the traits related to weed suppressive ability of 10 barley varieties grown in two organic (O2, O2) and two conventional (C1, C2) locations

Location	Intensity group/varieties									
	OL		MA			MH			ML	
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
Growth habit at tillering stage										
O1	6	6	2	4	7	5	6	6	2	4
O2	5	5	2	4	6	5	6	6	2	3
C1	5	5	2	4	7	5	7	7	2	3
C2	5	5	2	4	7	4	6	6	2	3
Mean	5	5	2	4	7	5	7	6	2	3
Development speed in the tillering stage										
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
O1	6	6	8	7	3	5	3	4	7	6
O2	5	5	7	7	3	4	3	3	7	5
C1	5	5	8	7	4	5	3	3	7	6
C2	6	6	8	7	4	5	3	4	7	5
Mean	5	6	8	7	4	5	3	3	7	5
Plant height at the beginning of the stem elongation stage										
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
O1	27	27	37	30	21	26	20	21	31	28
O2	21	22	26	23	20	22	18	19	25	24
C1	29	30	40	33	24	28	24	23	33	30
C2	28	30	39	36	26	30	25	25	33	32
Mean	28	30	39	34	25	29	24	24	33	31
Crop ground cover										
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
O1	68	72	77	76	54	61	45	60	73	66
O2	70	65	69	71	57	62	53	44	67	58
C1	68	67	73	69	54	59	55	52	64	61
C2	58	57	76	66	47	60	50	45	60	54
Mean	63	62	74	68	50	59	53	49	62	58
Length of flag leaf										
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
O1	12	10	12	11	10	12	8	9	10	10
O2	13	10	10	11	9	12	8	9	9	9
C1	12	10	12	12	9	12	8	9	11	9

Performance of various spring barley varieties for traits related to weed competitiveness under organic and conventional growing conditions

C2	13	10	11	10	10	12	8	9	10	10
Mean	12	10	11	11	10	12	8	9	10	10
Width of flag leaf										
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
O1	3	3	3	2	3	3	2	2	3	3
O2	4	3	3	3	3	4	3	3	3	3
C1	3	3	3	3	3	3	2	3	3	3
C2	3	3	3	3	3	3	3	3	3	3
Mean	3	3	3	3	3	3	3	3	3	3
Plant height before harvest										
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
O1	92	98	87	77	60	67	59	65	70	72
O2	74	82	70	68	54	61	54	58	61	62
C1	93	100	86	77	62	66	59	64	67	73
C2	96	101	84	78	65	70	63	66	68	73
Mean	94	101	85	77	63	68	61	65	68	73
Number of productive tillers per m ²										
	Latvijas vietējie	Primus	Dziugiai	Abava	Anni	Inari	Annabell	Ansis	Idumeja	Rubiola
O1	396	402	442	452	471	454	525	426	410	432
O2	321	322	351	396	381	372	436	352	373	349
C1	540	513	557	588	653	537	721	619	602	567
C2	463	471	541	515	614	545	669	565	567	549
Mean	540	513	557	588	653	537	721	619	602	567

Chapter 4

**Comparison of Selection Efficiency for Spring Barley
(*Hordeum vulgare* L.) under Organic and Conventional
Farming Conditions**

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Abstract

The main objective of this research was to analyse whether selection under conventional conditions (indirect selection) is as effective as selection under organic conditions (direct selection) to develop varieties suitable for organic farming systems. Two F_3 barley (*Hordeum vulgare* L.) populations 'Primus'/'Idumeja' (P/I) and 'Anni'/'Dziugiai' (A/Dz) targeted for organic variety development were selected in two organically and two conventionally managed environments during F_3 to $F_{3.6}$. From there, the performance of the five best $F_{3.6}$ lines selected in each of the four environments from each cross (in total, 40 lines) were compared at all four sites during 2 yr. For obtaining varieties adapted to organic conditions for the P/I cross, it did not matter at which condition the selection was performed. For A/Dz, the best selection results in terms of yield, combined with other traits included for an organic ideotype, were achieved under well-managed organic conditions. We conclude that direct and indirect selection in early breeding stages are equally suitable for the development of cultivars for organic conditions if (i) care is taken that selection considers not only yield, but also other traits important for organic growing conditions, (ii) selection is not performed under too stressful conditions, and (iii) testing in later stages of the breeding program is conducted under various organic farming conditions for the best recommended varieties for organic agriculture.

Key words:

breeding for organic farming, selection criteria, cross population, selection environments, organic crop ideotype

Abbreviations:

A/Dz, cross Anni/Dziugiai; BYDV, *Barley yellow dwarf virus*; C1, conventional medium-input site; C2, conventional high-input site; GS, growth stage; O1, organic institute site; O2, organic farmer's site; OIS, Organic ideotype scores; P/I, cross Primus/Idumeja; TWG, Thousand grain weight; WSA, Weed suppression ability.

4.1 Introduction

Organic agriculture has been developing in Europe during the past decades. In the EU, the total organic land area has increased from 9.6 million ha in 2011 up to 10.3 million ha in 2011. During the last decade in the EU, area under organic management increased by about 500,000 ha yr⁻¹ (Eurostat, 2015).

Organic farming in Latvia evolved slowly, but now Latvia is among the EU countries with the fastest-developing organic farming segment. In 2014, organically certified area in Latvia accounted for around 11.0% of the total agricultural land area, while in the EU, the whole organic area represents 5.9% of the total utilized agricultural area. In 2015, organically certified agricultural areas have increased by 12.5% and occupied 237,462 ha. (Zemkopības Ministrija, 2015).

Cereals are important crops in both organic and conventional crop rotations in Latvia, and organic cereal production covers approximately 36,900 ha. Organic growers refrain from chemical inputs such as artificial fertilisers and pesticides. Therefore, breeding programs should focus on special plant characters for weed suppression, such as early vigour, rapid canopy cover, planophile leaf angle, and straw length (Hoad et al., 2005). Also, attention needs to be paid to disease resistance and end-use quality by the improvement of nitrogen (N) use efficiency under low-N input conditions to achieve acceptable protein content, optimal crop performance, and stable yield (Baresel et al., 2005; Mueller, 2005).

Organic farmers can make use of three different types of breeding programs to find varieties suitable for organic farming systems: (i) varieties derived from conventional breeding programs aimed at conventional agriculture, (ii) varieties derived from conventional breeding programs aimed for organic farming by taking into account some traits that are important for organic farming systems, and (iii) varieties derived from breeding programs performed solely under organic conditions (Wolfe et al., 2008).

More and more conventional breeding companies are interested in responding to the need of organic farmers for better-adapted cultivars for their low-input farming systems. However, the market for organic varieties is limited, and managing two different breeding programs within one company may not always be economically feasible. Therefore breeding companies are looking

for the most efficient way to develop varieties suitable for organic growing conditions (e.g., Löschenberger et al., 2008).

Like soil fertility, crop management and other factors vary substantially among organic farms, and between organic farms and organically managed selection fields of breeding companies, it is essential to find the most appropriate selection environment for developing successful varieties for organic farms (Wolfe et al., 2008). For breeders who select under conventional agronomic conditions for both conventional and organic farming systems, it seems more economically efficient to combine the breeding programs (e.g., by performing the initial selection process in their conventional selection fields and conducting only the final steps in organic fields) (Löschenberger et al., 2008). However, one might lose valuable genotypes with specific adaptation to organic environments during selection under conventional conditions. Herbicide application at conventionally managed trials preclude efficient selection for weed suppressive ability, and relatively high levels of N fertilization prevent selection for high nutrient use efficiency under low-N conditions (Wolfe et al., 2008).

Studies comparing variety performance in organic and conventional farming systems have led to contrasting outcomes. Przystalski et al. (2008) concluded that there were high genetic correlations between both systems for most traits over all tested cereal varieties, but some cultivars showed different ranking between environments (genotype \times environment). Therefore, their recommendation was to combine information from both organic and conventional trials. In a study on winter wheat (*Triticum aestivum* L.), Murphy et al. (2007) presented evidence that improving yield for organic conditions will require selection within organic systems (in this paper, direct selection) rather than selection in conventional systems (in this paper, indirect selection) and should include traits important to organic farmers. Reid et al. (2009) also concluded that indirect selection of spring wheat would not give the best possible result for organically managed production. Kirk et al. (2012) came to the same conclusion from their experiment of comparing results of spring wheat selection under both organic and conventional growing conditions. Although the above-cited authors compared direct and indirect selection aiming at varieties suitable for organic farming, none of these trials

were based on parental combinations representing varieties suitable for organic agriculture.

With respect to the choice of an appropriate selection environment for varieties suitable for organic farming, breeders are concerned that prediction of potential gains from selection in organically managed fields can be difficult because of lower heritability for relevant traits than under conventional conditions (Wolfe et al., 2008). Kokare et al. (2014) compared 10 cultivars at two organically and two conventionally managed sites and found that varieties ranked similar for grain yield between the organically managed site institute site (O1) and both conventionally managed sites C1 (medium-input) and C2 (high-input), but less with the organic farmer's field (O2). They argued that, due to the higher heritability for yield at the conventional sites, selection for yield for varieties suitable for organic conditions could successfully be performed under medium-input conventional conditions. Burger et al. (2008) compared selection of maize breeding lines under organic and conventional conditions and concluded that heritabilities were comparable. Their explanation for this finding was that not only the environmental variation under organic conditions was larger (due to irregular weed pressure in the fields), but the genetic variance was also larger (as many hybrids failed under organic conditions).

The objective of our experiment was to compare selection results for organic farming by carrying out the selection process under organic conditions (direct selection) and conventional conditions (indirect selection). We performed the selection in two different F3 populations based on parental combinations made specifically to serve an organic breeding program. Lines were selected during F3 to F3:6 in parallel in two organically managed fields (one institute's research field and one farmer's field) and two conventionally managed fields (medium- and high-input levels). The five best lines selected in each separate environment were then compared at all four sites during two subsequent growing seasons. The main research question was whether selection under conventional conditions (indirect selection) is as effective as selection under organic conditions (direct selection) to develop varieties suitable for organic farming systems. We also investigated whether traits related to weed suppressive ability are likely to contribute to higher yield under organic growing conditions.

4.2 Materials and methods

Genetic Resources

Our choice for the parents was based on some specific morphological and biological traits that might be beneficial for organic farming. Based on prior research (Kokare et al., 2014), contrasting parents (e.g., early or late maturing, short or tall, rapid or slow early development) with stable yield and different yield potential were selected and crossed. The crossing combinations used for the present study were (i) 'Primus'/'Idumeja' (P/I) and (ii) 'Anni'/'Dziugiai' (A/D). Primus (NGB16806) is an old Swedish variety (1901), and despite the fact that it was known as not high yielding based on previously obtained research results, it was chosen as one of the parents for its weed suppressive characteristics, such as moderately planophile plant growth habit, long leaves, good plant ground cover at early development stages, very tall stature, and late maturity (Kokare et al., 2014). Idumeja (LVA00165) is a comparatively recently released variety (in 2000). In contrast to Primus, Idumeja is a medium tall variety with good early vigour, erectophile plant growth type, early maturity, and a medium yield under organic growing conditions. In the second cross, the modern short straw Estonian variety Anni (EST14, released in 1993) with good yield potential and wide adaptation across various growing conditions (see Kokare et al., 2014) was crossed with the Lithuanian variety Dziugiai (AGB0257) from the middle of the last century (1947) with tall plants and poor lodging resistance but good early vigour.

In breeding for organic farming, it is essential to include parents with resistance or a high level of tolerance to diseases such as powdery mildew [*Blumeria graminis* (DC.) Speer] and netblotch (*Pyrenophora teres* Drechs.), because comparatively high infection can occur in organic conditions, especially in the case of netblotch, because fungicides are not applied as in conventional farming; however, it was not a focus for parental choice for this project. The four parents included were considered moderately susceptible to powdery mildew and netblotch, except for Idumeja, which was highly susceptible to netblotch, and Dziugiai, which was moderately resistant to netblotch.

Selection Environments

Selection in the two populations was performed at two organic and two conventional sites during 2006 to 2009, and the final evaluation of the selected lines was conducted in 2010 and 2011 at all four growing sites (see Supplemental Table S1). The first organic site (O1) was situated in a research field of the plant breeding institute (with pea green manure as fertiliser). The second organic site (O2) was at an organic farmer's field (with stable manure application, ploughed in autumn). Both fields were certified organic for more than 5 years at the beginning of the experiment. The first conventional site (C1) was located in a barley (*Hordeum vulgare* L.) breeding field (with a medium level of mineral fertiliser input) and the second conventional site (C2) was in a seed production field of the institute (with higher mineral fertiliser input). Fields were located in and around Priekuli (57°19' N, 25°20' E) in the west region of Latvia. Distances between sites O1, C1, and C2 were 0.5 to 2 km; O2 was within a 5-km distance of the other three sites. Soil and crop management characteristics for the evaluation years 2010 and 2011 are summarised in Supplemental Table S1. To gain insight into weed pressure, the weed ground cover in the organic test sites was visually estimated as the percentage of plot area covered by weeds (at growth stage [GS] 31–32, according to Zadoks's decimal scale; Zadoks et al., 1974). At the organic sites O1 and O2, the weed ground cover was, on average, 13 and 12% in 2010 and 5.3 and 6.2% in 2011. For weed control, harrowing was applied at O1 after weed ground cover evaluation. At O1, the main annual weeds were *Chenopodium* spp., *Galeopsis* spp., *Matricaria perforata* Mérat, *Thlaspi arvense* L., *Capsella bursa-pastoris* (L.) Medik., *Fallopia convolvulus* (L.) Á. Löve, and *Stellaria media* (L.) Vill. Among the perennial weeds, *Cirsium* spp. and *Rumex crispus* L. were dominant. At O2, no weed control was applied (as occurs in many organic farms in Latvia due to lack of appropriate equipment), which resulted in extremely high weed pressure at later stages at this site. The range of perennial weed species was broader at O2 than at O1, and included mainly weeds such as *Rumex crispus* L., *Cirsium* spp., *Taraxacum officinale* F.H.Wigg., *Artemisia vulgaris* L., *Ranunculus repens* L., *Potentilla anserina* L., and *Achillea millefolium* L. At the two conventional sites, herbicides were used for weed control.

Selection Method and Selection Criteria

The same breeding team performed the selection during the whole project with the focus on traits of importance for organic production conditions at all four sites, aiming to select lines suitable for organic farming systems (Supplemental Table S2). From 2006 to 2011, the parents were included in the selection trials of their respective progenies each year at each site.

For good weed suppression ability, emphasis in selection was given to the genotypes with early vigour, a taller canopy height, rapid crop ground cover at the beginning of the stem elongation stage, long and wide leaves, and taller plants at harvest but good lodging resistance. The preference was also given to a lower level of infection by pathogens, such as powdery mildew and netblotch, early heading, and maturing genotypes with high thousand-grain weight (TGW) and grain volume weight. See Fig. 4.1 for the selection scheme. In F_2 , bulk multiplication was conducted to obtain enough seeds for the selection in four environments from F_3 onward. For F_3 , bulk plots were sown at approximately 2000 seeds plot^{-1} , with a density of 80 plants m^{-2} to allow single-plant evaluation. At each site, 100 plants cross^{-1} were individually harvested and progenies sown in two to five rows of 1-m length, with a density of approximately 170 plants m^{-2} for the $F_{3:4}$ generation at the same site.

In $F_{3:4}$ (selection intensity of 20%), lines of each cross were rated for rapid early growth, more prostrate leaf angle, and early heading (plants that are just as early or earlier than the parents Idumeja and Dziugiai, respectively). All plants in the plot were harvested in bulk. Following harvest, lines with poor-looking grains (according to visual appearance of size, hull roughness, awn adherence, and color) were discarded. Finally, the 20 best-performing breeding lines were selected per cross and per site according to the above-mentioned criteria. In $F_{3:5}$, the sowing rate was approximately 400 seeds m^{-2} , and 10 out of 20 lines of each cross per site were selected for $F_{3:6}$. In $F_{3:5}$ and $F_{3:6}$, grain yield was considered as the most important selection criterion. In $F_{3:5}$, we selected out of the best-yielding lines those that scored highest for rapid early growth (scores from 1 [low] to 9 [high]), earlier heading (lines earlier than the late parent), and more prostrate leaf angle (scored from 1 [erect] to 9 [prostrate]).

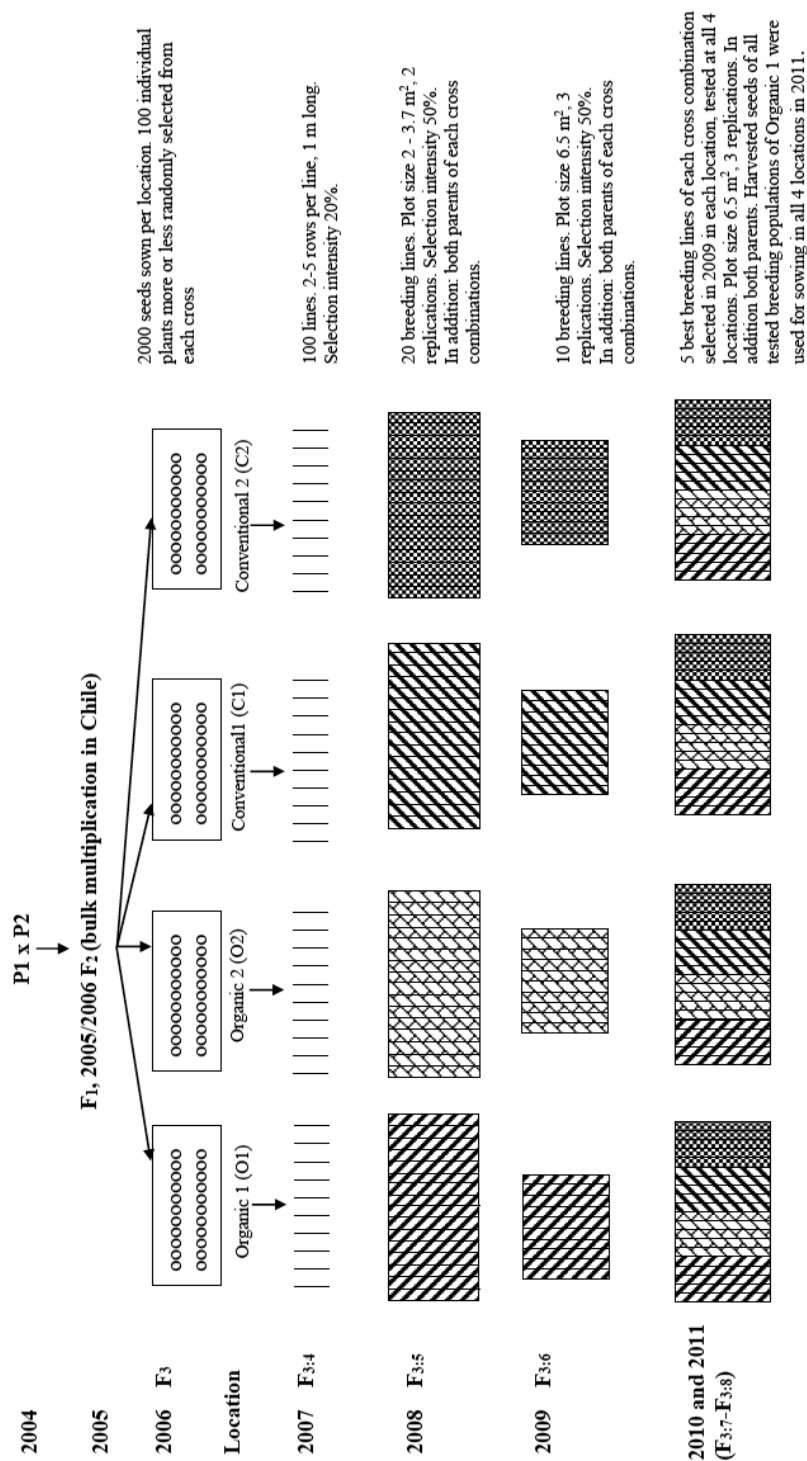


Figure 4.1 Scheme of the spring barley breeding experiment (showing one cross), Priekuli 2006 to 2011.

Lines with taller canopy height at stem elongation, better crop ground cover, longer and wider leaves that scored higher than the means of all lines of the cross, and medium tall or tall plants at harvest time were selected. With respect to the latter trait, we took care not to select for plant length taller than the tallest parent. Uniformity in plant type was also considered; lines segregating for plant height or for other easily visible traits were discarded. Also, grain quality traits (high TGW and volume weight) were a selection criterion. Disease infection levels (netblotch and powdery mildew) were not sufficient to select against high susceptibility.

In $F_{3:6}$, selection was done similarly to $F_{3:5}$. Starting from the $F_{3:6}$ generation, sowing rate was 400 seeds m^{-2} . Thousand-grain weight was used to prepare the required seed lots for sowing, but adjustments were made for germination rate.

Finally, from $F_{3:6}$, the five best lines of each cross per site were selected for the final comparison in 2010 and 2011. In total, 40 lines (i.e., five lines for each of four selection environments \times two crosses) plus the four parents were evaluated in 6.5- m^2 plots in three replications in a randomized complete block design. In 2011, seed material obtained in O1 was used for all lines in all test sites to eliminate the effect of site on seed quality and thus on yield and other plant traits in the following year.

Meteorological Conditions

The weather in the years of selection varied to a large extent (see Supplemental Tables S2 and S3). In 2006, the second half of growing period was extremely dry, resulting in a relatively low grain yield. In 2007, the growing conditions were favourable for growth and development of barley. In 2008, there was an insufficient amount of rainfall in the first part of growing period, but it was very rainy close to the maturity stage, and this decreased the grain quality. In 2009, average monthly air temperatures during the whole growing period were close to the long-term average.

During the comparison trials in both years (2010–2011), the meteorological conditions were, in general, favourable for barley cultivation. The summer of 2010 was the warmest in the history of Latvian meteorological observations and was characterized by thermal stability. The mean air temperature over the

whole growing period surpassed the long-term average and resulted in early maturity. The amount of rainfall at the end of July 2010 was 172% of the long-term average, which promoted lodging at both conventional sites, but not at the organic sites; in 2011, the amount of precipitation was close to the long-term average.

Evaluation of the Selected Breeding Lines

The following traits contributing to weed suppressive ability were scored in 2010 and 2011: plant growth habit (at GS 25–29,) with scores 1 (erect) to 9 (planophile); early vigour (at GS 25–29) with scores 1 (low) to 9 (high), as described by Donner and Osman (2006); plant canopy height (at GS 30–31) based on five measurements per plot (cm); crop ground cover (at GS 30–31), visually estimated as an overall percentage of plant-covered area per plot; length and width of flag leaf (at GS 47– 51), the average value of five randomly chosen and measured plants per plot (cm); plant height before harvest (at GS 90), the average of five plants per plot (cm); and resistance to lodging (at GS 90), with scores 0 (low) to (9) high. Length of growth periods from sowing to heading (GS 60) and to maturity (GS 90) was estimated (days); after harvest, TGW was assessed (g).

Resistance to leaf diseases powdery mildew and netblotch was scored 0 (high susceptibility) to 9 (high resistance) for each plot. In 2010 at C1, an extraordinarily strong infection with *Barley yellow dwarf virus* (BYDV) was observed. This disease was not noticed in the trials of the experiment in the years before and no selection was conducted for resistance to this virus. The effect of the virus infection could have influenced the results at C1 in 2010. In 2011 at O1, a high infestation by cockchafer (*Melolontha melolontha*) occurred.

The organic ideotype score (OIS), as is given in Table 4.1 was used to compare the selected breeding lines during the two testing years. This OIS served to consistently rate the phenotypic performance of the lines. A weight was given to each of the evaluated traits considered important for adaptation to organic farming systems, as discussed in the introduction and in accordance with the expert views of our breeders and some farmers with whom we spoke during field trial visits. In general, the higher the trait value, the better the suitability is considered for organic farming. The highest values were given to shorter

growth periods from sowing to heading and from sowing to maturity (standardized values were changed to negative values by multiplication with “-1”).

Table 4.1 The relative weight (%) of the traits included in the organic ideotype score (OIS) as applied in the comparison of the selected barley breeding lines in 2010 and 2011.

Trait†	OIS components‡	Growing stage§	Relative Weight %
Grain yield, Mg ha ⁻¹	Y	after harvest	40
Early vigour	WSA	GS 31–32	3
Canopy height, cm		GS 31–32	17 (20) [¶]
Crop ground cover, %		GS 31–32	10
Width of flag leaf, cm		GS 47– 51	2
Length, of flag leaf, cm		GS 47– 51	2
Lodging resistance		GS 90	2
Plant height, cm		GS 90	6
Resistance to leaf diseases	DR	From GS 32	6
Heading, d	GP	GS 60	5
Maturity, d		GS 90	5
TGW, g	Q	after harvest	2
Total			100

† Early vigour was scored 1 (low) to 9 (high); crop ground cover was visually estimated as an overall percentage of plant covered area per plot; lodging resistance was given using 1 to 9 scores with 0 = low to 9 = high; resistance to leaf diseases was scored 0 (high susceptibility) to 9 (high resistance); heading was days from sowing to heading; maturity was days from sowing to maturity.

‡ Y, grain yield; WSA, weed suppressive ability; DR, disease resistance; GP, growing period; TGW, thousand-grain weight; Q, grain quality.

§ Growing stages (GS) according to Zadoks et al. (1974) at which traits were scored.

¶ Because of a strong positive correlation ($r = 0.84–0.82$, $p < 0.01$, data not shown) between early vigour and canopy height in 2010, early vigour was not estimated in 2011 and, in the organic ideotype score, the relative weight of this trait was added to plant canopy height.

The OIS was calculated for each line at each test site based on the following formula described by Bänziger et al. (2000):

$$\text{OIS} = (b_1P_1 + b_2P_2 + \dots b_{12}P_{12})/100$$

where b_1 is the relative weight given to trait 1 in the organic ideotype score (Table 4.3), and P_1 is the observed standardized value of the trait, calculated as:

$$P_1 = \frac{(x_i - \bar{x}_{pl})}{s_p}$$

where x_i is the mean of the trait for each individual line in a test site, \bar{x}_{pl} is the mean of the traits of all lines in a test site, and σ_p the standard deviation over the mean trait values of all lines in a test site. We analysed OIS and its components to gain insight into which selection site was most effective for selection for the above-mentioned characteristics important for organic farming.

Statistical Analysis

Analysis of variance and the calculation of phenotypic correlation coefficients were performed using Genstat 14.0 (VSN International, 2011). Analysis of variance was used to determine the impact of the organic versus conventional growing sites and other factors such as year, selection site, and cross effects on the yield and traits associated with weed competitiveness. Pearson's correlations (r) were calculated to compare relationships among phenotypic traits. The consistency of ranking of the genotypes between test sites was assessed with Spearman's rank correlation coefficient (r_s). Statistical significance was assessed at the 95% confidence level ($\alpha = 0.05$).

4.3 Results

Grain Yield

Selection and Test Sites

Grain yield was significantly ($p < 0.01$) influenced by all main factors (year, test site, selection site, and cross combination) and two-way (test site \times selection site and test site \times cross), as well as three-way (year \times test site \times cross), interactions. In general, yield in 2010 was lower than in 2011 (Table 4.2), probably due to the high lodging in 2010 at both conventional sites and the BYDV infection at C1. The situation was opposite only for the test site O2, where extremely low yield was obtained in 2011 due to the high cockchafer infestation and high weed pressure. As expected, the selected lines yielded lowest at the test site O2 (with the lowest input and highest weed pressure) and the highest at the highest-input level of the conventional site C2. Over both crosses and years, differences in yield did not depend strongly on the site at which the lines were selected (Table 4.2). At test site O1, the lines selected in O1 and C1 yielded highest (see Table 4.2) row “Mean over crossing combinations”). At the O2 test site, there were no significant differences in yield between lines originating from the four different selection sites. Lines selected in the poor environment O2 were low yielding at all test sites. The lines selected from high-input site C2 yielded significantly higher at the conventional test site C2 than lines selected from the two organic sites. Spearman’s rank correlation coefficient was used to compare grain yield for the lines between testing sites. A moderate rank correlation coefficient was found between C1 and O1 ($r_s = 0.648$, $P < 0.001$) and between C2 and O1 ($r_s = 0.469$, $P = 0.02$), but not between the two organic sites ($r_s = 0.123$, $P > 0.05$).

These results show that, with respect to yield, selection under conventional conditions (indirect) and well-managed organic (direct) conditions both were about equally effective in developing varieties for organic farming. Selection in the poor organic environment (direct selection) did not lead to high-yielding genotypes at either organic test site

Crosses

In general, the lines of A/Dz yielded higher than lines of P/I, except at the organic test site O2, where significant ($P = 0.002$) year \times cross interaction was observed (Table 4.2., see for each testing site the mean over selection sites). Under the favourable conditions of 2010, lines of A/Dz yielded higher than lines of P/I in O2; however, in 2011 with a cockchafer incidence and weeds, lines of P/I achieved significantly higher yield than the lines of A/Dz. In the average ranking of the total set of 40 lines included in the testing experiment, the lines of the P/I cross often took higher ranking positions at O2 than at O1 and the two conventional sites (Supplemental Table S4). At the organic site O1 and both conventional sites, the lines of A/Dz generally ranked higher than P/I lines.

At both organic sites, parents Anni and Dziugiai ranked higher in comparison to Idumeja and Primus. At test sites O1 and O2, most of the lines ranked higher than parents and the grain yield of the tested lines was mainly at the level of the highest-yielding parents in each cross, Idumeja and Anni (Figure 4.2, Supplemental Table S4). Two lines selected at O1 and C1 significantly exceeded Idumeja at the evaluation in O2 (test for significance not shown), but it was due to low yield of Idumeja at this test site.

Table 4.2 The average grain yield (t ha^{-1}) for barley breeding lines grown at two organic (O1 and O2) and two conventional (C1 and C2) test sites, depending on selection site, cross combination and year \times test site \times cross combination interaction

Cross †	Year	Test sites‡																			
		O1				O2				C1				C2							
		Selection site§				Mean over selection sites				Selection site				Mean over selection sites							
		O1		C1		O2		C1		O1		C2		O1		C2		O1		C2	
		O1	O2	C1	C2	O1	O2	C1	C2	O1	O2	C1	C2	O1	O2	C1	C2	O1	O2	C1	C2
P/I	2010	2.25a‡	2.08a	2.34a	2.31a	2.24	2.77a	2.65a	2.54a	2.62a	2.61	1.95a	2.19a	2.18a	2.07a	2.15	3.43a	3.54a	3.74a	3.68a	3.65
	2011	4.33a	3.89b	4.12ab	4.06ab	4.02	1.40a	1.17a	1.51a	1.30a	1.34	2.72a	2.45a	2.66a	2.70a	2.60	5.40a	4.99b	4.90b	5.06ab	4.98
Mean over the years		3.29a	2.98b	3.23ab	3.17ab	3.13	2.09a	1.91a	2.02a	1.96a	1.99	2.34a	2.32a	2.42a	2.39a	2.38	4.41a	4.27a	4.32a	4.37a	4.32
A/Dz	2010	2.88a	2.57ab	2.77ab	2.5b	2.68	2.65a	2.79a	2.77a	3.08a	2.82	2.48a	2.48a	2.61a	2.67a	2.59	3.78a	3.36c	3.86ab	4.20a	3.81
	2011	4.60a	4.54a	4.88a	4.53a	4.64	1.13a	1.06a	0.97a	1.21a	1.09	2.91ab	2.73b	3.17a	3.17a	3.02	5.45a	5.34b	5.46ab	5.75a	5.52
Mean over the years		3.74ab	3.55ab	3.83a	3.51b	3.66	1.89a	1.92a	1.87a	2.15a	1.95	2.70ab	2.61b	2.89a	2.92a	2.81	4.61a	4.35c	4.66b	4.97a	4.66
Mean over combinations		3.51ab	3.27c	3.53a	3.35bc		1.99a	1.92a	1.95a	2.05a		2.52ab	2.46b	2.66a	2.65a		4.50b	4.31c	4.49abc	4.67a	

† Test sites: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site

‡ Cross: P/I = Primus /Idumeja, A/Dz = Anni/Dziugiai

§ Selection site: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site

¶ Mean values are compared per row between selection sites within each test site. No label in common indicates significantly different at $p < 0.05$

Comparison of Selection Efficiency for Spring Barley (*Hordeum vulgare* L.) under Organic and Conventional Farming Conditions

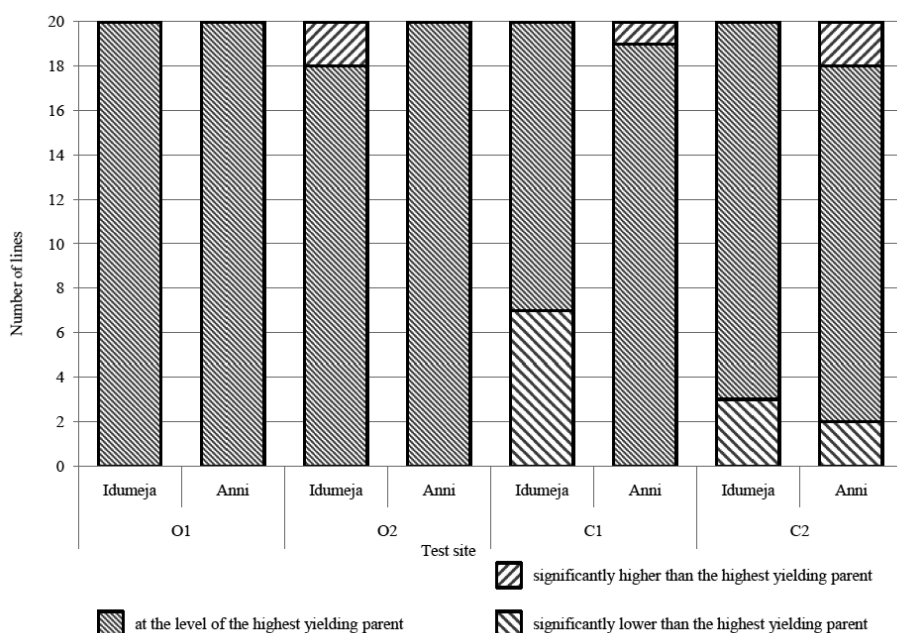


Figure 4.2 The number of lines of the barley crosses Primus/Idumeja (P/I) and Anni/Dziugiai (A/Dz) with yield at the level of best parent and lines significantly different from the best yielding parent at organic (O1 and O2) and conventional (C1 and C2) sites.

Organic Ideotype Score

Overall

As the goal was to select for cultivars adapted to organic growing conditions for yield and other traits important for organic farming such as early vigour, canopy height, crop ground cover and plant height before harvest, and TGW, the performance of the selected lines was analysed according the OIS.

Selection results for OIS differed over selection sites for both crosses. The most contrasting results for both crosses were obtained for the lines selected at farmer's site O2 (Table 4.3).

Table 4.3 Average OIS (organic ideotype scores) over all testing sites for the lines of two barley crosses derived from organic (O1 and O2) and conventional (C1 and C2) selection sites, tested in 2010-2011.

OIS components [‡]	Cross [§]	Selection site [†]			
		O1	O2	C1	C2
OIS	P/I	15.6	-15.2	9.5	4.1
	A/Dz	-0.3	27.4	-7.4	-2.3
	Mean OIS per selection site	7.7	6.1	1.1	0.9
Y	P/I	3.3	-27.1	-4.7	-11.9
	A/Dz	14.7	6.8	18.3	21.1
	Mean Y per selection site	9.0	-10.1	6.8	4.6
WSA	P/I	15.5	13.6	12.6	7.4
	A/Dz	-14.5	16.3	-20.6	-20.3
	Mean WSA per selection site	0.5	14.9	-4.0	-6.4
DR	P/I	-3.4	-2.8	-1.9	0.2
	A/Dz	1.8	1.9	2.1	2.0
	Mean DR per selection site	-0.8	-0.5	0.1	1.1
GP	P/I	-0.7	0.2	1.8	6.2
	A/Dz	-1.4	3.7	-5.8	-3.5
	Mean GP per selection site	-1.1	2.0	-2.0	1.3
TGW	P/I	1.0	0.9	1.6	2.1
	A/Dz	-0.8	-1.3	-1.4	-1.6
	Mean TGW per selection site	0.1	-0.2	0.1	0.3

[†] Selection site: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site.

[‡] OIS components: Y= yield, WSA = weed suppressive ability, DR = diseases resistance, GP = growing period, TGW = thousand grain weight.

[§] Cross: P/I = Primus /Idumeja, A/Dz = Anni/Dziugiai

Contrary to other sites, lines selected at site O2 had lower yield but higher weed suppressive ability, especially for the lines derived from cross A/Dz. At both organic test sites, the lines of P/I were observed to have a higher weed suppressive ability, earlier maturity, and higher TGW (Table 4.4) than lines of the A/Dz cross, which were high yielding and more resistant against diseases.

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Table 4.4 Organic ideotype scores (OIS) components for the lines of two barley crosses derived from organic (O1 and O2) and conventional (C1 and C2) selection sites tested at both organic sites in 2010-2011

		Test sites [†]									
OIS components ‡	Cross §	O1				Mean of cross per test	O2				Mean of cross per test site
		Selection site [¶]					Selection site				
		O1	O2	C1	C2		O1	O2	C1	C2	
Y	P/I	-13.0	-47.7	-19.3	-24.7	-26.2	19.6	-6.4	10.0	1.0	6.0
	A/DZ	39.0	17.6	48.9	13.0	29.6	-9.6	-4.0	-12.3	29.1	0.8
WSA	P/I	14.6	3.4	7.3	0.6	6.5	16.5	23.7	18.0	14.2	18.1
	A/DZ	-6.6	22.0	-12.6	-24.4	-5.4	-22.4	10.5	-28.6	-16.2	-14.2
DR	P/I	-2.7	-2.3	-0.7	0.7	-1.2	-4.1	-3.4	-3.2	-0.2	-2.8
	A/DZ	1.1	0.8	1.1	1.9	1.2	2.6	2.9	3.1	2.0	2.7
GP	P/I	-0.5	-0.1	1.3	6.7	1.9	-1.0	0.5	2.3	5.6	1.7
	A/DZ	-1.1	4.0	-7.3	-3.4	-2.0	-1.8	3.4	-4.3	-3.6	-1.6
TGW	P/I	0.9	0.7	1.5	2.1	1.3	1.0	0.9	1.8	2.1	1.5
	A/DZ	-0.6	-1.2	-1.0	-2.1	-1.2	-1.0	-1.3	-1.9	-1.1	-1.3
OIS	P/I	-0.7	-45.8	-9.9	-14.6	-17.7	31.9	15.3	28.8	22.7	24.7
	A/DZ	31.7	43.2	29.1	-15.0	22.3	-32.2	11.6	-43.8	10.3	-13.6

† Test sites: O1 = organic institute site, O2 = organic farmer's site.

‡ OIS components: Y = yield, WSA = weed suppressive ability, DR = diseases resistance, GP = growing period, TGW = thousand grain weight.

§ Cross: P/I = Primus /Idumeja, A/Dz = Anni/Dziugiai

¶ Selection sites: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site.

Relation between OIS Components and Selection Sites

Correlations (Pearson) between the phenotypic traits (included in OIS components) and grain yield mainly differed between organic and conventional test sites (Table 4.5). In the organic sites, weed suppressive traits such as early vigour, canopy height, crop ground cover and plant height before harvest, and TGW tended to have a positive correlation with grain yield in comparison with the conventional sites, where correlations tended to be negative (Table 4.5). Number of days to heading correlated negatively with grain yield, which indicated that late-heading genotypes may have lower yield under organic conditions. Days to heading were also negatively related to early growth vigour, canopy height, and crop ground cover (not shown).

Under organic conditions, the latter three traits were correlated positively with grain yield. That was the reason why we gave the highest value in OIS calculation to the genotypes with a shorter growth period from sowing to heading and maturity. These correlations suggest that early-heading genotypes should be favoured in selection programs for organic conditions.

Table 4.5 Pearson's correlation (r) between grain yield and traits included in the organic ideotype score (OIS): for two barley crosses in two organic sites (O1 and O2) and two conventional sites (C1 and C2) test sites.

Trait [†]	OIS component s [‡]	Test site [§]			
		O1	O2	C1	C2
Early vigour	WSA	0.23	0.06	-0.23	-0.31*
Canopy height, cm		0.23	0.28	-0.18	-0.43**
Crop ground cover, %		0.71**	0.23	-0.04	-0.07
Length of flag leaf, cm		0.39*	-0.13	0.14	-0.21
Width of flag leaf, cm		0.37*	0.15	0.19	0.06
Lodging resistance		-0.10	0.03	0.52**	0.40*
Plant height, cm	DR	0.46**	0.18	-0.23	-0.62*
Resistance to powdery mildew		0.21	0.31*	0.55*	0.34*
Resistance to netblotch	GP	0.10	0.03	-0.32*	-0.02
Heading, d		-0.38*	-0.19	0.01	0.11
Maturity, d	Q	0.25	0.03	0.07	0.34*
TGW		0.26	0.38*	-0.16	-0.02

[†] Early vigour: was scored 1 = low to 9 = high; crop ground cover: visually estimated as an overall percentage of plant covered area per plot; lodging resistance: 1-9 scores with 0 = low to 9 = high; resistance to leaf diseases: was scored 0 = high susceptibility to 9 = high resistance; heading: days from sowing to heading; maturity: days from sowing to maturity.

[‡] OIS components: WSA = weed suppressive ability, DR = diseases resistance, GP = growing period, TGW = thousand grain weight, Q = grain quality.

[§] Test sites: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site

* Significant at $p < 0.05$.

** Significant at $p < 0.01$.

The selection results for weed suppressive ability (WSA) differed between crosses. For P/I, no great differences were observed with respect to selection sites, but for A/Dz, the highest WSA was achieved at selection site O2 (Table 4.3). With respect to grain quality trait TGW, selection under conventional conditions gave the best selection results for P/I, while for A/Dz, organic conditions were slightly better than conventional conditions.

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In this study, yield and WSA were each taken to represent 40% relative weight of the OIS; to gain insight how these two components are connected at each test site, we performed a correlation analysis.

At O1 and C1, the correlations between yield and WSA tended to be positive for P/I lines and negative for the A/Dz lines (Table 4.6). However, at the most stressful environment, O2, A/Dz lines had stronger positive correlation in comparison with P/I lines. At the highest-input environment, C2, correlations tended to be negative. This indicates that it depends on the cross whether selection on WSA components contributes to higher-yielding varieties for organic farming.

Table 4.6 Pearson correlation coefficient (r) between grain yield and weed suppressive ability for two barley crosses selected at two organic (O1 and O2) and two conventional sites (C1 and C2) test sites.

Crosses [†]	Test sites [‡]			
	O1	O2	C1	C2
Over all lines	-0.19	0.29	-0.14	-0.34
P/I	0.35	0.02	0.39	-0.06
A/Dz	-0.20	0.37	-0.32	-0.38

[†] Crosses: P/I = Primus /Idumeja, A/Dz = Anni/Dziugiai

[‡] Test sites: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site

The ranking of lines for yield and WSA (Supplemental Tables S5 and S6) confirmed that, only for the P/I lines at O1, yield and WSA tended to be positively associated. At this site for this cross, three lines in the top five for yield were also in the top five for WSA. In the test site O2, there were two lines of this cross in common in the top five for both yield and WSA, but some lines that ranked high for one criterion ranked low for the other (Supplemental Table S5). For the A/Dz cross (Supplemental Table S6), only one line selected in C1 was in common in the top five for both criteria in O1, and the same line in O2, with addition of another line selected in C2. For both crosses, the lines that were selected in O2 and were in the top five of WSA did not rank in the top five for yield in either organic test site.

4.4 Discussion and Conclusion

Do Lines Selected under Conventional Conditions Differ in Yield from Lines Selected under Organic Conditions?

With respect to average yield, our results suggested that lines selected in O1 and the two conventional sites performed equally well when tested at O1 (Table 4.2). This means that indirect selection for yield under the conventional conditions of C1 and C2 showed similar results to direct selection under the organic conditions of O1. Also, Spearman's rank correlation for yield between the conventional and organic sites indicated that the high-yielding genotypes in the conventional sites (indirect selection) may be high yielding under organic conditions as well. This is in contrast to the findings of Murphy et al. (2007), who concluded from their trials with 35 wheat genotypes (breeding lines) that the highest-yielding genotypes in conventional fields did not rank the highest in organic systems. This difference could be due to the fact that the latter used lines originated from a conventional breeding program and were selected for high performance under conventional conditions. In our trial, we selected lines from crosses of which the parents were chosen for traits important for organic growing conditions, and in addition, the selection procedure was also aimed at selecting varieties for organic farming systems. The line selection at the organic farmer's field O2 did not lead to a high yield for any of the growing conditions, including O2 itself. This is in accordance with results we obtained in our previous experiment, where we compared the performance of 10 different varieties in the same four environments, as in these selection trials (Kokare et al., 2014). In that study, we found a high correlation between yield results of O1, C1, and C2, but not between O1 and O2, nor between O2 and the conventional sites. Heritability estimates for yield and yield components were very low for O2 because of high residual variance and genotype \times year interaction under these conditions.

Does Selection under Organic or Conventional Growing Conditions Result in Different Outcomes with Respect to Various Traits of Importance for Organic Farming?

Organic farmers do not always experience yield loss from weeds but nevertheless find weed suppressive traits of great importance to avoid building up weed seed banks for the following crops that do suffer from weeds, such as carrots (*Daucus carota* L.) and onions (*Allium cepa* L.) (Hoad et al., 2012).

By applying the OIS, we took into account traits of importance for organic agriculture other than merely yield. Such plant traits are length of growth period, tolerance to diseases, TGW, and traits that are known to enhance the weed suppressive ability, i.e. early vigour, canopy height, soil shading ability, and plant height at harvest (Coleman et al., 2001; Hoad et al., 2005, 2008; Mason and Spaner, 2006; Murphy et al., 2008).

The best selection result for OIS was achieved for the A/Dz cross by direct selection in the more variable organic selection site O2 (Table 4.4); this was due to very high weed suppressive ability but comparatively low yield for lines derived from O2. Anni and Dziugiai are, agronomically and morphologically, strongly contrasting parents. Of these, Dziugiai, with its tall canopy and high early vigour, may have contributed to the high weed suppressive ability in the generally weed-infested environment of O2. Selection of individual plants started in 2006, when weather conditions were extremely dry and hot. Growing conditions were relatively poor, especially in the organic farmer's field (O2) due to a high weed incidence. That provided us the opportunity to select for plants that stood out for traits of importance for organic farming, such as weed suppressive traits, and a shorter growing period in comparison to the other selection sites. However, it turned out in the final comparison that the yield of lines selected at O2 was lower than that of lines selected at other sites. From the ranking of O2 selected lines for yield and weed suppressive ability (see Supplemental Tables S5 and S6), we conclude that selection under extreme and variable conditions led to genotypes with outstanding weed suppressive ability in organic sites, but even in organically managed sites, these genotypes did not compete in productivity with genotypes that were selected under more optimal growing conditions. Lines with a better balance between weed suppression ability and yield were derived from selection at O1. Selection at the conventional sites also showed good results in regard to OIS, but the result obtained there was opposite of the result at O2: high yield but low weed suppressive ability. From this observation, we conclude that input level in both conditions played an important role in the selection process for organic farming. Direct selection under more favourable organic growing conditions, such as in O1, and also indirect selection under medium-input conventional conditions (C1) can thus be recommended for breeding for organic farming. Our results are also in agreement with those of Mikó et al.

(2014), who compared wheat varieties with different breeding origins (organic breeding, conventional, and combined strategies) under organic and low-input management and concluded that the environment where selection is performed has measurable effects on the performance of bread wheat varieties under organic and low-input growing conditions.

In our selection trials, the two crosses showed different results: the breeding lines of A/Dz were generally more productive under favourable organic conditions, but under unfavourable conditions (O2 in 2011), the lines of P/I yielded significantly higher ($p = 0.002$). For the P/I cross, the selection site did not have a large impact on weed suppressive traits, and selection at O2 resulted in a low yield and, finally, in low OIS. The parents of this cross were less contrasting for morphological traits. Primus is a very tall variety, while Idumeja is medium tall, but with rapid early growth. The largest differences were in the length of the growing period: Primus is very late maturing, while Idumeja is early maturing. The higher weed suppression ability, earliness, and larger grains of the P/I breeding lines ensured a higher yield under unfavourable organic conditions with high weed and pest pressure, like in O2 in 2011, in comparison with the performance of the lines selected from the cross A/Dz. The lines developed in our experiments are very useful for future studies on the effect of traits related to weed suppressive ability on yield under organically managed conditions.

This experiment also taught us that, for the breeding populations that are derived from parents that contrast for traits of importance for organic farming such as weed suppressive ability, organic selection sites can have an advantage over conventionally managed sites to identify genotypes with good expression of such traits. However, a negative impact of increased weed suppressive ability on grain yield cannot be ruled out. Our results support the results obtained by Reid et al. (2009), who concluded that creating a population from parents with different morphological and/or physiological traits of potential interest for organic systems may result in greater differences in selection results between the two systems. Results of our experiment indicated that whether direct selection or indirect selection is more effective depends on the properties of the parents that are crossed.

Our trial showed that, for the poorest organic site O2, it did not matter from which selection site the lines were derived, as they performed equally with

respect to yield. Our trials also revealed that there was a low-rank correlation of selected lines for yield between the two organic farms compared with better correlations between O1, C1, and C2. The line testing under different organic conditions contributes to identifying the best-performing cultivars for various organic farms and also allows us to recommend to farmers the most appropriate cultivars to particular sites after variety registration. This is in accordance with the findings and recommendations of Przystalski et al. (2008), who compared the performance of cultivars of cereal varieties under various organic and conventional growing conditions. Also, Osman et al. (2016) describes how conventional breeders, for reasons of cost efficiency, acknowledge that mixed breeding programs, where the demands of organic farming are prioritized and integrated into a conventional breeding program, can be a solution for obtaining spring wheat cultivars adapted to organic farming. Osman et al. (2016) recommended a mixed program that starts the first generations of breeding under conventional conditions and splits up in the final years of the program into organic and conventional parts.

Based on our selection experiment with barley, we finally conclude that direct and indirect selection in early breeding stages are about equally suitable for the development of cultivars performing well under organic conditions if (i) care is taken that selection not only considers yield, but also traits of importance for organic growing conditions, (ii) selection is not performed under too stressful of conditions, and (iii) (final) testing at later stages of the breeding program is conducted under various organic farming conditions for the best-recommended varieties for organic management.

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Supplementary material

Table S1 Description of the soil characteristics and crop management systems of the trials under organic (O1, O2), and conventional (C1, C2) test sites in Priekuli, 2010– 2011.

Site*	Year, sowing, harvest data	Soil type	Soil texture	pH _{KCl}	P ₂ O ₅ (mg kg ⁻¹)	K ₂ O (mg kg ⁻¹)	Humus content (g kg ⁻¹)	Available N content in soil in spring (kg ha ⁻¹)	Precrop	Amount of N-P-K	Fertility management	Diseases, pests	Management of Weeds
O1	2010 21 Apr 2 Aug	Sod podzolic	Loamy sand	5.7	111	144	28	75.6	Peas for green manure	N 26 kg ha ⁻¹	Green manure, 20 t ha ⁻¹ (to pre-crop)	No	1 x harrowing at tillering stage
	2011 21 Apr 3 Aug	Sod podzolic	Loamy sand	5.4	116	98	21	56.7	Peas for green manure	N 26 kg ha ⁻¹	Green manure, 20 t ha ⁻¹ (to pre-crop)	No	1 x harrowing at tillering stage
	2010 19 Apr 3 Aug	Sod podzolic	Loamy sand	6.5	265	173	35	94.5	Perennial grasses	N 41 kg ha ⁻¹	Stable manure 40 t ha ⁻¹	No	No
O2	2011 20 Apr 1 Aug	Sod podzolic	Loamy sand	6.6	394	167	30	81	Spring wheat	No	No	No	No
	2010 30 Apr 12 Aug	Sod podzolic	Loamy sand	5.5	100	132	26	70.2	Potatoes	N 80 kg ha ⁻¹ P 48 kg ha ⁻¹ K 84 kg ha ⁻¹	Inorganic (300 kg ha ⁻¹) N 5-P14-28), 200 kg ha ⁻¹ (NH ₄ NO ₃)	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator OD 0.15 l ha ⁻¹
	2011 27 Apr 9 Aug	Sod podzolic	Loamy sand	5.4	187	165	30	81	Potatoes	N 83 kg ha ⁻¹ P 45 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic (300 kg ha ⁻¹) N 5-P15-25), 200 kg ha ⁻¹ (NH ₄ NO ₃)	Herbicide Secator OD 0.15 l ha ⁻¹	
C2	2010 3.05 17.08	Sod podzolic	Loamy sand	5.6	115	159	28	84.3	Potatoes	N 120 kg ha ⁻¹ P 48 kg ha ⁻¹ K 84 kg ha ⁻¹	Inorganic (300 kg ha ⁻¹) N 5-P14-28), 200 kg ha ⁻¹ (NH ₄ NO ₃)	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator OD 0.15 l ha ⁻¹
	2011 29.04 10.08	Sod podzolic	Sandy loam	4.5	200	155	23	62.1	Potatoes	N 120 kg ha ⁻¹ P 45 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic (300 kg ha ⁻¹) N 5-P15-25), 200 kg ha ⁻¹ (NH ₄ NO ₃)	Herbicide Secator OD 0.15 l ha ⁻¹	Herbicide Secator OD 0.15 l ha ⁻¹

*Site: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site.

Table S2 Mean air temperature (°C) at Priekuli, 2006-2011.

	2006			2007			2008			2009			2010			2011		
	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C	Mean temperature °C	long term average °C
April	6.0	1.6	5.0	0.6	7.7	3.0	7.0	2.4	6.6	2.0	7.3	2.7						
May	11.6	0.9	12.3	1.6	11.1	0.1	11.7	0.7	13.0	2.0	11.6	0.6						
June	16.4	1.9	16.6	2.1	13.7	-1.0	14.1	0.5	15.0	0.3	17.6	2.9						
July	19.8	3.2	16.5	-0.2	16.8	0.2	17.2	0.6	22.1	5.4	20.3	3.6						
August			18.2															
t	17.8	2.1		2.6	16.7	-0.2	15.7	0.1	18.7	3.0	16.8	1.1						

Table S3 Amount of precipitation (mm) at Priekuli, 2006-2011.

	2006			2007			2008			2009			2010			2011		
	Sum, mm	% of long term average	Sum, mm	% of long term average	Sum, mm	% of long term average	Sum, mm	% of long term average	Sum, mm	% of long term average	Sum, mm	% of long term average	Sum, mm	% of long term average	Sum, mm	% of long term average	Sum, mm	% of long term average
April	24	60	37	93	77	0	40	19	41	104	19	48						
May	60	109	70	127	9	16	54	42	90	161	76	136						
June	34	44	79	103	66	86	134	174	111	142	46	59						
July	7	7	119	124	65	68	122	118	116	124	80	85						
August	40	44	113	126	156	173	82	92	113	131	115	134						

Table S4 Average ranking for yield of lines of the P/I (marked in grey) and A/Dz (no marking) crosses at the organic (O1 and O2) and conventional (C1 and C2) test sites.

Rank	O1	Yield t ha ⁻¹	Test sites [†]		Yield t ha ⁻¹	C1	Yield t ha ⁻¹	C2	Yield t ha ⁻¹
			O2	Yield t ha ⁻¹					
1	C1 [‡] -A/Dz [§] -34 [¶]	3.94	C2-A/Dz-66	2.34	C2-A/Dz-36	3.22	C2-A/Dz-36	5.43	
2	C1-A/Dz-52	3.93	C1-P/I-35	2.28	C2-A/Dz-66	3.03	C1-A/Dz-62	5.23	
3	O1-A/Dz-12	3.90	O1-P/I-93	2.27	C1-A/Dz-5	3.02	C2-A/Dz-66	4.96	
4	O1-A/Dz-92	3.89	C2-A/Dz-37	2.24	C1-A/Dz-34	2.98	C2-A/Dz-84	4.96	
5	C1-A/Dz-62	3.89	O1-P/I-63	2.23	C2-A/Dz-37	2.95	C2-A/Dz-85	4.89	
6	O1-A/Dz-79	3.85	C2-A/Dz-84	2.21	C1-A/Dz-52	2.95	O1-A/Dz-79	4.83	
7	C1-A/Dz-5	3.83	C1-A/Dz-52	2.20	C1-A/Dz-62	2.90	O1-A/Dz-65	4.74	
8	O1-A/Dz-65	3.76	O1-A/Dz-12	2.15	O1-A/Dz-12	2.88	C1-A/Dz-52	4.73	
9	O2-A/Dz-57	3.69	O2-A/Dz-57	2.12	O1-A/Dz-92	2.88	O1-P/I-63	4.71	
10	C2-A/Dz-36	3.63	C2-P/I-98	2.12	O2-A/Dz-57	2.87	C1-A/Dz-5	4.71	
11	C2-A/Dz-66	3.62	C2-A/Dz-85	2.11	O1-A/Dz-79	2.86	O2-A/Dz-57	4.69	
12	O2-A/Dz-49	3.58	O2-P/I-51	2.05	C2-A/Dz-84	2.83	O1-P/I-34	4.69	
13	O2-A/Dz-54	3.57	O1-P/I-34	2.04	Idumeja	2.75	C1-P/I-32	4.69	
14	Anni	3.57	C1-P/I-25	2.04	C1-P/I-32	2.73	O2-A/Dz-54	4.67	
15	O2-A/Dz-52	3.55	C2-P/I-72	2.04	O2-P/I-58	2.71	O1-A/Dz-92	4.64	
16	C2-A/Dz-37	3.54	C1-P/I-32	2.03	O2-P/I-97	2.67	C2-A/Dz-37	4.64	
17	C1-A/Dz-20	3.54	O2-A/Dz-54	1.99	O2-A/Dz-69	2.64	C2-P/I-72	4.63	
18	C2-A/Dz-84	3.52	O1-A/Dz-92	1.99	O2-A/Dz-52	2.64	C1-A/Dz-34	4.62	
19	C2-P/I-98	3.48	O1-P/I-86	1.98	C1-P/I-35	2.62	Idumeja	4.61	
20	O1-P/I-93	3.44	C2-P/I-92	1.96	O1-P/I-63	2.61	Anni	4.61	
21	O1-P/I-75	3.42	C2-P/I-40	1.95	C1-A/Dz-20	2.60	O2-P/I-58	4.54	
22	C1-P/I-83	3.41	C1-A/Dz-62	1.94	O2-A/Dz-54	2.58	C2-P/I-92	4.53	
23	Dziugiai	3.40	O2-P/I-58	1.93	Anni	2.58	C1-P/I-25	4.52	
24	O1-P/I-63	3.40	O2-A/Dz-52	1.92	C2-A/Dz-85	2.55	C2-P/I-98	4.44	
25	O2-A/Dz-69	3.37	O2-P/I-40	1.92	C2-P/I-24	2.55	O1-A/Dz-12	4.44	
26	C2-P/I-40	3.37	Anni	1.92	O1-A/Dz-65	2.54	O1-A/Dz-99	4.42	
27	C1-P/I-35	3.32	O1-P/I-75	1.91	C2-P/I-40	2.53	C1-P/I-20	4.41	
28	O1-A/Dz-99	3.31	C1-P/I-20	1.90	C2-P/I-72	2.49	O1-P/I-75	4.39	
29	O2-P/I-97	3.27	O1-A/Dz-65	1.89	Dziugiai	2.45	O2-P/I-40	4.36	
30	C2-A/Dz-85	3.26	O2-P/I-91	1.87	C1-P/I-20	2.45	C2-P/I-40	4.34	
31	C1-P/I-25	3.22	C1-P/I-83	1.86	O1-P/I-75	2.41	O2-A/Dz-52	4.31	
32	C2-P/I-72	3.16	O2-A/Dz-49	1.86	O1-P/I-93	2.41	O2-P/I-91	4.25	
33	C1-P/I-32	3.16	C1-A/Dz-34	1.86	O1-A/Dz-99	2.32	O1-P/I-93	4.18	

Comparison of Selection Efficiency for Spring Barley (*Hordeum vulgare* L.)
under Organic and Conventional Farming Conditions

34	O1-P/I-86	3.12	C1-A/Dz-5	1.83	O2-A/Dz-49	2.30	O2-P/I-97	4.15
35	C2-P/I-92	3.08	C2-A/Dz-36	1.82	C2-P/I-92	2.28	O1-P/I-86	4.10
36	O1-P/I-34	3.06	O2-P/I-97	1.78	C1-P/I-83	2.22	C1-P/I-35	4.09
37	Idumeja	3.06	O1-A/Dz-99	1.75	O2-P/I-51	2.19	Dziugiai	4.09
38	C1-P/I-20	3.05	C2-P/I-24	1.75	O1-P/I-34	2.18	O2-A/Dz-69	4.08
39	O2-P/I-40	3.02	O2-A/Dz-69	1.74	C1-P/I-25	2.09	O2-P/I-51	4.03
40	Primus	2.97	Dziugiai	1.73	C2-P/I-98	2.08	C1-A/Dz-20	4.02
41	O2-P/I-51	2.93	Idumeja	1.69	O1-P/I-86	2.07	O2-A/Dz-49	3.98
42	O2-P/I-58	2.85	O1-A/Dz-79	1.67	O2-P/I-91	2.04	Primus	3.96
43	O2-P/I-91	2.85	Primus	1.56	O2-P/I-40	1.99	C2-P/I-24	3.91
44	C2-P/I-24	2.84	C1-A/Dz-20	1.52	Primus	1.93	C1-P/I-83	3.88

[†] Test site: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site. [‡] Selection site: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site.

[§] Cross: P/I = Primus /Idumeja, A/Dz = Anni/Dziugiai

[¶] Number of line.

Table S5. The ranking of lines of P/I cross between grain yield (Y) and weed suppression ability (WSA) at the organic (O1 and O2) test sites. Lines connect the top five lines for grain yield (Y) with the ranking position for the weed suppression ability (WSA) and vice versa.

Test site [†]									
O1					O2				
Y	Rank	Line	Line	Rank	WSA	Rank	Line	Rank	WSA
3.48	1	C2-P/I-98 [‡]	C1-P/I-83	1	32.8	1	C1-P/I-35	1	28.8
3.44	2	O1-P/I-93[#]	C1-P/I-35	2	20.8	2	O1-P/I-93	2	28.2
3.42	3	O1-P/I-75	O1-P/I-63	3	20.7	3	O1-P/I-63	3	28.0
3.41	4	O1-P/I-83	O1-P/I-93	4	20.6	4	C2-P/I-98	4	27.9
3.40	5	O1-P/I-63	O1-P/I-86	5	18.7	5	O2-P/I-51	5	27.4
3.37	6	C2-P/I-40	C2-P/I-24	6	17.5	6	O1-P/I-34	6	26.3
3.32	7	C1-P/I-35	O2-P/I-97	7	12.6	7	C1-P/I-25	7	26.1
3.27	8	O2-P/I-97	O1-P/I-75	8	12.1	8	C2-P/I-72	8	22.4
3.22	9	C1-P/I-25	C2-P/I-98	9	9.1	9	C1-P/I-32	9	21.9
3.16	10	C2-P/I-72	C2-P/I-72	10	8.7	10	O1-P/I-86	10	20.5
3.16	11	C1-P/I-32	C1-P/I-32	11	7.3	11	C2-P/I-92	11	19.8
3.12	12	O1-P/I-86	O2-P/I-58	12	3.9	12	C2-P/I-40	12	16.2
3.08	13	C2-P/I-92	O2-P/I-40	13	3.8	13	O2-P/I-58	13	15.0
3.06	14	O1-P/I-34	O2-P/I-91	14	3.1	14	O2-P/I-40	14	13.7
3.05	15	C1-P/I-20	O1-P/I-34	15	0.6	15	O1-P/I-75	15	12.4
3.02	16	O2-P/I-40	C1-P/I-20	16	-5.8	16	C1-P/I-20	16	9.9
2.93	17	O2-P/I-51	O2-P/I-51	17	-6.1	17	O2-P/I-91	17	8.7
2.85	18	O2-P/I-58	C2-P/I-40	18	-9.5	18	C1-P/I-83	18	7.7
2.85	19	O2-P/I-91	C1-P/I-25	19	-18.7	19	O2-P/I-97	19	4.1
2.84	20	C2-P/I-24	C2-P/I-92	20	-22.7	20	C2-P/I-24	20	-3.0
$R_s = 0.442$ ($p = 0.013$)					$R_s = -0.005$ ($p = 0.246$)				

[†] Test sites: O1 = organic institute site, O2 = organic farmer's site.

[‡] Selection site: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site.

[§] P/I = Primus/Idumeja cross.

[¶] Number of line

[#] Selected lines in bold are in the top five for Y as well as for WSA (per site)

^{††} Spearman's rank correlation coefficient

Table S6. The ranking of lines of A/Dz cross between grain yield (Y) and weed suppression ability (WSA) at the organic (O1 and O2) test sites. Lines connect the top five lines for grain yield (Y) with the ranking position for the weed suppression ability (WSA) and vice versa.

Test site [†]									
O1					O2				
Y	Rank	Line	Line	Rank	WSA	Rank	Line	Line	Rank
3.94	1	C1-A/Dz-34 [‡]	C1-A/Dz-52	1	45.8	1	C2-A/Dz-66	O2-A/Dz-52	1
3.93	2	C1-A/Dz-52 [#]	O2-A/Dz-49	2	42.4	2	C2-A/Dz-37	O2-A/Dz-49	2
3.90	3	O1-A/Dz-12	O2-A/Dz-69	3	38.1	3	C2-A/Dz-84	C2-A/Dz-37	3
3.89	4	O1-A/Dz-92	O2-A/Dz-54	4	33.1	4	C1-A/Dz-52	C1-A/Dz-52	4
3.89	5	C1-A/Dz-62	O2-A/Dz-52	5	25.9	5	O1-A/Dz-12	O2-A/Dz-69	5
3.85	6	O1-A/Dz-79	O1-A/Dz-92	6	22.2	6	O2-A/Dz-57	O2-A/Dz-54	6
3.83	7	C1-A/Dz-5	O1-A/Dz-99	8	19.4	7	C2-A/Dz-85	O1-A/Dz-92	7
3.76	8	O1-A/Dz-65	C1-A/Dz-5	9	14.0	8	O2-A/Dz-54	C2-A/Dz-85	9
3.69	9	O2-A/Dz-57	O1-A/Dz-12	10	7.8	9	O1-A/Dz-92	C2-A/Dz-66	10
3.63	10	C2-A/Dz-36	C2-A/Dz-37	11	5.6	10	C1-A/Dz-62	O1-A/Dz-12	11
3.62	11	C2-A/Dz-66	C2-A/Dz-85	12	-0.3	11	O2-A/Dz-52	O1-A/Dz-99	12
3.58	12	O2-A/Dz-49	C2-A/Dz-66	13	-15.8	13	O1-A/Dz-65	C1-A/Dz-5	13
3.57	13	O2-A/Dz-54	O2-A/Dz-57	14	-29.4	14	O2-A/Dz-49	O2-A/Dz-57	14
3.55	15	O2-A/Dz-52	C1-A/Dz-20	15	-30.4	15	C1-A/Dz-34	C1-A/Dz-34	15
3.54	16	C2-A/Dz-37	O1-A/Dz-79	16	-39.0	16	C1-A/Dz-5	C2-A/Dz-84	16
3.54	17	C1-A/Dz-20	C1-A/Dz-62	17	-39.5	17	C2-A/Dz-36	O1-A/Dz-79	17
3.52	18	C2-A/Dz-84	O1-A/Dz-65	18	-43.4	18	O1-A/Dz-99	C1-A/Dz-20	18
3.37	20	O2-A/Dz-69	C1-A/Dz-34	19	-52.7	19	O2-A/Dz-69	C1-A/Dz-62	19
3.31	21	O1-A/Dz-99	C2-A/Dz-36	20	-55.1	21	O1-A/Dz-79	O1-A/Dz-65	20
3.26	22	C2-A/Dz-85	C2-A/Dz-84	21	-56.6	22	C1-A/Dz-20	C2-A/Dz-36	21
$R_s = -0.099$ ($p = 0.168$)					$R_s = 0.313$ ($p = 0.044$)				

[†] Test sites: O1 = organic institute site, O2 = organic farmer's site.

[‡] Selection site: O1 = organic institute site, O2 = organic farmer's site, C1 = conventional medium input site, C2 = conventional high input site.

[§] A/Dz = Ami/Dziugiai cross.

[#] Number of line

^{*} Selected lines in bold are in the top five for Y as well as for WSA (per site)

^{††} Spearman's rank correlation coefficient

Chapter 5

Effect of four selection procedures conducted in conventional and organic management on organic barley productivity and weed suppressive ability

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Abstract

The use of varieties with high competitiveness against weeds is of high importance in organic crop production. Breeding programs aiming at developing varieties adapted to organic farming should consider weed suppressive ability (WSA) as key selection criterion beside grain yield (GY). The aim of this study was to analyse under which conditions and with which selection procedure the best selection is achieved to improve both GY and WSA for organic farming systems. We used data on the performance of 134 barley genotypes in two organic and two conventional sites over three years in all possible year orders (six) to perform direct and indirect selection procedures in this panel according to selection criteria that took GY and WSA into account, either alone or in combination. We found that selection at organically managed sites was the most effective to identify highly suitable genotypes for organic farming with respect to both GY and WSA. The selection procedure where mild selection for weed suppressive ability was followed by strict selection for grain yield gave the best result. Selection of genotypes which combined high GY and WSA was also possible under conventional conditions. However, this occurred less frequently and the fraction of selected genotypes that performed well for both traits was smaller than by direct selection under organic conditions. Selection based on organic ideotype scores (OIS), comprising various important traits for organic farming including GY and WSA, resulted in the highest number of well-performing genotypes for grain yield and weed suppressive ability in both organically and in conventionally managed selection fields in comparison to two-stage selection on GY and WSA.

Keywords:

breeding for organic farming, grain yield, weed suppressive ability, direct and indirect selection, organic ideotype score

5.1 Introduction

Plant breeding aimed specifically at organic farming systems is a relatively new field of research. Organic farms represent a much more heterogeneous population of target environments than conventional farms that have options to mask negative growing conditions with high inputs of mineral fertilisers and crop protectants (Wolfe et al., 2008; Dawson & Goldringer, 2009). Organic farming refrains from chemical-synthetic inputs, and challenges growers to deal with different limiting factors such as irregular nitrogen availability from organic fertilisers, as mineralisation depends on soil microbial activity and soil temperature. Also the competition with weeds for resources and stress resulting from diseases might lead to a less stable yield across years and can affect grain quality such as thousand grain weight and grain protein content (Wolfe et al., 2008; Hoad et al., 2008; Kristensen & Ericson, 2008; Lammerts van Bueren et al., 2011). Nutrient uptake and nutrient use efficiency, weed suppressive ability (WSA) and disease resistances are therefore of high priority in breeding programs for organic farming and this requires a different breeding approach compared to breeding for conventional, high input systems (Lammerts van Bueren et al., 2002; 2011; Osman et al., 2016).

The ability to suppress weeds in a cereal crop depends on various plant traits, such as plant growth habit, early vigour, canopy height at early growing stage, crop ground cover, plant canopy growth habit, the length and width of the flag leaf, leaf area index, leaf inclination angle and plant height at harvest and their interaction (Lemerle et al., 1996; Liebman & Davis, 2000; Davies et al., 2004; Bertholdsson, 2005; Hoad et al., 2005; Köpke, 2005; Mason et al., 2008; Wolfe et al., 2008; Mahajan & Chauhan, 2013; Piliksere et al., 2013; Worthington & Reberg-Horton, 2013; Andrew et al., 2015; Kissing Kucek et al., 2021). The high WSA also helps to diminish the negative impact of weeds on crop yields as well as to decrease the weed seed bank in the soil for the following crops (Harker & O'donovan, 2013; Lutman et al., 2013; Mahajan et al., 2020).

Solutions have been sought for an effective and robust assessment method of WSA that could be used for as an important tool in genotype selection and variety testing. For example, Mahajan et al. (2020) suggested that plant height and high panicle production are desirable traits for weed suppression ability

in barley genotypes. Hansen et al. (2008) found a valuable suppressive index for barley variety screening, which includes growth traits such as leaf area index, reflectance, leaf angle, and culm length.

Yield potential and stability are also important in organic crop production. Therefore, organic breeding programs should use the combination of adequate weed suppressive ability and yield potential as key selection objective (Feledyn-Szewczyk & Jończyk (2017). However, Andrew et al. (2015) pointed at a possible trade-off between GY and WSA, especially in weed free conditions. Also Lazzaro et al. (2019) reported in their experiment with a wide set of wheat accessions a trade-off between GY and traits related to WSA under integrated weed management conditions.

In our previous breeding experiment with two barley populations at two organically and two conventionally managed sites (Kokare et al., 2017, Chapter 4), the results suggested that in a conventionally managed selection environment with high input, the correlation between the GY and WSA was negative compared to the selection in a medium input, conventional selection environment and under organic conditions. In the nutrient poor organic environment without weed control, a focus on weed suppressive, vegetative characteristics of the plants such as greater canopy height at stem elongation, dense crop ground cover and greater plant height (at harvest time), led to a high WSA. However, the GY of these genotypes was lower than for the genotypes selected under more optimal growing conditions. This indicates a possible trade-off between GY and WSA when selecting genotypes under extreme and variable organic conditions as well as under high input conventional conditions, in which chemical weed control is combined with high nitrogen input. On the other hand, Löschenberger et al. (2008) and Miko et al. (2014) observed that conventionally managed conditions may be used for assessing and selecting qualitative traits which are highly heritable such as soil coverage at end of tillering and at booting stage, but that these conditions would not be useful for quantitative traits such as GY which are more affected by environmental effects and genotype \times environment interactions.

Literature is not conclusive on the most appropriate selection environments and criteria for varieties adapted to organic farming conditions. Therefore, the aim of this study was to find out what selection procedure provides the best combination of both GY and WSA for genotypes intended to grow in organic

farming systems. For this study, we used experimental data of a three year variety trial with 134 barley genotypes in two organic and two conventional sites to perform direct and indirect selection procedures in this panel.

5.2 Materials and methods

Experimental sites and growing conditions

The dataset for our study is based on the performances of spring barley trials carried out in two management systems, conventional (C) and organic (O) during three seasons, 2010-2011-2012. Within each management system there were two sites. All fields were located within a radius of 5 km of each other and were chosen for their different management. The largest differences between the two organic sites were in weed management practices (Supplemental Table S1). For weed management, harrowing was applied in O1, while in O2, which was a farmer's field, no weed control measure was used, resulting in high weed pressure. Over three years, the weed ground cover at the barley stem elongation stage in the organic site O1 was in average 8% at O1 and 17% at O2 organic site respectively. O2 was mainly dominated by perennial weeds, but in the research centre's O1 site mainly annual weeds were present. The most important difference among the two conventionally managed sites was in the level of fertilization: in C1 a medium level and in C2 a relatively high level of mineral N fertilisers were applied (Supplemental Table S1). In both C1 and C2 herbicides were applied against weeds.

Both 2010 and 2011 had warm summers. In the second part of the growing period, July and August 2010, the mean air temperatures considerably exceeded the long term average. (Supplemental Figure S1). In site C1 (2010) an unusual incidence of Barley Yellow Dwarf Virus (BYDV) occurred. On average, the infection level with BYDV was 1.1 per plot, with a min value of 0 and a max of 8. (scored from 0 - no infection to 9 – high infection). In 2011, there was a dry spell during three weeks at the beginning of the growth period, from the middle of April until the middle of May. In site O2 (2011) substantial damage caused by cockchafer (*Melolontha melolontha*) was observed. The degree of damage was estimated in scores (0 – no beetle larvae damage, 5- all plants are damaged). The average damage per plot scored 3, which fluctuated from 0.5 to 4.5 over the plots.

In 2012 during the whole growing period the air temperature was close to the long term average, but in July the precipitation considerably exceeded the long term average.

Genetic resources

For this experiment a panel consisting of 134 spring barley genotypes was used. The panel consisted of genotypes from various gene banks and other breeders plus local breeding material (Supplemental Table S2). Out of the total number of genotypes, 94 were of Latvian origin (of which 24 varieties bred in the period from 1930 to 2011 and 70 breeding lines).

Experimental design and multiple use of the data from three years

The 134 genotypes were sown at each of the four sites in each of the three years in unreplicated plots of 3.7 m² in a completely randomized design where randomization was independent per year-location combination; mimicking selection performed in the early breeding stages when the amount of breeding material often is too limited to have replicates. We carried out selection based on the experimental dataset of the three years' performance of the 134 genotypes in four sites (twelve environments in total). We considered the years as independent environments, allowing us to use the three years' data per site in all possible (six) year orders: 2010/2011/2012; 2010/2012/2011; 2011/2010/2012; 2011/2012/2010; 2012/2010/2011; 2012/2011/2010. These data combinations allowed us to make the results more robust against specific conditions, with the presence of biotic and abiotic stress, under which mild and strict selection were done and the specific conditions of the evaluation year. They act as repetitions of the selection procedures under different conditions. In each year order, we performed selection in the first two years at the respective organic and conventional sites, and in the third year we evaluated the selection results by using the performance data of these selected genotypes at each of the organic sites in that third year. Each year was used twice to carry out the "first year" of selection, twice to carry out the "second year" of selection and twice to carry out the "third year" for evaluation at the organically managed sites of the genotypes selected in the other two selection years.

Field observations and traits evaluated

At each test site/year combination the grain yield (GY) and a number of traits contributing to WSA were evaluated. As weed competitiveness could play a crucial role in the early growth period, we focused on canopy height at stem elongation stage (CH) and crop ground cover (CGC) to help limit weed establishment. Additionally, we took into account the traits which could help to withstand weeds from the heading/flowering stage to later stages, such as the length and width of the flag leaf (LFL and WFL), plant height before harvest (PH), and resistance to lodging (LOD). The details on the evaluation methodology are presented in Table 5.1 (based on Kokare et al., 2017, Chapter 4).

The WSA index was calculated from the above mentioned traits. Each trait was first standardized by autoscaling where: $S_i = \frac{(x_i - \bar{x})}{\sigma}$, with: [1]

S_i - is the standardized trait value for individual genotype i in an environment (test site/year combination)

x_i – the trait value of individual genotype i in an environment (test site/year combination)

\bar{x} – the mean of the trait over all genotypes in that environment,

σ – the standard deviation of the trait values over all genotypes in that environment.

The standardization allows a set of desired traits with different units to be combined in a WSA index, see Table 1.

After that, the WSA over all traits considered was calculated for each genotype i at each environment, based on the following formula:

$$WSA_i = b_1 S_{1i} + b_2 S_{2i} + \dots b_t S_{ti} \quad [2]$$

where the b 's are the relative weights given to traits $1, 2, \dots, t$ (see Kokare et al., 2017) and S_i is the observed standardized trait value for an individual genotype i .

After that, WSA was standardized as

$$WSA_i = \frac{(WSA_i - \overline{WSA})}{\sigma}, \text{ where} \quad [3]$$

WSA_i – the WSA of individual genotype in an environment (test site/year combination)

\overline{WSA} – mean of the WSA over all genotypes in that environment (which is 0),

σ – the standard deviation of the WSA values over all genotypes in that environment.

The organic ideotype scores (OIS) was developed in Kokare et al. (2017) (see Chapter 4). The OIS was calculated by applying autoscaling in the same way as WSA (see [1] and [2], but with the traits and weights mentioned in Table 5.1.

Weed ground cover (WEED) was measured as a visual assessment of the percentage of the plot area covered by weed plants at the crop tillering stage as another estimate of weed suppressive ability by the barley genotypes.

Applied selection procedures

At all four sites the selection was performed with the aim to select genotypes suitable for organic farming. The selection at the organic sites (O1 and O2) was considered direct selection, and selection at the conventional sites (C1 and C2) was considered indirect selection.

The selection of genotypes was focused on the two traits GY and WSA. Four selection procedures were applied to select genotypes suitable for organic farming, out of the dataset of 134 barley genotypes grown each year: 1) selection for GY alone (GY); 2) selection for WSA in combination with more strict selection for GY (WSA+GY); 3) selection for GY in combination with more strict selection for WSA (GY+WSA); and 4) selection for the OIS.

1 - GY. In the first year at each of the four growing sites the 20 highest yielding genotypes were selected out of the 134 genotypes. In the second year, from these 20 genotypes per growing site, at each site the 10 genotypes with the highest GY were selected.

2 and 3 - GY and WSA.

The combination of these two criteria in one genotype is considered highly relevant for organic farming. Two different selection procedures were applied to find out the best method to avoid undesired trade-offs while selecting for both GY and WSA:

In the first year, we selected at each growing site, out of the 134 genotypes those that had moderate to high WSA (higher than the average over all

134 genotypes) and then, out of that selection the twenty with the highest GY. In the second year, we selected at the same site, from those 20 the ten genotypes with the highest GY. We refer to this procedure as WSA+GY

Alternatively:

- 2) In the first year we first selected at each growing site, out of the 134 genotypes the moderately to high yielding genotypes (GY higher than the average of all 134 genotypes) and then, out of that selection the twenty with the highest WSA. In the second year, we selected at the same site, from those 20 the ten genotypes with the highest WSA. We refer to this procedure as GY+WSA.

In these two procedures WSA+GY and GY+WSA, we consider the first criterion (i.e. higher value than the average over 134 genotypes) to ensure that the material will be “reasonably good” for the respective aspect, while the second criterion (the top twenty and top ten) will imply a strict selection for that criterion. So in WSA+GY, the strict selection is for GY; in GY+WSA, the strict selection is for WSA.

4 – OIS. In the first year at each of the four growing sites out of the 134 genotypes, the 20 genotypes with the highest OIS were selected. In the second year from these 20 genotypes per growing site, at each site the 10 genotypes with the highest OIS were selected.

In the third year, the performances of the ten best genotypes from each selection procedure (GY, WSA+GY, GY+WSA and OIS) per selection site (O1, O2, C1, C2) were compared at both organic evaluation sites (O1 and O2). Many genotypes were selected at multiple environments and by multiple selection procedures.

For GY, the gain by selection was expressed as relative difference of the mean GY of 10 selected genotypes and the overall mean GY over all 134 genotypes; for WSA, the gain by selection was expressed as the difference of the average value of WSA of 10 selected genotypes and the average value over the total set of 134 genotypes (which is equal to 0). A WSA value above the average WSA value of all 134 genotypes (> 0) is considered as high WSA (higher than average), whereas a negative WSA value is considered as low (lower than average).

Table 5.1 Traits included in the organic ideotype score (OIS) (2010–2012), and the relative weight (%) of each trait included in OIS, adapted from Kokare et al. (2017)

Traits	Abbreviation	Growing stage (GS) according to Zadoks et al. (1974)	Evaluation methodology	Relative weight (%) when included in the Organic Ideotype Score (OIS) ¹
Grain yield	GY	After harvest	Measured per full plot (3.7 m ²), the yield was expressed in tonnes ha ⁻¹ after drying and cleaning with 1.8 mm sieve	40
Traits contributing to suppressive ability against weeds (WSA)				
Canopy height at stem elongation stage	CH	GS 31–32	Measured in cm from the soil to the top of the leaf canopy. One measurement in each plot	20
Crop ground cover	CGC	GS 31–32	Visually estimated % of plot area covered by plants	10
Width of flag leaves	WFL	GS 47–51	Leaves for 5 plants in each plot were measured in cm and mean was calculated	2
Length of flag leaves	LFL	GS 47–51	Leaves for 5 plants in each plot were measured in cm and mean was calculated	2
Lodging resistance	LOD	GS 90	Visually estimated and scored from 1 (low) to 9 (high)	2
Plant height at maturity	PH	GS 90	Five plants per plot in cm were measured and the mean has calculated	6
Traits related to growing period (GP)				
Thermal time to heading	HED	GS 51	Days from sowing to heading were estimated and the thermal time expressed in cumulated degree days was calculated	5
Thermal time to maturity	MAT	GS 90	Days from sowing to full ripening (maturity) were estimated and the thermal time expressed in cumulated degree days was calculated	5
Diseases resistance (DR)				
Resistance to leaf diseases (powdery mildew and net blotch)	DR	From GS 32 onwards	Visually estimated and scored from 1 (low resistance) to 9 (high resistance)	6
Traits related to grain quality (Q)				
Volume weight	VOL	After harvest	Volume weight g L ⁻¹ determined by Infratec Analyser 1241 (Foss, Högenås, Sweden)	2
				Total 100

Statistical analysis

The phenotypic data were summarized using descriptive statistics. An analysis with a linear mixed model was performed using REML. In that analysis all terms were random and it was only used to quantify the relative sizes of the variance components for phenotypic traits. REML model included genotypic main effects (G) and relevant environmental effects of: management system (M); year (Y); growing site (M/S) within a management system as well as the interactions of genotype with year (G×Y), management system (G×M) and growing site within a management system (G×M/S), and a residual term e:

$$y = \mu + G + M + Y + (M/S) + M \times Y + (M/S) \times Y + G \times M + G \times Y + G \times (M/S) + G \times M \times Y + G \times Y \times (M/S) + e.$$

A two-sample t-test was used to compare differences between the mean of the selected set of genotypes according to each selection procedure and the mean of the original population of 134 genotypes in each testing site separately. Pearson correlation coefficients were used to quantify the relationships between traits at each growing site.

5.3 Results

Grain yield and Weed suppressive ability.

The average GY under conventional conditions was higher by approximately 56% compared to organic conditions (Supplemental Table S3). The management system was the largest variance component for GY (44.7%), followed by growing site-by-year interaction (M/S×Y) (22.6%). The genotype effect was highly significant ($p < 0.001$), but explained only 4.6% of the variation in GY, some of the environmental variances were much larger (Supplemental Table S3). Therefore, we have to consider that it will be challenging to select the same set of high-yielding genotypes across different environments. The genotypes would respond differently to differences in growing sites and years in terms of their yield.

In contrast, for most of the traits contributing to the WSA: CH, WFL, PH, but not CGC and LFL, the variation is mainly explained by the genotype (Supplemental Table S3). For CGC we found considerable variation due to the management system. The management system was also the main source of variation for the traits contributing to GP, viz. HED and MAT. In terms of

HED and MAT the genotypes headed and matured later under organic than under conventional environments (Table 5.2). The year effect was large for MAT (close to 38% of the variance) and for VOL (21.7%). For VOL, the genotype main effect explains a large proportion of variation (26.3%).

The correlation across the 134 barley genotypes between the growing sites for GY was positive (0.16 - 0.31) (Supplemental Table S4a). The highest correlation coefficients were between the organic site O1 and the two conventional sites C1 and C2. However, the weakest correlation was observed between the farmer's site O2 and the other sites. For most traits contributing to WSA, but not for CGC and LFL, the correlation coefficient across genotypes between the organic and conventional sites was high (up to 0.79).

The yield level within the growing sites fluctuated considerably over the years (Table 5.2) In our experiment, the average GY was the highest at high input site C2, with the highest value 5.52 t ha⁻¹ in 2011 and the lowest 4.60 t ha⁻¹ in 2012. The lowest GY was at O2 and fluctuated from 0.92 t ha⁻¹ in 2011 to 2.89 t ha⁻¹ in 2010). Besides, the lack of rank correlations for the genotypes for yield at O2 between the years (Supplemental Table S4b) indicates considerable changes in variety ranking between environments. The rank correlation among the environments at both organic sites O1 and O2 was relatively low. This is because, the two organic sites differed in crop rotation, fertilization, and weed management, and additional unforeseen circumstances (e.g. diseases, bad weather) affected barley crop development, resulting in large yield differences in some years.

The genotypes ranks between environments are changing to a large extent, which indicates that performance of superior genotypes in one environment could be unreliable in another.

Relation between the traits contributing to weed suppressive ability, weed ground cover and grain yield

Average weed ground cover (WEED) was higher at O2 than at O1 in the years of testing: 12% in 2010, 6% in 2011, and 22% in 2012 at O2, and 13.1%, 5%, and 6%, respectively at O1. Most of the barley traits contributing to WSA tended to correlate negatively with WEED at both organic sites (Supplemental Figure S3).

The correlation analysis between WSA and the individual traits contributing to WSA and GY showed that CH and CGC correlated positively with GY at both organic sites (Supplemental Figure S4). Similar correlations were observed in the conventional site C1 with medium input, whereas in the conventional, high input field C2 the correlations between GY and the traits associated with WSA were lower, and for WFL and PH even slightly negative. WSA correlated significantly and positively with GY at O1 and O2, as well as at conventional site C1. The correlation between WSA and GY was consistent with the correlations of traits CH and CGC with GY which could be explained by the relatively high weight given to these traits (CH 20% and CGC 10%) in our WSA index.

Table 5.2 Mean values and standard error for the traits of 134 barley varieties grown at two organic sites (O1 and O2) and at two conventional sites (C1 and C2), in 2010, 2011, 2012

Environment	Traits																	
	GY	s.e.	CH	s.e.	SSH	s.e.	LFL	s.e.	WFL	s.e.	PH	s.e.	HED	s.e.	MAT	s.e.	VOL	s.e.
O1_2010	3.11	0.06	25.46	0.47	33.85	0.77	11.51	0.17	7.56	0.15	76.52	1.19	63.37	0.20	96.34	0.19	63.67	0.31
O2_2010	2.89	0.06	21.55	0.39	25.99	0.53	11.85	0.16	8.82	0.14	75.13	1.07	66.07	0.19	98.96	0.19	65.47	0.28
C1_2010	2.82	0.08	26.63	0.47	38.62	0.92	11.74	0.18	8.36	0.14	80.22	1.25	56.11	0.22	87.57	0.19	67.2	0.21
C2_2010	4.29	0.08	28.01	0.41	47.44	0.86	14.88	0.22	9.66	0.15	92.60	0.87	54.60	0.22	87.94	0.21	68.07	0.19
O1_2011	3.65	0.07	16.64	0.33	24.46	0.71	10.89	0.16	7.80	0.13	70.93	0.93	59.48	0.25	97.06	0.19	69.33	0.17
O2_2011	0.92	0.05	16.46	0.34	22.24	0.61	10.04	0.15	7.32	0.13	58.06	0.76	63.11	0.29	95.19	0.16	67.65	0.18
C1_2011	3.80	0.07	21.72	0.47	53.81	1.47	10.86	0.15	7.99	0.13	78.37	0.98	55.16	0.24	90.87	0.22	68.31	0.17
C2_2011	5.52	0.08	21.62	0.58	53.51	1.56	10.94	0.16	8.02	0.13	73.07	0.92	54.17	0.23	90.93	0.25	-	-
O1_2012	1.70	0.05	15.34	0.40	17.39	0.61	10.96	0.16	7.75	0.14	76.22	0.85	63.44	0.26	104.35	0.25	69.92	0.20
O2_2012	1.81	0.06	19.19	0.50	20.41	0.61	9.97	0.17	7.48	0.13	74.28	0.92	63.66	0.25	106.20	0.33	69.59	0.15
C1_2012	4.10	0.08	23.54	0.65	33.58	0.93	11.74	0.15	8.65	0.14	92.42	1.06	58.80	0.23	99.07	0.31	68.13	0.20
C2_2012	4.60	0.07	24.33	0.64	37.91	0.69	9.76	0.13	7.55	0.12	78.44	1.01	53.57	0.19	96.96	0.37	70.39	0.14

^a The values expressed: GY = grain yield, t ha⁻¹; CH = canopy height, cm; CGC = crop ground cover, %; LFL = length of flag leaf, cm; WFL = width of the flag leaf, mm; PH = plant height before harvest, cm; HED = days from sowing to heading; MAT = days from sowing to maturity; VOL = volume weight g l⁻¹

^b Mean of the trait is calculated over 134 genotypes.

The effect of evaluation year on selection results for high grain yield and weed suppressive ability

The performances of genotypes in the third year in each combination of three years were used to quantify the yield increase at both organic sites obtained by the different selection procedures applied (based on the first two years of each of the year combinations) at the selection sites under organic and conventional conditions. The results for GY are presented in supplemental Table S5, in which GY is calculated as the relative difference of the mean GY of the 10 selected genotypes and the overall mean GY over all 134 genotypes included in the experiment (100%).

Across all procedures, two year orders resulted more frequently in a significant increase in GY by selection at the O1 site: 10/12/11 and, to a lesser extent 12/10/11 (Supplemental Table S5). This may be because the evaluation year 2011 was the most productive year for O1. When the lowest yielding year for O1 (2012) was the evaluation year, the least number of procedures resulted in a significant gain in GY by selection when selected under O1. When 2012 was one of the selection years under O1, then the gain by selection was moderate to good when evaluated at O1.

The pattern was slightly different at the O2 evaluation site. Here the year order 10/11/12 stood out in which almost all procedures resulted in a significant increase in GY and also for 11/10/12 (Table 5.3). In both year orders, the evaluation year 2012 had a moderate yield level at O2 (Table 5.2). However, no significant GY (per selection site/procedure) increase was obtained in those year orders where 2011 was the evaluation year.

The year orders 11/12/10 and 12/11/10 stood out for many negative values at both organic evaluation sites across all procedures implying that the selected set of genotypes evaluated in 2010 yielded lower than all 134 genotypes tested. When selection was carried out under conventional conditions, the year orders influenced the increase in GY at organic evaluation sites O1 and O2 to a lesser extent compared to direct selection.

The average values of WSA of the 10 selected genotypes obtained by four selection procedures at four selection sites in the six-year orders are presented in Table S6 (Supplemental material). We compared the difference between the average value of WSA of the 10 selected genotypes and the average value

of the total set of 134 genotypes (which is equal to 0). WSA values ranged from a score of 1.53 (for the genotype with the highest WSA) to -2.09 (for the genotype with the lowest WSA) at O1, and from 1.63 to -2.26 at O2 (Supplemental Table S2).

The same year orders as for GY turned out to be particularly effective for the direct selection for high WSA: 10/12/11 and 12/10/11 for evaluation site O1 and 10/11/12 and 11/10/12 for evaluation site O2 (Supplemental Table S6). The year order 11/12/10 mainly led to no significant increase or even negative results in WSA when selection was performed under O2 or conventional conditions.

The selection for merely GY at conventional high input site C2 resulted in negative WSA values at organic evaluation sites in almost all year orders; in two year orders 11/10/12 and 12/10/11, WSA was even significantly lower than at organic site O1

The efficiency of selection per site

The results presented in Supplemental Table S5 and S6 are summarized in Table 3 which shows in how many of the six different year orders the average GY, WSA, and both these traits simultaneously (GY/WSA) of the ten selected barley genotypes per each selection procedure was in the third (evaluation) year significantly higher than the average GY and WSA of all 134 barley genotypes.

Table 5.3 The number of year orders out of six in which the average grain yield (GY) and weed suppressive ability (WSA) of the ten selected barley genotypes was in the third year significantly higher than the average GY and WSA of all 134 barley genotypes. GY/WSA presents the number of year orders in which the selected set of ten genotypes had both a significantly higher GY and a significantly higher WSA than the average of all 134 genotypes, evaluated at two organic sites (O1 and O2) obtained in direct (at two organic sites O1 and O2) and indirect (at two conventional C1 and C2 sites) selection according to four selection procedures (GY; WSA+GY; GY+WSA; OIS)^a, based on Supplemental Table S3, S4

Selection traits	Selection procedure	Evaluation site O1				Average across all selection sites	Evaluation site O2				Average across all selection sites
		Selection site					Selection site				
		Direct		Indirect			Direct		Indirect		
		O1	O2	C1	C2		O1	O2	C1	C2	
GY	GY	2	3	2	1	2	2	2	0	2	2
	WSA+GY	2	3	2	2	2	3	2	1	0	2
	GY+WSA	3	0	0	0	1	1	1	1	2	1
	OIS	5	4	0	1	3	1	1	1	2	1
Average across the procedures		3	3	1	1		2	2	1	2	
WSA	GY	1	1	0	0 (-2) ^b	1	1	0	0	0	0
	WSA+GY	4	3	2	0	2	4	2	1	1	2
	GY+WSA	6	5	6	5	5	6	4	4	4	5
	OIS	6	4	2	1	3	3	2	2	4	3
Average across the procedures		4	3	2	2		4	2	2	2	
GY/WS	GY	1	1	0	0	1	1	0	0	0	0
	WSA+GY	2	3	1	0	2	3	1	1	0	1
	GY+WSA	3	0	0	0	1	1	1	0	1	1
	OIS	5	4	0	0	2	1	1	1	1	1
Average across the procedures		3	2	0	0		2	1	1	1	

^a GY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score

^b In brackets shown the number of year ranks when the WSA value of the 10 selected genotypes was significantly lower than the average of all 134 genotypes

Direct versus indirect selection

Across all procedures, the direct selection at organic sites gave in more year orders a significant increase in GY (on average three out of six year orders over all procedures at both organic selection sites versus one year order over all procedures at both conventional selection sites) and in WSA (on average

three to four out of six year orders) when evaluated at O1 than indirect selection at conventionally managed sites (two to three out of six year orders) (Table 5.3). A similar tendency, but less clearly so, was seen at evaluation site O2. The direct selection at the more optimally managed O1 site was more effective than indirect selection to achieve a higher increase of both traits (GY/WSA): three to four versus zero year orders out of six at evaluation site O1 and two versus one year order at evaluation site O2. Within the organic management system, the selection at O1 tended in more year orders than the selection at O2 to result in a significant improvement in GY and WSA.

A two-sample t-test was performed to test whether the differences of GY and WSA obtained by direct selection at the respective organic site and other selection sites were significant per each selection procedure (Supplemental Table S7 and Table S8).

There were no significant differences in the average GY and WSA between the set of ten genotypes selected at organic site O1 and the ten genotypes selected at other places (at another organic O2, conventional C1, and C2) in most of the year orders when evaluated at organic site O1. However, in three-year orders, the average WSA of the ten genotypes selected for merely GY at the conventional C2 site was significantly lower than for the ten genotypes selected at evaluation site O1, but did not differ from the genotypes selected at the organic evaluation site O2. According to the results of the comparison, in principle, the mean GY and WSA of the set of genotypes selected under conventional conditions did not differ from the set of directly selected genotypes under organic conditions, if in the selection, both GY and WSA were taken into account.

Selection procedures

When the selection was carried out at the organic sites, OIS was the selection procedure that more frequently resulted in a significant increase in GY at the organic evaluation site O1 (in four to five year orders out of six) than the other selection procedures. When the selection was carried out at conventionally managed sites, the procedure WSA+GY resulted most frequently (in two year orders) in a significant increase in GY at evaluation site O1 (Table 5.3). At O2, a significant increase in GY was achieved less frequently than at O1, with no clear difference between organic and conventional selection sites and no clear differences between the selection procedures.

To improve WSA, the selection for GY+WSA resulted in the highest number of year orders in which the selected genotypes had a significantly improved WSA, with not much difference between O sites and C sites (six versus five year orders for O/C when evaluated at O1 and four to six versus four year orders when evaluated at O2), see Table 5.3. The selection on GY alone carried out at organic and conventional sites resulted in the lowest number of year orders with significant improvement in WSA at both evaluation sites O1 and O2.

If the selection was aimed at reaching both a significantly higher GY and a significantly higher WSA (GY/WSA), then the direct selection at organic sites for OIS resulted in the highest number of successful average year orders (four to five) when evaluated at site O1; when evaluated at site O2, the highest number of year orders (three) in which the goal was achieved was based on the selection for WSA+GY carried out at organic site O1. Not one procedure under indirect selection at both conventional sites appeared very effective: the number of year orders with a significant increase in both GY and WSA at both organic evaluation sites was low (zero to one).

Selection of genotypes that combine high GY and good WSA

For each evaluation environment (site and year order combination) we determined how many of the 10 selected genotypes according to each of the selection procedures had both a GY and a WSA higher than the average of the set of 134 genotypes. Direct and indirect selection by the procedures GY+WSA and OIS appeared to have been the most effective, leading to the highest number of genotypes out of the 10 genotypes which performed high for GY and WSA (Table 5.4) at O1. For O2 the highest number of genotypes performing well for GY and WSA was obtained by applying the selection procedure WSA+GY.

Direct and indirect selection for merely GY was less effective than other selection procedures and resulted on average in the lowest number of genotypes that had higher than average GY and WSA of the set of 134 genotypes at the organic evaluation sites. There was an exception of direct selection carried out at farmers site O2, where the selection for merely GY was the most effective leading to the highest number of genotypes performing high for both traits when evaluated again at O2.

The frequency of the selected genotypes

In our study with six year orders, the frequency by which certain genotypes appear in the selection can show how consistently some genotypes are selected by certain selection procedures in a certain environment. We applied a threshold of being selected at least three times in six year orders at a site/procedure combination. These 'most frequently' selected genotypes are presented in Table 5.5 per selection site and selection procedure. Out of the set of 134 genotypes, 97 were selected at least in one combination of year orders/site/selection procedure. Out of those, 42 genotypes were selected at least in three out of six year orders in at least one of the selection procedures and selection site combinations.

The number of frequently selected genotypes listed in Table 5.5 was slightly higher when selection was performed under organic conditions than when carried out under conventional conditions. Besides, the most frequently selected individual genotypes differed between the selection sites with few genotypes in common. The most frequently selected genotypes at the organically managed sites were Druvis and Rubiola.

Table 5.4 The number of barley genotypes from among the 10 genotypes selected per selection site/year order and selection procedure) for grain yield (GY) and weed suppressive ability (WSA) that were higher for both traits than the average of 134 genotypes according to the selection procedures: GY, WSA+GY, GY+WSA and OIS.

Year order	Selection procedure	Evaluated in O1				Evaluated in O2			
		Selection procedure				Selection procedure			
		Direct		Indirect		Direct		Indirect	
		O1	O2	C1	C2	O1	O2	C1	C2
10/11/12 ^b	GY ^a	7	4	5	4	4	6	3	4
	WSA+GY	6	6	6	5	6	6	6	4
	GY+WSA	7	6	7	6	6	6	6	6
	OIS	6	7	4	5	7	4	5	5
10/12/11	GY	7	7	6	4	4	2	4	4
	WSA+GY	7	6	5	6	4	4	5	4
	GY+WSA	9	7	6	5	3	2	5	5
	OIS	7	8	6	4	3	1	2	5
11/10/12	GY	6	6	3	3	5	6	2	4
	WSA+GY	7	6	6	4	6	6	4	4
	GY+WSA	7	6	7	6	8	8	5	7
	OIS	9	8	5	6	6	7	2	5
11/12/10	GY	6	1	4	4	6	4	5	4
	WSA+GY	5	1	4	6	7	3	5	6
	GY+WSA	7	4	6	6	5	2	4	7
	OIS	7	3	4	5	5	3	5	6
12/10/11	GY	5	6	5	2	3	6	5	3
	WSA+GY	6	7	6	3	3	5	6	6
	GY+WSA	7	7	6	6	5	2	3	4
	OIS	7	6	6	5	4	5	5	5
12/11/10	GY	5	2	3	3	5	4	3	4
	WSA+GY	6	1	3	6	6	3	4	6
	GY+WSA	7	2	5	4	4	4	5	6
	OIS	8	1	5	5	4	4	4	7

	GY	6.0	4.3	4.3	3.3	4.5	4.7	3.7	3.8
Average over the year orders	WSA+GY	6.2	4.5	5.0	5.0	5.3	4.5	5.0	5.0
	GY+WSA	7.3	5.3	6.2	5.5	5.2	4.0	4.7	5.8
	OIS	7.3	5.5	5.0	5.0	4.8	4.0	3.8	5.5

^a GY = selection according to GY alone, GY+WSA= first mild selection for GY followed by strict selection for WSA, WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score

^b year order 10/11/12 indicates selection in 2010 and 2011 then final evaluation using the 2012 data

Of these, Druvis, which is one of the four six-row varieties, was particularly often selected at O1, and Rubiola about equally often at O1 and at O2. These two genotypes were frequently selected (at least 3×) by almost all selection procedures at both organic sites, but for none of the selection procedures at C1, and only for one procedure frequently selected at C2. Another striking genotype is Golf, which was selected by all four procedures at C1 in at least four year orders. The breeding line PR-4181 was most often selected at conventional high input site C2, but it was not among the frequently selected genotypes at other sites.

Within a management system/site combination, the most frequently selected genotypes often appeared in multiple selection procedures (Table 5.5; Supplemental Table S2). This finding suggests that the frequency of the genotypes to be selected did not mainly depend on particular selection procedures but mainly on the management system and site within that management system.

We ranked the 42 genotypes listed in Table 5 according to their total frequency of selection, i.e. including site/selection procedure combinations for which they were selected only in one or two year orders. The top 20 of these 42 frequently selected genotypes were further checked for their performance for high GY and high WSA relative to the 134 genotypes at both organic sites O1 and O2 (Table 5.6).

Effect of four selection procedures conducted in conventional and organic management on organic barley productivity and weed suppressive ability

Table 5.5 The most frequently (at least 3 times in six-year orders at a site/procedure combination) selected genotypes at the four selection sites (two organic O1 and O2 and two conventional C1 and C2) and by four selection procedures per site.

Selection site	Selection procedure	Names of individual genotypes selected at least 3 times in 6 year orders									
O1	GY ^a	Druvis (4x ^b)	Rubiola (4x)	PR-3282 (3x)	PR-5105 (3x)	Abava (3x)	PR-4814 (4x)	1079488-45 (4x)	Kristaps (4x)		
	WSA+GY	Druvis (6x)	Rubiola (4x)	PR-3282 (3x)	PR-5105 (3x)	Abava (3x)	PR-4814 (4x)				
	GY+WSA	Druvis (6x)	Rubiola (3x)	PR-3282 (6x)	Klinta (3x)	PR-3605 (6x)	827580-15 (3x)	PR-3297 (6x)	BZ12-63 (4x)		
	OIS	Druvis (6x)	H130 (4x)	PR-3282 (4x)	Klinta (3x)	PR-3605 (4x)	PR-4814 (6x)	Imula (4x)			
O2	GY	PR-3005 (4x)	Rubiola (4x)	Sencis (4x)	Klinta (3x)	PR-5135 (4x)	PR-3223 (3x)	PR-5105 (3x)	Tocada (4x)		
	WSA+GY	PR-3005 (6x)	Rubiola (3x)	12825 (3x)	Klinta (3x)	PR-5135 (4x)	PR-3223 (3x)	PR-4810 (3x)			
	GY+WSA	Dziugiai (4x)	H130 (4x)	12825 (3x)	BZ14-90 (4x)	L-2985_1 (4x)	Ula (3x)				
	OIS	PR-3005 (4x)	Rubiola (3x)	12825 (3x)	BZ14-90 (3x)	PR-4810 (4x)					
C1	GY	Golf (4x)	H130 (4x)	Leeni (4x)	Tocada (4x)	L-2735 (4x)					
	WSA+GY	Golf (4x)	H130 (5x)	L-2735 (4x)	Klinta (4x)	PR-5131 (3x)					
	GY+WSA	Golf (6x)	768678-28 (4x)	BZ12-63 (4x)	827580-15 (4x)	PR-5135 (3x)	PR-5117 (4x)	Alsa (3x)			
	OIS	Golf (6x)	768678-28 (3x)	BZ12-63 (4x)	Klinta (4x)	Dzintars (4x)	PR-5117 (4x)				
C2	GY	743-09 (6x)	PR-5135 (4x)	PR-4181 (6x)	Ula (3x)	PR-3515 (6x)	PR-3885 (4x)				
	WSA+GY	1163691-34 (4x)	Rubiola (3x)	PR-4181 (6x)	Ula (3x)	1267199-30 (3x)	Eunova (3x)				
	GY+WSA	1163691-34 (3x)	Abava (3x)	PR-4814 (4x)	Klinta (4x)						
	OIS	1163691-34 (4x)	PR-5135 (3x)	PR-4181 (4x)	Druvis (4x)	1272500-36 (3x)					

^a GY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score

^b The number shows the number of year orders in which the genotype was selected at that selection site by that selection procedure

Table 5.6 The performance of the most frequently selected genotypes at organic and conventional sites for GY and WSA at evaluation sites O1 and O2 during three testing years (2010, 2011, and 2012).

Most frequently selected genotypes ^a	Total times selected at O (max 48)	Total times selected at C (max 48)	Number of times (max 3) the GY and WSA of the genotype was higher than average at organic sites		Rank of average GY ^b	Rank of average WSA ^b	Rank of average GY ^b	Rank of average WSA ^b
					O1	O2	O1	O2
Druvis	24	8	3	1	1	4	55	37
Rubiola	23	8	3	3	5	13	4	26
PR-3005	20	5	3	3	2	64	3	41
H130	19	15	2	2	16	21	28	4
Klinta	17	22	2	2	27	6	12	33
PR-3282	17	7	3	1	9	10	87	14
PR-4814	16	8	3	1	4	9	57	53
PR-5105	16	0	3	2	8	32	8	24
PR-3605	13	11	3	2	21	1	40	9
PR-5135	11	18	1	3	85	25	1	35
BZ12-63	10	12	3	1	26	8	89	66
Abava	10	9	3	1	3	5	88	51
Sencis	9	9	0	0	22	90	9	59
827580-15	8	18	3	1	29	22	73	82
Ula	6	11	2	2	56	40	39	6
PR-5117	5	15	2	1	89	45	59	22
Golf	4	25	3	2	49	12	64	34
PR-4181	4	18	1	1	6	77	42	57
768678-28	4	14	1	0	35	37	110	94
L-2735	0	18	0	1	82	81	63	76

^aThe 20 most frequently selected genotypes over all selection procedures at organic and conventional selection sites

^bAveraged over all 134 genotypes, according Supplemental Table S2

The genotypes Druvis, Rubiola and PR 3005 were the most frequently selected under organic, and Golf, Klinta were most frequently selected under conventional conditions. The genotypes H130 and Klinta were selected with similar high frequency at both the organically and conventionally managed sites. The most frequently selected genotype over all environments (at organic and conventional sites) was 'Klinta'.

Genotypes selected frequently under organic conditions ranked higher for their average GY as well as for their average WSA than the genotypes particularly frequently selected under conventional conditions when tested at both organic O1 and O2 sites. The genotypes similarly frequently selected at both the organically and at the conventionally managed sites performed high in two out of three years of testing at O1 and O2 and ranked high for GY and WSA at both organic sites. However, they did not outperform for GY the genotypes most frequently selected under organic conditions, such as Rubiola and PR-3005. Furthermore, H130 and Klinta were mainly superior for GY and the traits with the highest weights in WSA, such as CH and CGC, than those most consistently selected under conventional sites (data not shown).

5.4 Discussion

Breeders aiming to develop varieties for the organic sector often face the question of whether they can use conventionally managed breeding fields to select varieties appropriate for organic conditions or whether they should invest in a parallel, organically managed breeding program. To address this question, we analyzed a dataset of three years of performance of 134 barley genotypes in two conventional and two organic environments. During two years, the selection was performed in all four environments, followed by one year of evaluation at both organic sites, and we repeated this procedure in six different year orders. We compared different selection procedures under direct and indirect selection and focused on two traits highly important for organic farming: grain yield (GY) and weed suppressive ability (WSA). We wanted to find out whether combining high GY and WSA is possible without a significant reduction in one of these traits, and which selection procedure carried out under direct and indirect selection provided best results concerning both these important traits.

How does the direct and indirect selection of barley genotypes affect traits relevant for performance in organic farming systems?

One of the reasons to do this more extensive experiment with a large set of genotypes is that the literature is not conclusive about whether a direct or indirect selection is more effective in developing cereal varieties suitable for organic farming.

The results of our experiment show that a significant increase in GY or WSA by the selection, as well as the increase in both these traits together, was more frequently achieved under direct selection than under indirect selection (Supplemental Tables S5, S6, Table 5.3). That is in line with the experimental evidence of several other authors that direct selection in the target environment is most effective for cereal varieties aimed at low-input or organic conditions (Ceccarelli et al., 1994; Brancourt-Hulmel et al., 2005; Murphy et al., 2007). It also confirms the results of our earlier selection experiment on two segregating populations (two crosses) aimed to obtain varieties suitable for organic farming comparing selection in two conventionally and two organically managed selection fields, where we found that the genotypes with the best combination of GY and WSA were selected under organic conditions (Kokare et al., 2017).

In addition, we observed differences for direct selection between the two quite different organic selection sites. The set of selected high-performing genotypes differed between both organic sites, only with some genotypes in common, which reflects the rather large genotype \times environment (year and growing site within the management ($G \times Y \times (M/S)$) interaction. For all procedures, a significant increase in GY and WSA occurred more frequently for the genotypes selected at organic site O1 than for those selected at the organic site O2. Besides, the selection in low-yielding environments (O2/2011 and O2/2012) in subsequent selection year orders (11/12/10 and 12/11/10) at organic O2 site led to significantly low GY or WSA or both, depending on the selection procedure, when evaluated in the more favourable year 2010, at organically managed O1 site (Supplemental Tables S5, S6). Also, a low number of high performing genotypes were selected in those unstable environments (O2/2011 and O2/2012) (Table 5.4). This can be explained by a lower heritability that often occurs under low-input conditions and high weed incidence. In that previous experiment carried out at the same organic

sites O1 and O2, the heritability observed for GY at O2 site was nearly half (24%) of the heritability observed at O1 site (45%) (Kokare et al., 2014; Chapter 2). We attained a similar result in another experiment with two segregating populations, where the selection in the organic commercial farm site with high weed incidence led to very high WSA but low GY in comparison to other selection sites when tested in a more optimal organic environment (Kokare et al., 2017). Therefore, selection conducted in poor and unstable or extreme environments may be less effective to obtain high yielding barley genotypes for more favourable organic farming conditions. For poor, more unfavorable conditions, such as organic site O2, decentralized selection for specific adaptation could be a useful approach (Annicchiarico et al., 2005; Ceccarelli & Grando, 2007; Desclaux et al., 2008; Dawson et al., 2008; Döring et al., 2011).

Which selection procedure is most effective to obtain genotypes with the combination of high grain yield and high weed suppressive ability?

Our results show that in general the direct and indirect selection according to the procedures WSA+GY, GY+WSA, and OIS were more effective than selection for GY alone for a significant increase in GY and WSA, in terms of the number of year orders (Table 5.3) as well as in terms of the number of selected genotypes that combined high GY and high WSA (Table 5.4) under organic conditions. This confirms that it is possible to select for both high grain yield and weed suppressive ability in barley, as was shown by other studies in wheat (Coleman et al., 2001; Bertholdsson et al., 2016).

Focusing merely on GY will not lead to significant improvement in WSA, and in the case of the C2 selection site, even to decrease in WSA. Although, the selection for only high GY may lead to lower WSA under organic conditions than when selection is also based on WSA characters, the number of selected genotypes that combine high GY and good WSA in direct selection procedures was slightly higher compared to the indirect selection results.

Direct selection

Our results show that selection under organic conditions more often resulted in the highest number of selected genotypes with a high GY and high WSA when the selection was performed for WSA+GY and OIS in comparison to

the selection for merely GY and GY+WSA (Table 5.3, 5.4). Such selected genotypes performed well for both WSA and GY at the optimally managed organic site O1 and at the low yielding organic site O2. Following the selection for merely GY, the selection carried out under organic conditions resulted more frequently in barley genotypes that were above average for WSA than selection for only GY at the conventionally managed high input selection site. This may be explained by the fact that the correlation coefficients between GY and the traits constituting the highest proportion in WSA, such as CH and CGC, were significantly positive at organic sites but not at conventional high input sites. This is in agreement with other studies in rice, wheat, and barley, where a more dense crop canopy resulted in less weed pressure and higher GY (Kruepl et al., 2007; Hoad et al., 2012; Worthington and Reberg-Horton, 2013; Mahajan et al., 2020).

Interestingly, our results show that the variety Tocada was among the frequently selected genotypes by selection for only GY at organic site O2 and conventional site C1 and had the second highest average GY at O2 of all barley genotypes, despite a low WSA at both organic sites. Its high GY may be due to 'weed tolerance' - the ability of the variety to maintain high GY despite the high presence of weeds (Lemerle et al., 2006; Fradgley et al., 2017; Mahajan et al., 2020). The fact that Tocada was quite an exception for having high GY despite low WSA at O2 suggests that the weed tolerance was not a common phenomenon in the set of barley genotypes used in this experiment. Therefore, selection for WSA+GY is presumably more effective than selection for merely GY in order to obtain barley varieties for organic farming, especially if the genotypes are targeted for more unfavourable conditions with a high presence of weeds.

The other two selection procedures (GY+WSA and OIS) applied under organic conditions also seemed an effective option, because they often resulted in the highest number of high performing genotypes for organic farming (Table 5.3, 5.4). These two selection procedures resulted in frequent selection of genotypes, such as H130, Klinta, PR 3605 and Rubiola (Table 5.5, 5.6), which were characterized by rapid early development, tall CH, high CGC, high GY and high grain quality. Genotypes such as PR 4814, and PR 5135, for which Rubiola was used as one of the parents, were also among the frequently selected genotypes when selecting for GY+WSA or OIS at

organic sites. Thus, the application of the procedures GY+WSA or OIS succeeded in selecting genotypes suitable for organic farming, i.e. genotypes that combine high GY and WSA. This confirms the conclusions of our earlier selection experiment in Kokare et al. (2017, Chapter 4), where also a combination of GY and WSA gave better selection results than selection for GY alone.

The selection by OIS score also included other traits desirable for organic farming such as earliness, disease resistance, and grain quality of barley, in combination with good GY and WSA. However, the high OIS of some selected genotypes may have been due to a relatively high WSA score compensating a medium GY score and vice versa as an example with PR-3605 at O2 and Sencis at O1 and O2 shows (Table 5.6). A possible trade-off between GY and WSA could occur within OIS, for which our previous selection experiment found some evidence (Kokare et al., 2017; Chapter 4). In order to avoid too much emphasis on WSA (42% of OIS) and a possible trade-off with GY (40% in OIS), the weight in OIS given to the individual traits should be carefully reconsidered. Possibly, more weight should be given to GY (e.g. 50%) and less weight to WSA (e.g. 30% for WSA).

The use of OIS as a selection criterion requires a much larger number of traits to be scored compared to other selection procedures and may be considered too laborious and time-consuming. Besides, some genotypes frequently selected by OIS were also frequently selected by the procedure GY+WSA. Therefore, selection based on GY+WSA or WSA+GY would be adequate and more efficient than selection based on OIS under organic conditions.

Indirect selection

Indirect selection rarely showed a significant increase for both GY and WSA simultaneously, particularly if the selection was performed at high input site C2. The selection for high GY only under high input conventional conditions leads to a significant decrease in WSA under organic conditions in various year orders. Therefore, the trade-off between GY and WSA cannot be ruled out, when the emphasis is on GY without taking traits related to WSA into account. Our finding that the correlation coefficient between traits contributing to WSA and GY was lower at conventional sites than at organic selection sites also indicates that it may be difficult to combine these two traits

under an indirect breeding approach. Although most of the genotypes selected in the conventional environment differed from those selected under organic conditions, the procedures in which the selection for both traits (GY and WSA) were combined (WSA+GY, GY+WSA, and OIS) resulted in GY and WSA which were not significantly lower than the average of all selected genotypes at both organic evaluation sites O1 and O2 (Supplemental Tables S7, S8). However, as shown by an analysis of genotypes concerning GY and WSA (Table 5.4), only approximately half of the 10 genotypes selected in conventional environments had GY and WSA above the mean of all 134 genotypes when tested under organic conditions. As pointed out above, it could be explained by large genotype \times environment ($G \times Y \times (M/S)$) interaction. Nevertheless, our results suggest that it could be possible to achieve the combination of both high GY and high WSA for organic conditions by performing selection under conventional conditions.

This can be illustrated by H130 and Klinta, which performed well under organic conditions and were among the most frequently selected genotypes under conventional conditions as well, especially by the procedures in which GY was combined with WSA. Besides, some genotypes such as Druvis and Rubiola, which mainly were selected under organic conditions, to some extent also appeared among the frequently selected genotypes under conventional. Rubiola also featured in an earlier experiment with 10 barley varieties at the same organic and conventional testing sites as in the present study, and was found among the highest yielding varieties over a wide range of environments (year-location combinations) (Kokare et al., 2014).

In our approach, we selected fixed numbers of genotypes (20 or 10) for GY and WSA, based on their ranking, not considering the size of the differences between genotypes for these traits. This probably left out some good genotypes in some years where they performed only slightly worse than the 20th or 10th selected genotype. We suggest that in further breeding for organic farming, a larger number of genotypes from the initial set of material should be selected; the genotypes with very similar productivity levels or weed suppressive ability should not be left out and should be taken for further evaluation. In addition, we recommend the testing of the selected genotypes at several organic sites to select the genotypes with a high GY and WSA for a diverse range of environments.

5.5 Conclusions

The results of this selection experiment suggest that the selection in organically managed fields was the most effective approach to identify highly suitable genotypes for organic farming.

In order to obtain genotypes suitable for a wide range of organic environments combining high GY and high WSA, the most effective selection was when mild selection for weed suppressive ability was followed by strict selection for grain yield (WSA+GY) under organic conditions. Because of the low correlation for yield between organic sites and between years, additional multi-year and multi-location testing is recommended. Our results also offer perspectives for breeders who aim to serve organic farming in their conventionally managed breeding programs, if the selection is performed not only for grain yield but also for traits contributing to weed suppressive ability (WSA+GY, GY+WSA, and OIS). In that case, an increase in both GY and WSA for genotypes selected could be achieved for the different organic environments, but is less effective than direct selection under organic conditions, because the indirect selection resulted in a smaller number of genotypes that performed high in GY and WSA than direct selection under organic conditions.

Selection based on organic ideotype scores (OIS), comprising various important traits for organic farming including GY and WSA, resulted in the highest number of well-performing genotypes for grain yield and weed suppressive ability in both organically and in conventionally managed selection fields. OIS could be applied as an alternative to both previously mentioned selection procedures in programs for barley breeding for organic farming. Depending on the demands of farmers and food producers, the combination of traits and their weight in the selection score of OIS may be adapted according to their specific requirements.

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Supplementary material

Table S1 Characterisation of soil and crop management practices in two organic (O1 and O2) and two conventional (C1 and C2) growing sites

Location	Year, sowing data	Soil type	Soil texture	pH KCl	P ₂ O ₅ (mg kg ⁻¹)	K ₂ O (mg kg ⁻¹)	Humus content (g kg ⁻¹)	Available N content in soil in spring (kg ha ⁻¹ according to the humus content)	Fertility management			Management of
									Precrop	Amount of N, P, K	Type	Diseases, Weeds pests
O1	2010 21.04	Sod	Loamy podzolic sand	5.7	111	144	28	75.6	Peas for green manure	N 26.5 kg ha ⁻¹	Green manure, approx. 20 t ha ⁻¹ (precrop)	No 1 × harrowing at tillering stage
	2011, 21.04	Sod	Loamy podzolic sand	5.4	116	98	21	56.7	Peas for green manure	N 26.5 kg ha ⁻¹	Green manure, approx. 20 t ha ⁻¹ (precrop)	No 1 × harrowing at tillering stage
	2012 28.04	Sod	Loamy podzolic sand	5.7	160	93	23	62.1	Peas for green manure	N 26.5 kg ha ⁻¹	Green manure, approx. 20 t ha ⁻¹ (precrop)	No 1 × harrowing at tillering stage
O2	2010, 19.04	Sod	Loamy podzolic sand	6.5	265	173	35	94.5	Perennial grasses	N 41 kg ha ⁻¹	Stable manure 40 t ha ⁻¹	No No
	2011 20.04	Sod	Loamy podzolic sand	6.6	394	167	30	81.0	Spring wheat	-	- -	No No
	2012 28.04.	Sod	Loamy podzolic sand	5.6	171	129	23	62.1	Perennial grasses	-	- -	No No

C1	2010 30.04	Sod podzolic	Loamy sand	5.5	100	132	26	70.2	Potatoes	N 81 kg ha ⁻¹ P 48 kg ha ⁻¹ K 84 kg ha ⁻¹	Inorganic	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
	2011 27.04	Sod podzolic	Loamy sand	5.4	187	165	30	81.0	Potatoes	N 83 kg ha ⁻¹ P 45 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
	2012 2.05	Sod podzolic	Sandy loam	5.7	264	155	27	72.9	Potatoes	N 80 kg ha ⁻¹ P 10 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic	Insecticide Decis 2.5 e.c. 0.15 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
	2010 3.05	Sod podzolic	Loamy sand	5.6	115	159	28	84.3	Potatoes	N 120 kg ha ⁻¹ P 48 kg ha ⁻¹ K 84 kg ha ⁻¹	Inorganic	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
C2	2011 29.04	Sod podzolic	Sandy loam	4.5	200	155	23	62.1	Potatoes	N 120 kg ha ⁻¹ P 45 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
	2012 11.05	Sod podzolic	Sandy loam	5.4	166	175	17	45.9	Potatoes	N 120 kg ha ⁻¹ P 30 kg ha ⁻¹ K 45 kg ha ⁻¹	Inorganic	Herbicide Secator 0.15 l ha ⁻¹ and Estets 600 e.k. 0.5 l ha ⁻¹	

Table S2 Information on country of origin and performance of the 134 barley genotypes included in the present investigation. Yield data (GY) and weed suppressive ability (WSA) are averaged over three years (2010-2012) at the two organic sites (O1 and O2) separately. For sites O1 and O2 we also present the average ranking numbers of the genotypes for yield over three years, and the selection procedure and site (organic O1 and O2 and conventional C1 and C2 sites) where the respective genotype was selected in three or more year orders

Genotype	Country of origin	Evaluation site O1			Evaluation site O2			Selection procedure ^a		
		Rank of GY	Rank of average GY	WSA	Rank of GY	Rank of average GY	WSA	GY + WSA	GY+ WSA	OIS
Druvis	Latvia	3.88	1 ^b	1.36	1.97	55	0.58	O1	O1	O1 C2
PR-3005	Latvia	3.84	2	0.11	2.87	3	0.52	O2	O2	O2
Abava	Latvia	3.81	3	1.32	1.76	88	0.35	O1	O1	C2
PR-4814	Latvia	3.80	4	1.2	1.95	57	0.32	O1	O1	C2 O1
Rubiola	Latvia	3.75	5	1.13	2.73	4	0.77	O1 O2	O1 O2 C2	O2
PR-4181	Latvia	3.75	6	-0.01	2.04	42	0.27	C2	C2	C2
PR-3518	Latvia	3.64	7	-1.06	1.86	72	-1.40			
PR-5105	Latvia	3.62	8	0.61	2.58	8	0.79	O1 O2	O1	
PR-3282	Latvia	3.53	9	1.17	1.76	87	0.95	O1	O1	O1 O1
Imula	Latvia	3.51	10	0.98	1.16	129	-0.40			O1
Kristaps	Latvia	3.46	11	0.07	1.42	115	-1.20	O1		
1079488-45	Latvia	3.46	12	0.91	1.89	68	1.40	O1		
PR-5137	Latvia	3.45	13	0.39	2.62	5	0.53			
PR-3297	Latvia	3.44	14	1.43	1.67	96	0.50		O1	
Otira	Denmark	3.43	15	-0.49	2.19	23	-0.90			
H130	Latvia	3.39	16	0.91	2.15	28	1.29	C1	C1	O1
Balga	Latvia	3.36	17	0.04	2.32	15	0.06			
797877-39	Latvia	3.33	18	0.06	1.9	66	0.46			
PR-4835	Latvia	3.32	19	0.36	2.19	25	1.04			
Stendes	Latvia	3.32	20	0.63	2.05	41	0.15			
PR-3605	Latvia	3.32	21	1.53	2.06	40	1.01		O1	O1
Sencis	Latvia	3.31	22	-0.29	2.58	9	0.21	O2		
Inari	Finland	3.31	23	0.05	1.89	69	-0.20			
PR-3223	Latvia	3.31	24	-0.49	2.59	6	0.43			
Malva	Latvia	3.3	25	0.68	2.09	36	0.27			
BZ12-63	Latvia	3.29	26	1.24	1.75	89	0.08		O1 C1	C1
Klinta	Latvia	3.28	27	1.31	2.46	12	0.67	O2 C1	O2 C2	O1 C1
Rasa	Latvia	3.26	28	0.70	1.66	98	-0.40			
827580-15	Latvia	3.26	29	0.89	1.86	73	-0.20		O1 C1	

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1012786-41	Latvia	3.25	30	0.16	2.29	17	0.48				
754277-27	Latvia	3.2	31	-0.15	1.62	103	-0.30				
PR-4121	Latvia	3.17	32	0.08	1.58	105	-0.00				
1271100-26	Latvia	3.15	33	-0.17	2.21	20	0.77				
PR-3522	Latvia	3.12	34	-0.46	1.95	58	0.23				
768678-28	Latvia	3.11	35	0.57	1.49	110	-0.30		C1	C1	
PR-5131	Latvia	3.11	36	0.37	2.13	31	0.39	C1			
BZ14-12	Latvia	3.1	37	0.58	2.53	10	1.01				
3280-14-1-4	Estonia	3.09	38	-0.74	2.02	45	-0.80				
Mik1	Russia	3.09	39	-0.57	1.85	74	-1.10				
Anni	Estonia	3.08	40	-1.16	1.8	80	-1.00				
12820	Latvia	3.05	41	0.10	1.82	79	0.94				
G-131	Latvia	3.04	42	0.31	2.2	22	0.00				
SZD4748	Austria	3.03	43	-0.56	2.13	30	-0.40				
PR-3512	Latvia	3.02	44	-0.86	1.98	49	-1.10				
Iakub	Belarus	3.02	45	0.46	1.98	52	-0.30				
PR-3885	Latvia	3.01	46	-1.09	2.07	38	-0.30	C2			
B-93	Latvia	3.01	47	0.57	1.89	70	0.32				
Ilga	Latvia	3.01	48	0.47	1.9	65	-0.10				
Golf	United Kingdom	3.01	49	1.15	1.91	64	0.60	C1	C1	C1	C1
1272500-36	Latvia	3.00	50	0.01	1.98	51	-0.50			C2	
PR-4822	Latvia	3.00	51	0.57	2.17	26	0.68				
Hellana	Austria	3.00	52	-0.24	1.96	56	-0.20				
L-2630	Latvia	2.99	53	0.05	1.41	117	0.14				
1163691-34	Latvia	2.99	54	0.04	1.85	75	0.55	C2	C2	C2	
PR-3300	Latvia	2.99	55	-1.17	1.98	53	-0.60				
Ula	Lithuania	2.98	56	0.55	2.07	39	1.11	C2	C2	O2	
Vienna	Austria	2.97	57	-0.55	2.21	21	-1.00				
718676-19	Latvia	2.97	58	1.08	1.44	114	-0.50				
BZ14-99	Latvia	2.95	59	0.59	2.04	43	0.99				
813380-13	Latvia	2.95	60	0.41	1.85	76	0.31				
Ruja	Latvia	2.95	61	0.37	1.92	62	0.70				
Ansis	Latvia	2.94	62	-1.36	1.45	113	-1.20				
L-2985 1	Latvia	2.94	63	0.18	2.07	37	0.88		O2		
Alsa	Lithuania	2.92	64	-0.36	1.92	61	0.13		C1		
L-2544	Latvia	2.91	65	-0.05	1.31	123	0.42				
Leeni	Estonia	2.9	66	-0.76	1.63	100	-1.60	C1			
Nuevo	Denmark	2.9	67	-1.11	1.33	122	-1.20				
L-3101	Latvia	2.9	68	0.10	2.31	16	0.41				
PR-4812	Latvia	2.89	69	1.10	2.34	14	0.81				
Aura	Lithuania	2.88	70	0.34	2.12	32	-0.10				
Idumeja	Latvia	2.88	71	0.60	2.01	47	0.41				
Linga	Latvia	2.86	72	0.71	2.17	27	0.83				

Tocada	Germany	2.86	73	-1.08	2.87	2	-0.60	O2 C1
HeilsHanna	Czech Republic	2.85	74	1.17	1.31	124	-0.10	
PR-4115	Latvia	2.85	75	-1.73	1.71	92	-1.70	
Peggy	Germany	2.83	76	-0.78	1.98	50	-0.90	
12825	Latvia	2.82	77	-0.07	2.59	7	1.09	O2 O2 O2
Vada	Netherlan ds	2.82	78	0.37	1.42	116	-0.50	
69-CIho11319	United Kingdom	2.81	79	1.29	2.11	33	0.97	
Pervonez	Ukraine	2.79	80	0.89	1.67	97	0.95	
BZ12-83	Latvia	2.79	81	0.89	1.77	86	0.83	
L-2735	Latvia	2.78	82	-0.07	1.91	63	-0.0	C1
Priekuļu	Latvia	2.77	83	0.62	1.37	120	-0.1	
PR-3636	Latvia	2.76	84	0.15	1.92	60	0.05	
PR-5135	Latvia	2.76	85	0.85	2.97	1	0.60	O2 O2 C2 C1 C2
Pallas	Sweden	2.75	86	0.17	1.62	102	0.02	
743-09	Latvia	2.75	87	-1.53	1.79	82	-1.2	C2
PR-3134	Latvia	2.71	88	0.18	1.77	85	0.33	
PR-5117	Latvia	2.71	89	0.45	1.93	59	0.82	C1 C1
Verena	Germany	2.7	90	-0.25	1.69	95	-0.2	
Dziugiai	Lithuania	2.7	91	1.42	2.02	44	1.58	O2
12819	Latvia	2.69	92	-0.38	1.54	106	-0.4	
M9	Latvia	2.68	93	-0.94	1.84	78	-1.7	
PR-4803	Latvia	2.66	94	0.35	1.45	112	0.84	
Eunova	Austria	2.64	95	0.21	2.00	48	0.03	C2
250-PI436150	Chile	2.63	96	-0.27	2.19	24	0.15	
PR-3520	Latvia	2.61	97	-0.41	1.8	81	-0	
PR-5127	Latvia	2.61	98	0.65	2.11	34	0.59	
Priekuļu 60	Latvia	2.6	99	0.18	1.66	99	-0.2	
BZ12-93	Latvia	2.58	100	-0.28	1.27	125	-0.3	
1263098-13	Latvia	2.58	101	0.01	1.85	77	0.19	
Primus	Sweden	2.54	102	1.11	1.63	101	0.7	
Gate	Latvia	2.52	103	-0.83	1.26	126	-0.3	
L-2295	Latvia	2.51	104	-1.03	2.5	11	-0.5	
Betzes	Germany	2.47	105	-0.19	2.27	18	0.45	
Bor88377	Finland	2.47	106	-0.32	1.72	91	-0.2	
Roxana	Germany	2.44	107	-0.22	1.9	67	0.05	
Divosnoje	Belarus	2.44	108	-0.69	1.5	109	-0.6	
PR-4810	Latvia	2.42	109	0.54	2.38	13	0.96	O2 O2
PR-5145	Latvia	2.4	110	0.38	2.24	19	0.75	
BZ14-90	Latvia	2.35	111	1.02	2.1	35	1.65	O2 O2
PR-3351	Latvia	2.32	112	-0.63	1.7	94	-0.8	
BZ12-86	Latvia	2.29	113	0.60	1.52	108	0.55	

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Kombainie ris	Latvia	2.28	114	0.47	1.78	84	0.79	
Camila	Germany	2.28	115	-1.52	2.02	46	-0.2	
1273300-50	Latvia	2.22	116	-0.94	1.38	119	-0.8	
Agra	Latvia	2.21	117	1.12	1.53	107	0.87	
1267199-30	Latvia	2.2	118	-0.41	1.97	54	0.04	C2
Maaren	Sweden	2.18	119	-1.53	1.7	93	-0.9	
Heris	Czech Republic	2.07	120	-1.03	1.75	90	-1.0	
PR-3245	Latvia	2	121	-1.38	2.15	29	-0.9	
Lysiba	Denmark	1.98	122	-1.67	0.9	134	-2.3	
PR-3515	Latvia	1.96	123	-1.45	1.87	71	-0.6	C2
PR-4144	Latvia	1.95	124	-0.01	1.46	111	0.11	
12811	Latvia	1.93	125	-0.81	0.95	132	-0.9	
Justina	Germany	1.91	126	-1.72	1.4	118	-2.0	
Dzintars	Latvia	1.87	127	0.38	1.59	104	0.73	C1
Vairogs	Latvia	1.82	128	0.06	1.13	130	1.27	
PR-4832	Latvia	1.81	129	-1.17	1.33	121	-0.8	
Latvijas vietejie	Latvia	1.66	130	-0.7	1.04	131	0.07	
Annabell	Germany	1.57	131	-2.09	0.94	133	-1.9	
Thuringia	Germany	1.47	132	-1.01	1.78	83	-1.0	
Steffi	Germany	1.35	133	-1.47	1.22	128	-1.3	
Danuta	Germany	1.25	134	-1.87	1.22	127	-1.8	

^aGY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score.

^b The 134 genotypes in the table are ordered from the highest to the lowest average yield over three years at organically managed site O1.

Table S3 Partitioning of variance components (%) for the traits observed of 134 barley genotypes grown in two organically and two conventionally managed growing sites, in the years 2010 – 2012

Trait	G ^a	M	Y	M/S	M×Y	M/S×Y	G×M	G×Y	G×(M/S)	G×M×Y	G×Y×(M/S)
GY ^b	4.6	44.7	0.0	11.9	0.2	22.6	0.2	0.3	0.7	0.4	14.4
Traits contributing weed suppressive ability (WSA)											
CH	31.6	23.0	17.9	0.0	0.0	4.3	1.7	2.7	0.9	1.9	16.0
CGC	5.3	51.7	4.1	0.5	9.6	3.3	1.8	2.2	0.0	0.7	20.8
LFL	16.4	2.2	15.1	0.0	0.0	19.2	0.5	6.3	1.0	1.2	38.2
WFL	51.1	3.9	4.2	0.0	0.0	10.0	1.4	1.8	0.5	1.6	25.7
PH	29.7	20.8	11.8	0.0	0.0	13.8	0.0	1.5	0.0	1.4	21.1
Traits related to growing period (GP)											
HED	12.1	70.2	1.9	5.3	0.0	5.0	0.3	0.6	0.2	0.0	4.5
MAT	5.8	43.7	37.5	0.0	3.2	2.4	0.3	1.1	0.0	0.0	5.9
Trait contributing diseases resistance (DR)											
NET	0.6	0.0	8.3	0.0	1.1	12.7	0.0	14.6	0.1	0.0	62.6
PWD	23.5	0.0	1.0	0.0	0.0	41.0	0.0	2.1	0.0	0.0	32.4
Trait related to grain quality (Q)											
VOL	26.3	0.0	21.7	0.0	10.9	11.3	2.4	3.5	0.0	0.0	23.9

^a Genotype main effect (G), management system (organic vs. conventional) (M), year main effect (Y), growing site nested within management system (M/S), Management system by year interaction (M×Y), growing site nested within management system by year interaction (M/S×Y), genotype by management (G×M) interaction, genotype by year interaction (G×Y), genotype by site (within management) interaction (G×M/S), genotype by management system and by year interaction G×M×Y and genotype by year, by growing site within management interaction (G×Y×(M/S)) and all remaining variation as residual variance.

^b GY = grain yield; CH = canopy height; CGC = crop ground cover; LFL = length of flag leaf; WFL = width of the flag leaf; PH = plant height before harvest; HED = length of the growth period from sowing to heading; MAT = length of the growth period from sowing to maturity; NET = resistance to net blotch; PWD = resistance to powdery mildew; VOL = volume weight

Table S4a Pearson correlations (r) for the traits between sites and between management systems (across all 134 barley genotypes and traits averaged over the three years)

Traits	Correlation (r) between sites, and management systems ^a						
	O1/O2	O1/C1	O1/C2	O2/C1	O2/C2	O2/C	O/C
GY ^b	0.20 ^{*c}	0.31 ^{**}	0.31 ^{**}	0.21 [*]	0.16	0.25 ^{**}	0.25 ^{**}
CH	0.58 ^{**}	0.69 ^{***}	0.65 ^{***}	0.62 ^{***}	0.60 ^{***}	0.63 ^{***}	0.63 ^{***}
CGC	0.36 ^{**}	0.38 ^{**}	0.30 ^{**}	0.29 ^{**}	0.20 [*]	0.29 ^{**}	0.29 ^{**}
LFL	0.34 ^{**}	0.32 ^{**}	0.30 ^{**}	0.39 ^{***}	0.43 ^{***}	0.35 ^{***}	0.35 ^{***}
WFL	0.66 ^{***}	0.63 ^{***}	0.60 ^{***}	0.67 ^{***}	0.77 ^{***}	0.66 ^{***}	0.66 ^{***}
PH	0.55 ^{***}	0.61 ^{***}	0.55 ^{***}	0.58 ^{***}	0.56 ^{***}	0.58 ^{***}	0.58 ^{***}
HED	0.72 ^{***}	0.79 ^{***}	0.73 ^{***}	0.71 ^{***}	0.66 ^{***}	0.73 ^{***}	0.73 ^{***}
MAT	0.61 ^{***}	0.57 ^{***}	0.60 ^{***}	0.49 ^{***}	0.52 ^{***}	0.54 ^{***}	0.54 ^{***}
VOL	0.67 ^{***}	0.46 ^{***}	0.57 ^{***}	0.40 ^{***}	0.67 ^{***}	0.60 ^{***}	0.60 ^{***}

^a Pearson correlation coefficient (r) across 134 barley genotypes for averages per trait over years between sites and between conventional (C) and organic (O) management systems

^b The values expressed: GY = grain yield, t ha⁻¹; CH = canopy height, cm; CGC = crop ground cover, %; LFL = length of flag leaf, cm; WFL = width of the flag leaf, mm; PH = plant height before harvest, cm; HED= days from sowing to heading; MAT = days from sowing to maturity; VOL = volume weight g l⁻¹

^c * Significant at $P = 0.05$, ** significant at $P = 0.01$, ***significant at $P= 0.001$

Table S4b Genotypic rank correlation coefficient (Rs)^a for the grain yield (GY) between 12 environments (two organic O1 and O2 and two conventions C1 and C2 sites in three years 2010, 2011, 2012)

	O1_2010	O2_2010	C1_2010	C2_2010	O1_2011	O2_2011	C1_2011	C2_2011	O1_2012	O2_2012	C1_2012	C2_2012
O1_2010	1.000											
O2_2010	0.026	1.000										
C1_2010	-0.011	-0.024	1.000									
C2_2010	0.278**b	0.085	0.060	1.000								
O1_2011	0.264**	0.225**	0.154	0.280**	1.000							
O2_2011	0.035	-0.040	0.132	0.160	0.124	1.000						
C1_2011	0.240	0.153	0.189*	0.522***	0.386***	0.197*	1.000					
C2_2011	0.146	0.140	-0.040	0.360***	0.360***	0.163	0.363***	1.000				
O1_2012	0.225**	0.171	0.151	0.171*	0.360***	0.229**	0.231**	0.076	1.000			
O2_2012	0.178*	0.087	0.255**	0.181*	0.547***	0.258**	0.255**	0.165	0.378***	1.000		
C1_2012	0.227**	0.197	0.112	0.333***	0.397***	0.235**	0.423***	0.235**	0.393***	0.428***	1.000	
C2_2012	0.237**	0.319***	0.026	0.302***	0.376***	0.185*	0.355***	0.389***	0.251**	0.224**	0.454**	1.000

^a Spearman rank correlation coefficient

^b *Significant at $P = 0.05$, ** significant at $P = 0.01$, ***significant at $P = 0.001$

Table S5 The relative grain yield (GY) of ten selected barley genotypes, obtained in direct (at two organic sites O1 and O2) and indirect (at two conventional C1 and C2 sites) selection according to four selection procedures (GY; WSA+GY; GY+WSA; OIS)^a evaluated in the two organic sites (O1 and O2) in six year orders

Year order	Trait	Selection procedure	Evaluation in O1				Evaluation in O2			
			Direct		Indirect		Direct		Indirect	
			O1	O2	C1	C2	O1	O2	C1	C2
10/11/12 ^d	GY	GY	21 ^{*b,c}	13	19 [*]	-6	6	14	8	7
		WSA+GY	12	25 [*]	16	-3	23 [*]	22 [*]	28 [*]	2
		GY+WSA	12	12	17	0	15	4	22 [*]	12
		OIS	10	26 [*]	9	-9	28 ^{**}	2	26 ^{**}	6
10/12/11	GY	GY	13 [*]	15 [*]	6	5	5	3	-5	4
		WSA+GY	12 [*]	17 [*]	7	8	4	21	18	-7
		GY+WSA	15 [*]	11	11	0	-2	-2	25	39 [*]
		OIS	12 [*]	14 [*]	8	3	1	-6	-11	39 [*]
11/10/12	GY	GY	7	19 [*]	1	-12	21 [*]	34 ^{**}	-4	-6
		WSA+GY	7	13	26 [*]	3	24 [*]	37 [*]	9	0
		GY+WSA	13	10	10	-1	21 [*]	25 [*]	11	17 [*]
		OIS	29 [*]	17 [*]	7	4	15	26 [*]	-1	10
11/12/10	GY	GY	9	-6	-1	13 [*]	14 [*]	-4	10	13 [*]
		WSA+GY	0	-1	0	23 ^{**}	15 [*]	-6	8	12
		GY+WSA	18 [*]	3	4	4	4	-4	-2	10
		OIS	11 [*]	1	-1	13 [*]	5	-4	8	6
12/10/11	GY	GY	8	19 [*]	11 [*]	4	1	40 ^{**}	28	-11
		WSA+GY	10 [*]	17 [*]	11 [*]	2	-5	16	14	28
		GY+WSA	10	6	5	6	6	-8	-2	7
		OIS	15 [*]	14 [*]	8	7	-2	18	19	28
12/11/10	GY	GY	0	1	5	8	2	-2	4	15 [*]
		WSA+GY	6	-3	8	16 [*]	5	-8	7	8
		GY+WSA	20 ^{**}	-5	-4	1	3	-11	2	7
		OIS	13 [*]	-1	10	8	-2	-4	4	14 [*]

^a GY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score

^b GY of 10 selected genotypes expressed as a relative difference of the mean GY of the 10 selected genotypes and the mean GY over all 134 genotypes (100%);

^c* Significant at the 0.05 probability level; ** significant at the 0.01 probability level, marked in grey

^d Year order 10/11/12 means: selection in 2010 and 2011 followed by the final evaluation of the selection results using the 2012 data

Table S6 The average increase in weed suppressive ability (WSA) of ten selected barley genotypes, obtained in direct (at two organic sites O1 and O2) and indirect (at two conventional C1 and C2 sites) selection according to four selection procedures (GY; WSA+GY; GY+WSA; OIS)^a evaluated in the two organic sites (O1 and O2) in six year orders

Year order	Trait	Selection procedure	Evaluation in O1				Evaluation in O2			
			Direct		Indirect		Direct		Indirect	
			O1	O2	C1	C2	O1	O2	C1	C2
10/11/12 ^d	WSA	GY	0.30 ^b	0.27	0.29	-0.16	0.25	0.48	0.13	-0.03
		WSA+GY	0.52	0.53* ^c	0.63*	0.30	0.53*	0.72**	0.60*	0.12
		GY+WSA	0.82*	0.86*	0.53*	0.45	0.76*	0.85*	0.48	0.75*
		OIS	0.66*	0.77*	0.51	0.51	0.73*	0.64*	0.61*	0.58*
		WSA+GY	0.78*	0.72*	0.49	-0.33	0.27	0.18	0.12	0.14
10/12/11	WSA	GY	0.86*	0.58*	0.47	0.33	0.29	0.55*	0.23	0.46
		GY+WSA	1.00**	0.87*	0.71*	0.80*	0.57*	0.63**	0.57*	0.35
		OIS	0.71*	1.14**	1.03**	0.31	0.22	0.35	0.45	0.18
		WSA+GY	0.45	0.36	-0.40	-0.58*	0.50	0.33	-0.46	-0.12
		GY+WSA	0.65*	0.35	0.37	-0.07	0.70*	0.38	0.29	0.16
11/10/12	WSA	OIS	1.01**	0.68*	0.64*	0.73*	0.88**	0.77*	0.52*	0.88**
		OIS	1.06**	0.62*	0.32	0.42	0.85*	0.57*	0.29	0.52*
		GY	0.44	0.12	0.24	-0.04	0.82*	-0.03	0.10	-0.30
		WSA+GY	0.26	-0.14	0.33	0.30	0.89*	-0.07	0.38	0.41
		GY+WSA	0.75*	0.29	0.75*	0.80*	0.80*	0.05	0.19	0.66
11/12/10	WSA	OIS	0.50*	0.25	0.20	0.46	0.52*	0.24	0.42	0.34
		GY	0.39	0.44	0.43	-0.59*	-0.14	0.08	0.21	-0.11
		WSA+GY	0.61*	0.63*	0.69*	0.05	0.04	0.05	0.42	0.60*
		GY+WSA	0.73*	0.91*	0.76*	1.29*	0.65*	0.52	0.61*	0.16
		OIS	0.92*	0.69*	0.61*	0.35	0.34	0.15	0.54*	0.54*
12/10/11	WSA	GY	0.36	-0.03	0.17	-0.17	0.21	-0.03	-0.26	-0.25
		WSA+GY	0.49*	0.15	0.20	0.46	0.63*	0.11	0.05	0.43
		GY+WSA	0.65*	0.41*	0.43*	0.50*	0.64*	0.61*	0.56*	0.48*
		OIS	0.66*	0.09	0.34	0.77*	0.50	0.21	0.31	0.70*
		WSA	0.36	-0.03	0.17	-0.17	0.21	-0.03	-0.26	-0.25
12/11/10	WSA	WSA+GY	0.49*	0.15	0.20	0.46	0.63*	0.11	0.05	0.43
		GY+WSA	0.65*	0.41*	0.43*	0.50*	0.64*	0.61*	0.56*	0.48*
		OIS	0.66*	0.09	0.34	0.77*	0.50	0.21	0.31	0.70*
		WSA	0.36	-0.03	0.17	-0.17	0.21	-0.03	-0.26	-0.25
		WSA+GY	0.49*	0.15	0.20	0.46	0.63*	0.11	0.05	0.43

^a GY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score

^b Increase in WSA expressed as the difference between the average value of WSA of 10 selected genotypes and an average value of the total set of 134 genotypes (which is equal to 0)

^c * Significant at the 0.05 probability level; ** significant at the 0.01 probability level, marked in grey

^d Year order 10/11/12 means: selection in 2010 and 2011 followed by the final evaluation of the selection results using the 2012 data

Table S7 The significances of the differences between mean GY of barley genotypes (n=10) selected by direct selection at the organic site (O1 or O2) and mean GY of barley genotypes (n=10) obtained by indirect selection at conventional sites C1 and C2, when tested at each organic evaluation sites O1 and O2 according to four selection procedures: GY; WSA+GY; GY+WSA and OIS over the six-year orders; based on p-values of the independent-samples t-test

Year order	Trait	Selection procedure	Differences from O1			Differences from O2		
			O2	C1	C2	O1	C1	C2
10/11/12 ^d	GY	GY ^a	NS ^b	NS	(-) ^{c*}	NS	NS	NS
		WSA+GY	NS	NS	NS	NS	NS	NS
		GY+WSA	NS	NS	NS	NS	NS	NS
		OIS	NS	NS	NS	(+) [*]	NS	NS
10/12/11	GY	GY	NS	NS	NS	NS	NS	NS
		WSA+GY	NS	NS	NS	NS	NS	NS
		GY+WSA	NS	NS	(-) [*]	NS	NS	NS
		OIS	NS	NS	NS	NS	NS	NS
11/10/12	GY	GY	NS	NS	NS	NS	(-) [*]	(-) [*]
		WSA+GY	NS	NS	NS	NS	(-) [*]	(-) [*]
		GY+WSA	NS	NS	NS	NS	NS	NS
		OIS	NS	NS	(-) [*]	NS	(-) [*]	NS
11/12/10	GY	GY	NS	NS	NS	(+) [*]	(+) [*]	(+) [*]
		WSA+GY	NS	NS	(+) [*]	(+) ^{**}	(+) [*]	(+) [*]
		GY+WSA	(-) [*]	NS	NS	NS	NS	(+) [*]
		OIS	NS	NS	NS	NS	NS	NS
12/10/11	GY	GY	NS	NS	NS	NS	NS	NS
		WSA+GY	NS	NS	NS	NS	NS	NS
		GY+WSA	NS	NS	NS	NS	NS	NS
		OIS	NS	NS	NS	NS	NS	NS
12/11/10	GY	GY	NS	NS	NS	NS	NS	(+) [*]
		WSA+GY	NS	NS	NS	NS	NS	(+) [*]
		GY+WSA	(-) [*]	(-) [*]	NS	NS	NS	NS
		OIS	(-) [*]	NS	NS	NS	NS	(+) [*]

^a selection procedures: GY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score

^b * Significant at the 0.05 probability level; ** significant at the 0.01 probability level, NS no significant P < 0.05

^c significantly lower (-) or (+) higher value in comparison to direct selection carried out at respective organic site O1 or O2

^d Year order 10/11/12 means: selection in 2010 and 2011 followed by the final evaluation of the selection results using the 2012 data

Table S8 The significances of the differences between mean WSA of barley genotypes (n=10) selected by direct selection at the organic site (O1 or O2) and mean WSA of barley genotypes (n=10) obtained by indirect selection at conventional sites C1 and C2, when tested at each organic evaluation sites O1 and O2 according to four selection procedures: GY; WSA+GY; GY+WSA and OIS over the six-year orders; based on p-values of the independent-samples T-test

Year order	Trait	Selection procedure	Evaluation in O1			Evaluation in O2		
			O2	C1	C2	O1	C1	C2
10/11/12 ^d	WSA	GY ^a	NS ^b	NS	NS	NS	NS	NS
		WSA+GY	NS	NS	NS	NS	NS	(-) ^{c*}
		GY+WSA	NS	NS	NS	NS	NS	NS
		OIS	NS	NS	NS	NS	NS	NS
10/12/11	WSA	GY	NS	NS	(-) ^{c**}	NS	NS	NS
		WSA+GY	NS	NS	NS	NS	NS	NS
		GY+WSA	NS	NS	NS	NS	NS	NS
		OIS	NS	NS	NS	NS	NS	NS
11/10/12	WSA	GY	NS	(-) ^{c*}	(-) ^{c*}	NS	(-) ^{c*}	NS
		WSA+GY	NS	NS	(-) ^{c*}	NS	NS	NS
		GY+WSA	NS	NS	NS	NS	NS	NS
		OIS	NS	(-) ^{c*}	NS	NS	NS	NS
11/12/10	WSA	GY	NS	NS	NS	(+) ^{c*}	NS	NS
		WSA+GY	NS	NS	NS	(+) ^{c**}	NS	NS
		GY+WSA	NS	NS	NS	(+) ^{c**}	NS	(+) ^{c*}
		OIS	NS	NS	NS	NS	NS	NS
12/10/11	WSA	GY	NS	NS	(-) ^{c**}	NS	NS	NS
		WSA+GY	NS	NS	(-) ^{c*}	NS	NS	NS
		GY+WSA	NS	NS	(+) ^{c*}	NS	NS	NS
		OIS	NS	NS	(-) ^{c*}	NS	NS	NS
12/11/10	WSA	GY	NS	NS	NS	NS	NS	NS
		WSA+GY	NS	NS	NS	NS	NS	NS
		GY+WSA	NS	NS	NS	NS	NS	NS
		OIS	(-) ^{c*}	NS	NS	NS	NS	NS

^a selection procedures: GY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score

^b * Significant at the 0.05 probability level; ** significant at the 0.01 probability level, NS no significant P < 0.05

^c significantly lower (-) or (+) higher value in comparison to direct selection carried out at respective organic site O1 or O2

^d Year order 10/11/12 means: selection in 2010 and 2011 followed by the final evaluation of the selection results using the 2012 data

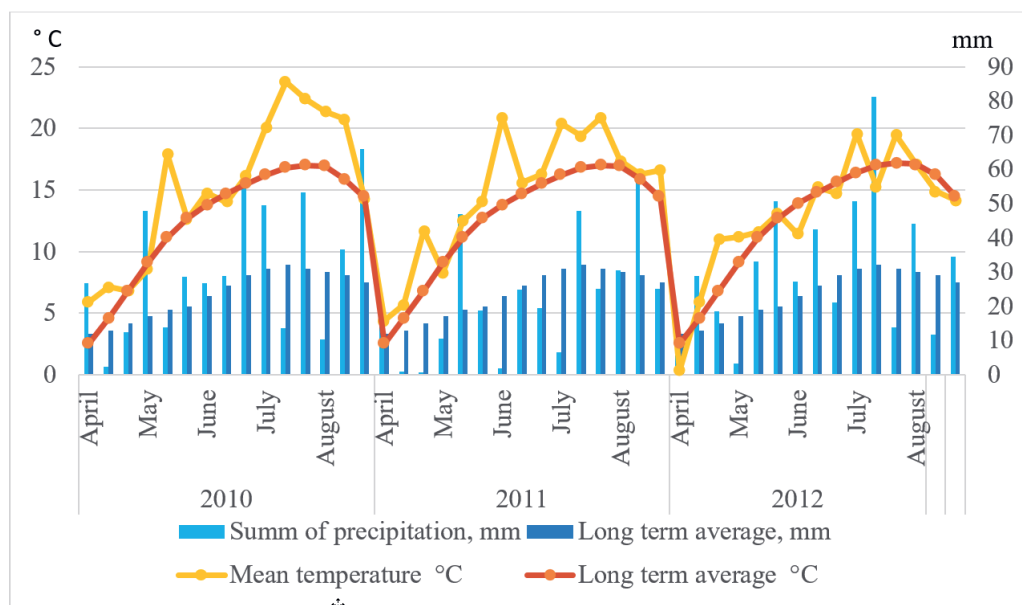


Figure S1 Mean temperatures and amount of precipitation at Priekuli, for the period from April to August during the years 2010 – 2012

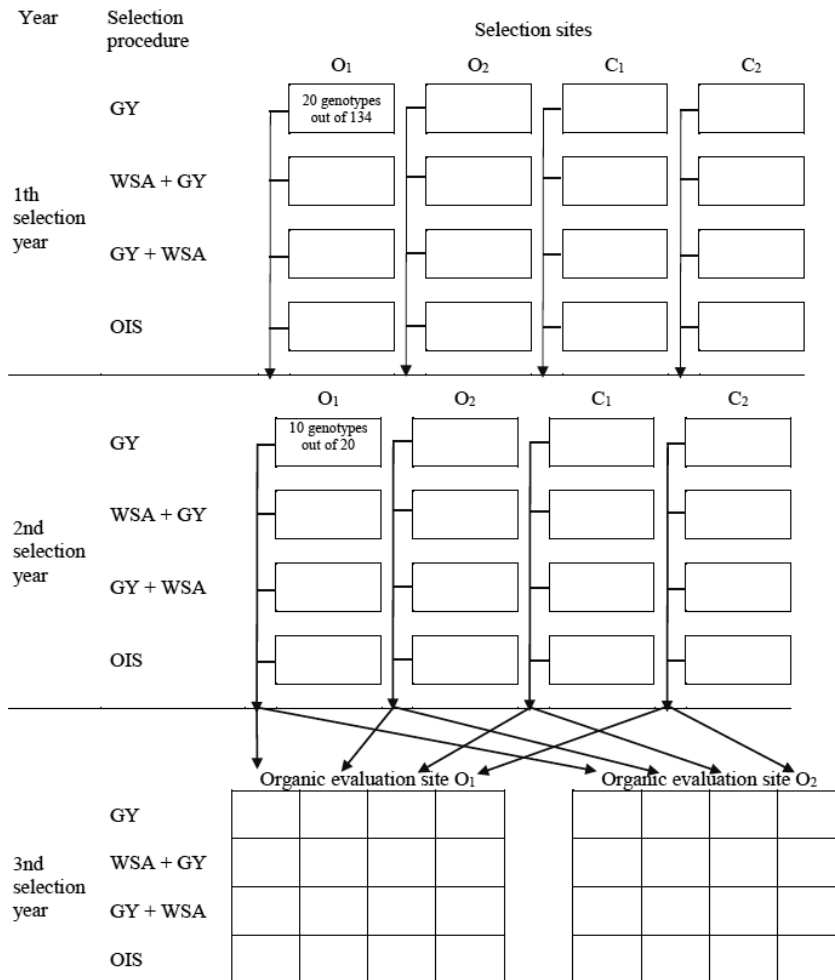
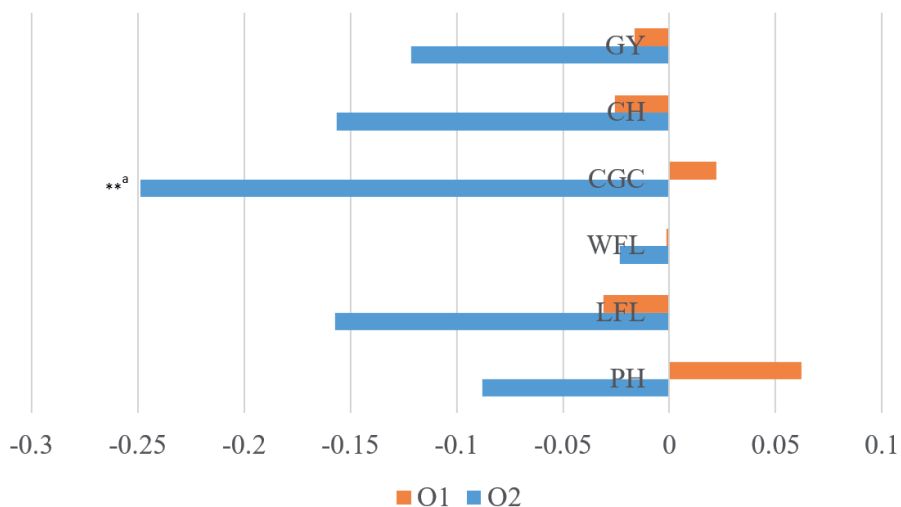
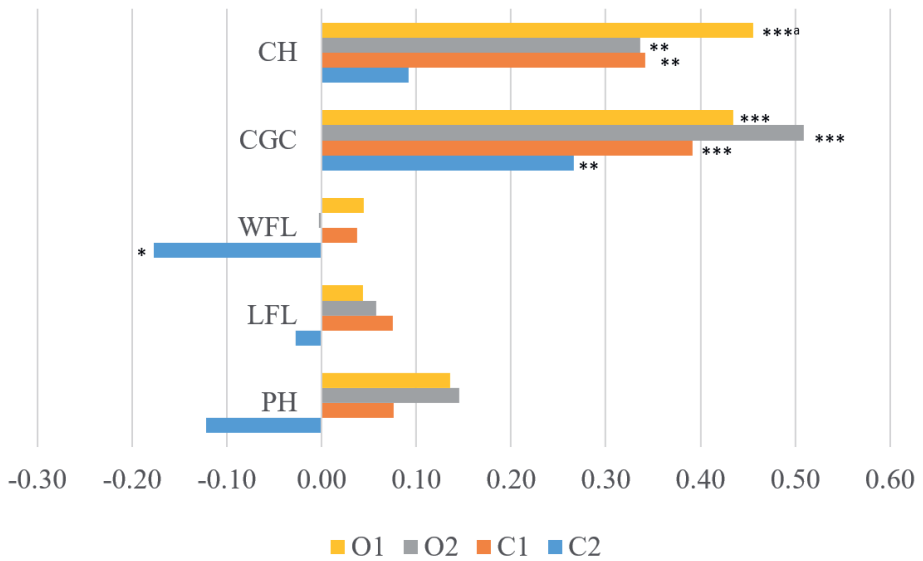


Figure S2 Scheme of selection procedures carried out at the organic sites (O₁ and O₂) and at the conventional sites (C₁ and C₂). Selection procedures: GY=selection for GY alone; GY+WSA= first mild selection for GY followed by strict selection for WSA; WSA+GY = first mild selection for WSA followed by strict selection for GY; OIS = organic ideotype score.



^a the asterisks show the significance at ** $p < 0.01$

Figure S3. Correlation between the weed ground cover (WEED) and the traits associated with competitiveness against weeds: canopy height (CH), crop ground cover (CGC), length of flag leaf (LFL), width of flag leaf (WFL), plant height (PH) and the grain yield (GY) in two organic (O1 and O2) sites over three years



*the asterisks show the significance at * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

Figure S4 Correlation between the barley yield and the traits associated with competitiveness against weeds: canopy height (CH), crop ground cover (CGC), length of flag leaf (LFL), width of flag leaf (WFL), plant height (PH) in two organic (O1 and O2) and two conventional (C1 and C2) sites over three year

Chapter 6

Genome wide association mapping for grain yield and weed suppressive ability in barley (*Hordeum vulgare* L.) under conventional and organic conditions

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Abstract

Organic farming has become increasingly important for the sustainable use of resources. Varieties combining high yield and weed suppressive ability are desirable for organic farming. The expression of the genetic factors contributing to the traits related to yield and weed suppressive ability could be different under organic/conventional conditions, and also their importance could differ under the different conditions. In this study, we used a genome-wide association study (GWAS) to identify quantitative trait loci (QTL) for grain yield (GY) and nine traits contributing to weed suppressive ability (WSA) in organic and conventional farming systems and determine their differences. An association mapping population panel consisting of 153 barley varieties and breeding lines relevant for Latvian farming was phenotyped over three growing seasons in two conventionally and two organically managed sites and genotyped with 1536 SNPs. Overall, 35 QTLs were identified for four traits contributing to weed suppressive ability, and 80% of these QTLs were management-specific. The QTLs for canopy height (CH), leaf inclination angle (LAN), width of flag leaf (WFL) and plant height at harvest (PH), were mapped in the organic system on chromosomes 3H, 7H, 6H, 2H, respectively. One QTL was found associated with several traits (CH, LAN and PH) in both systems on chromosome 3H. Those identified markers may be combined by breeders to develop barley cultivars with improved weed suppressive ability in organic farming.

Keywords:

GWAS, organic farming, QTL, spring barley, traits contributing to weed suppressive ability

6.1 Introduction

In organic farming crops have to cope with biotic and abiotic stresses and large variation in environmental conditions and limited nutrient availability. In breeding for organic farming, it is a challenge to develop varieties that maintain good and stable yields under low input conditions (Wolfe et al., 2008; Lammerts van Bueren et al., 2011; Lammerts van Bueren et al., 2012). Variety performance often differs between organic and conventional farming systems (Kokare & Legzdina, 2010; Kokare et al., 2014; Murphy et al., 2007; Sturite et al., 2019), Combining information from variety testing studies in both organic and conventional systems may help to identify traits that are particularly valuable for organic farming, and that could also be relevant under conventional management (Przystalski et al., 2008).

Beside the traits that are important for both conventional and organic farming systems, e.g. yield, grain quality and disease resistance, additional traits relevant specifically to organic farming, such weed suppressive ability, need to be considered.

Weeds competing with the crop is one of the most urgent problems in organic farming where herbicides are not applied (Hoad et al., 2008; Lammerts van Bueren et al., 2011). Therefore, high weed suppressive ability in varieties is particularly relevant (Lemerle et al., 2006; Murphy et al., 2008); moreover, weed suppressive ability of crops can also be regarded as a low-cost option for reducing the dependence on herbicides in conventional farming, at least if yield is not negatively affected. Weed suppressive ability is based on the plant's morphological characteristics (Hoad et al., 2008). Many traits can help to suppress weeds, including early vigour, increased plant height, higher canopy density, and good tillering capacity (Lammerts van Bueren et al., 2011; Mikó et al., 2014; Worthington et al., 2015; Mahajan et al., 2020; Kissing Kuseck et al., 2021a; 2021b)

Although the use of molecular markers is not self-evident for the organic sector and often debated, organic standards do not prohibit the use of molecular markers as diagnostic tool; therefore, marker-assisted selection (MAS) can be used in addition to phenotypic selection, if markers would become available for certain traits that are time consuming, complicated and expensive to phenotype directly (Lammerts van Bueren et al., 2010).

Formerly, biparental QTL linkage mapping was widely used to determine the genetic location of QTLs associated with relevant traits in barley (Alqudah et al., 2020). However, markers indicating QTLs for a particular trait in one mapping population can often not be applied in a different set of accessions, since those accessions may vary for other genes for that trait than those detected in the biparental mapping population. Genome-wide association studies (GWAS) have become a powerful alternative approach (Waugh et al., 2009). In GWAS a genetically diverse set of germplasm is analyzed for associations of markers with phenotypic traits so that more QTLs from different origins can be detected (Lorenz et al., 2010). GWAS requires a high-throughput molecular marker system to detect genetic variation for many markers in a large number of accessions which are simultaneously phenotyped for the traits of interest. The number of markers required for GWAS depends on the level of level of linkage disequilibrium (LD) - the non-random association of two or more alleles which is mostly due to linkage between loci. In cultivated barley LD extends up to 1–10 cM depending on the set of germplasm (Caldwell et al., 2006; Kraakman et al., 2004; Rostoks et al., 2006). Currently, available high-throughput genotyping platforms exceeding 1000 single nucleotide polymorphism (SNP) markers are sufficient to cover the barley genome. SNP markers are abundantly available in the genome and can be run in high-throughput assays (Bayer et al., 2017). Several resequencing efforts, e.g. by Rostoks et al. (2006) and eSNPs from expressed sequence tag data from different barley varieties (Close et al., 2009) identified a large number of barley SNPs allowing to establish a high-throughput SNP array for barley using Illumina Golden Gate technology (Rostoks et al., 2006) and to develop a high-density SNP map of barley (Close et al., 2009). Recently an iSelect platform and corresponding linkage map have been developed (Munoz-Amatriain et al., 2014).

A complicating issue in GWAS is that population structure can lead to spurious associations. To correct for these, it has been proposed to use a linear mixed model (Yu et al., 2006) in which information on population structure (Q-matrix) and/or differences in genetic relatedness (kinship or K-matrix) are included (Zhao et al., 2007). More recently, several approaches have been developed with the aim of increasing QTL power detection of the mixed model GWAS approach (e.g. Wang and Zhang, 2021).

Another issue is the correction of significance for multiple-testing in GWAS, which can be addressed with a Bonferroni correction (Balding, 2006), combined with estimating the effective number of independent SNPs by accounting for LD (Pe'er et al., 2008). Alternatively, a significance threshold could be adjusted by controlling the false discovery rate (FDR) (Benjamini et al., 2001) or by employing a permutation test (Dudbridge et al., 2008) or a Bayesian approach (Sebastiani et al., 2009).

In a GWAS on winter wheat, Tsai et al. (2020) revealed that some markers associated with grain yield were location-specific and usually significantly associated with one location only. The authors also indicate that it would be important in an analysis to include genotype-location information to identify QTLs having an effect at specific locations. There are not many studies that analyse quantitative traits of barley under organic and conventional conditions using GWAS. In a study on wheat, Zou et al. (2017) pointed out that not all identified QTLs were in common between conventional and organic management systems. Therefore, it is of interest to further study whether QTLs detected in conventional management systems can be used directly in the selection of the genotypes for organic farming. To this end, a spring barley association mapping population was used for this GWAS study. The aim of our study was to identify, in a GWAS of barley, QTLs for traits favourable for organic farming, such as yield and weed suppressive ability under both organic and conventional conditions, and to assess whether there are differences in identified QTLs between these different farming systems.

6.2 Materials and Methods

Plant material and field trials

In this study, 153 spring barley (*Hordeum vulgare* L.) accessions were included in an association mapping panel and evaluated in a field experiment in Priekuli, Latvia, during 2010- 2012 (Additional file 1). The accessions were obtained from various gene banks and from breeding material. All historical Latvian varieties, one Latvian landrace and older breeding lines stored at Latvian Gene Bank were included. Most of the accessions are hulled two-row barleys, but the set included 19 hulless barleys and 4 six-row barleys. The countries of origin of these barley accessions were Baltic or Nordic regions such as Latvia, Germany, Finland and Sweden.

Field trials were carried out in two organically (O1 medium input; O2 low-input farmer's field) and two conventionally managed (C1 medium input; C2 high input) environments/fields during three seasons: 2010-2012 (see for details Chapter 5, Additional file 2).

In these trials, the following traits were measured: grain yield (GY) and traits contributing to weed suppressive ability: plant growth habit (GWH), canopy height (CH); crop ground cover (CGC); length (LFL) and width (WFL) of flag leaf; leaf inclination angle (LAN); plant height before harvest (PH); number of productive tillers (NT) (Additional file 3).

SNP genotyping and data curation

DNA for genotyping was extracted from leaves of single plants using "DNeasy Plant Min Kit" ("Qiagen", Germany). Illumina high- throughput genotyping was done as described by Rostoks et al. (2006). The barley oligo pooled assay, BOPA1, contains 1536 SNPs selected from pilot assays (Close et al., 2009). Quality control of genotyping data involved removal of heterozygous SNP calls and markers with more than 10% missing data points as well as removal of markers with minor allele frequency less than 5%. The barley consensus linkage map based on these SNP markers (Close et al., 2009) was used throughout the study. 1055 markers were left with known map positions, then 74 duplicated SNPs were excluded. Finally, 981 markers remained in the GWAS analysis. 97% of the markers are less than 5 cM apart from each other, and there are 27 marker intervals of 5-15 cM on the map.

Statistical analysis of phenotyping data

Pearson correlation coefficients among the averages per trait over three years within each farming system were calculated for the data using GENSTAT 18.0

Association analysis

To calculate genotype adjusted means for yield and the traits evaluated as input for GWAS, ANOVA was performed using R (version 4.0.3) with this model:

$$\mu + G + Y + L + G \times Y + G \times L + L \times Y + e, \text{ where} \quad [5]$$

the genotype (G), year (Y), and locations (L) (two locations (O1 and O2) nested within the organic farming system and two locations (C1 and C2) nested within the conventional farming system) were fixed factors in the model, and the residuals (e) were the random term. After that, per trait, the best linear unbiased estimates (BLUEs) of genotypes were calculated for each of the four locations and later used for association analyses. The BLUEs of leaf inclination angle (LAN) were calculated without the year 2010, since LAN was scored differently in 2010.

A Principal Components Analysis (PCA) on the marker genotypes was performed by GAPIT version 3 (GAPIT3) or by the *prcomp* R function (R Core Team, 2021) to analyze population structure.

GAPIT3 was used to perform genome-wide association analysis in R using the multi-locus Bayesian-information and Linkage-disequilibrium Iteratively Nested Keyway (BLINK). The first 3 principal components were included as covariates in the BLINK model, to correct for population structure.

Trait heritability (h^2) was estimated by GAPIT3 for each location based on the variance components of the mixed linear model (MLM) (Yu et al., 2006), as the ratio of additive genetic variance over the total variance.

GWAS analysis for the trait hullessness was first conducted to check the reliability of the methods and software.

GWAS analyses for all 12 traits in four locations (C1, C2, O1, O2) were conducted. Two significance thresholds for markers were used ($\alpha = 0.05$), based on a Bonferroni correction (BC) (Armstrong, 2014) and on a less stringent false discovery rate (FDR) method (according to Benjamini and Hockberg, 1995).

Phenotypic variation explained by SNPs was estimated in two ways: i) as the difference between the R square of the full regression model (with all significant SNPs) and the R square of the same model without a SNP; or ii) using the R square of a regression model including one significant SNP only, to obtain the phenotypic variation explained by that SNP. Besides, the beta

coefficient of the model with only one significant SNP was used to estimate the effect of a SNP.

Linkage disequilibrium (LD) decay was estimated per chromosome based on the 90th percentiles of marker correlations (r^2) in distance bins of 2.5 cM. Significant SNPs were grouped (as being linked to the same QTL) when their distance was less than the LD decay distance, the distance at which LD decayed below 0.2.

6.3 Results

Difference in phenotypic traits between the organic and conventional locations

The grain yield was higher in the locations within the conventional farming systems than within organic system. The yield level within the organic system fluctuated considerably (Table 6.1). The main effect of the location (L), which includes two organic locations and two conventional locations, was significant for all traits contributing to weed suppressive ability (Additional file 4). The largest differences between organic and conventional locations were observed for CH, CGC, PH, and NT. Within the farming systems, fewer differences were pronounced for any traits.

The interaction of genotype by location (G×L) was significant for GY, CH and WFL (Additional file 4).

Heritability of the traits

Heritabilities of the traits were calculated per growing location (Table 6.2). For most of the traits, the heritability was rather similar in both farming systems and all four locations. In all locations, heritability values were high (> 0.50) for GWH, WFL, LAN, and HED. Heritabilities were very low for NT and CGC.

Correlation among the traits

Pearson's correlations (r) between the traits were calculated within each farming system over the three years to evaluate relationships among phenotypic traits (Additional file 5). The traits involved in WSA such as CH and CGC had a stronger positive correlation with GY in the organic (0.38 and 0.53, respectively) than in the conventional farming system (0.19 and 0.32, respectively).

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Table 6.1. Mean values for grain yield and traits contributing to weed suppressive ability, of 153 barley genotypes grown in two organic (O1, O2) and two conventional (C1, C2) locations over three years (2010-2012).

Traits	Growing location			
	O1	O2	C1	C2
GY ¹	2.74	1.8	3.44	4.66
Traits contributing to weed suppressive ability (WSA)				
GWH	5.2	5.0	4.8	4.9
CH	19	19	24	25
CGC	25	23	42	46
LAN	4.4	4.3	4.1	4.7
LFL	11.2	10.6	11.5	11.9
WFL	7.8	7.9	8.4	8.5
PH	75	70	84	82
NT	459	380	497	529

¹ Traits: GY = grain yield; GWH = plant growth habit; CH = canopy height; CGC = crop ground cover; CGH = plant canopy growth habit; LAN = leaf angle; LFL = length of flag leaf; WFL = width of the flag leaf; PH = plant height before harvest; NT = number of tillers

Table 6.2. Heritability (h^2) for the traits contributing to weed suppressive ability over 153 barley genotypes grown in two organic (O1, O2) and two conventional (C1, C2) locations.

Traits	h^2 (O1)	h^2 (O2)	h^2 (C1)	h^2 (C2)
GY ¹	0.35	0.38	0.34	0.43
GWH	0.46	0.55	0.53	0.58
CH	0.50	0.33	0.68	0.45
CGC	0.02	0.10	0.08	0.06
LAN	0.33	0.41	0.63	0.57
LFL	0.26	0.26	0.35	0.38
WFL	0.45	0.50	0.52	0.54
PH	0.29	0.28	0.22	0.26
NT	0.02	0	0.01	0.03

¹ Traits: GY = grain yield; GWH = plant growth habit; CH = canopy height; CGC = crop ground cover; CGH = plant canopy growth habit; LAN = leaf angle; LFL = length of flag leaf; WFL = width of the flag leaf; PH = plant height before harvest; NT = number of tillers.

Other traits included in the WSA, such as LAN, LFL, WFL, and PH have little effect on GY in both farming systems. In organic farming systems, NT had a slightly positive correlation with GY and with CGC. The NT may either directly contribute to GY because of more spikes per m² or indirectly because high tillering may help to suppress weeds. However, the NT had a slightly negative correlation with WSA in both farming systems. WSA includes

morphological traits CH, LAN, LFL, WFL, and PH with which NT correlates negatively. In this trial the genotypes that developed faster, reaching tall canopy in early growing stages and having long and broad leaves, tended to tiller less than short genotypes with narrow and stature leaves.

Population structure and linkage disequilibrium analyses

A Principal Components Analysis (PCA) was performed on the genotypic dataset using GAPIT3 to study population structure. Four individuals (the 6-row Latvian accessions) had a strong relationships and formed a compact cluster in the PCA plots, separate from the other accessions (Fig. 6.1). To mitigate genetic stratification and reduce the risk of false positives in our GWAS, these four 6-row Latvian accessions were removed. Then 137 individuals remained in the GWAS analysis. After removing the four 6-row Latvian accessions the first three principal components explained only about 16% of the total genetic variance.

According to the linkage disequilibrium (LD) analysis performed for each of the seven chromosomes, LD decayed below 0.2 in 6, 5, 5, 5, 4, 5 and 6 cM, respectively. According to these distances, significant SNPs were grouped and considered to be linked to the same QTL when their distances fell within these estimated LD distances.

Multi-locus GWAS analysis

In order to test whether the statistical model reliably detects QTLs of traits, a GWAS analysis for hullessness was conducted using the BLINK model. Hullessness in barley has been mapped and is controlled by the *Nud* gene 1 on chromosome 7H bin 7 (Taketa et al., 2006; Mezaka et al., 2011). Ten SNPs were significantly associated with hullessness in the BLINK model (Additional file 6).

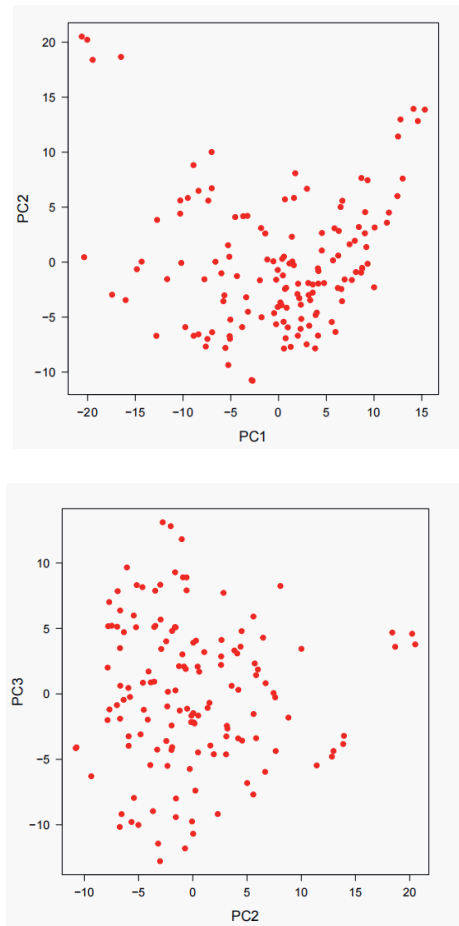


Fig. 6.1. Principal Components Analysis (PCA) on the genotypes of the barley GWAS accession panel.

The presence of a hull was significantly associated with markers located on chromosomes 1H, 3H, 6H, and 7H. The peak marker was located on chromosome 7H, in the *Nud* gene location region, which is consistent with Mezaka et al. (2011) thus suggesting that the BLINK model can detect significant SNPs well. Markers detected on other chromosomes were less significant.

For each trait, significant markers (FDR adjusted p-value < 0.05 as threshold) mapping within a LD decay distance up to 4 - 6 cM, depending on the

chromosome, were combined into a single QTL. Thus, in total 35 QTLs were identified for the four traits at four locations (C1, C2, O1 and O2) (Table 6.3).

Table 6.3. Summary of the number of QTLs discovered in the GWAS analyses (FDR adjusted p-value < 0.05) for all traits under organic and conventional farming systems.

Trait	Total number of QTLs	The number of QTLs found in organic farming only	The number of QTLs found in conventional farming only	The number of QTLs found in both farming systems
CH ¹	9	2	6	1
LAN	9	3	3	3
WFL	13	3	8	2
PH	4	2	1	1
Total	35	10	18	7

¹Traits: canopy height at stem elongation stage (CH); leaf inclination angle (LAN); the width of flag leaf (WFL); plant height at harvest (PH);

More detailed information on all significant marker-trait associations is provided in Additional file 8.

Most of the QTLs (80%) were identified only in one farming system, of which 28% in the organic farming system only, 52% in the conventional farming system only. 20% of the QTLs were found in both farming systems (Table 6.3).

The peak marker for grain yield was on chromosome 7H, in the region of the *Nud* gene (Additional file 8) (Mežaka et al., 2011). The *Nud* gene has previously been reported to be associated with grain yield, with hulled barley having higher yields than hulless barley (Barabaschi et al., 2012). We found several recombinant accessions for the *Nud* locus and the peak marker ConsensusGBS0132-4. Marker ConsensusGBS0132-4 allele A was associated with hulled grains and higher grain yield. We suppose the significant QTL identified for GY was contributing to hulled *versus* hulless grains.

Nine QTLs for **CH** were found on chromosomes 2H (2 QTLs), 3H (3 QTLs), 5H (1 QTL), 6H (2 QTLs) and 7H (1 QTL) (Additional file 7). Among these QTL 4025-300 explained the highest proportion of phenotypic variations (> 15%) and had the greatest effect on CH (Additional file 7). This QTL 4025-300 mapped on chromosome 3H at 117 cM and was identified in the conventional farming system. The allele A of QTL 4025-300 was associated

with higher canopy height at the stem elongation stage (CH) (Additional file 9).

Three QTLs were identified in the organic farming system, among these QTLs ABC38781-pHv2346-01 explained >15% of the phenotypic variation, while the other two had small effects.

Nine QTLs on chromosomes 2H (3 QTLs), 3H (2 QTLs), 4H (1 QTL), 6H (2 QTLs) and 7H (1 QTL) were significantly associated with **LAN** in both farming systems (Additional file 7, Table 6.3). The same marker QTL 4025-300 which explained the largest proportion of the variation for CH was also significantly associated with LAN, both in organic and conventional locations. Besides, another QTL 3718-1026 on chromosome 3H at 132 cM also explained a considerable fraction (>18%) of the phenotypic variation for LAN at two locations (O2, and C1). The allele A of QTL 4025-300 and the allele C of QTL 3718-1026 were related to more prostrate leaf inclination angle (Additional file 9).

Thirteen significant QTLs were found for **WFL** and were located on the chromosomes 1H (2 QTLs), 2H (3 QTLs), 3H (3 QTLs), 4H (1 QTL), 5H (2 QTLs), 6H (1 QTL) and 7H (1 QTL) (Additional file 9). Most of them were found under conventional cultivation. A QTL 5202-1199 on chromosome 7H at 77.85 cM explained the largest amount of phenotypic variation (16%) and had a relatively small (-0.63 mm) additive effect on WFL in the conventional farming system (Additional file 7). Another QTL 5286-486 mapped on chromosome 5H at 142 cM also had a large explained variance (14%) and had a small additive effect on WFL in conventional farming (+0.55 mm). In the organic farming system, only QTL 5251-184 on 6H at 64 cM had the highest phenotypic variation (> 13%) and a considerable additive effect (-0.82 mm) on WFL.

Four QTLs located on chromosomes 2H (2 QTLs) and 3H (2 QTLs) were significantly associated with **PH** (Table 6.3, Additional file 7). The previously mentioned QTLs 4025-300 and another QTL 3718-1026 were found associated with plant height at harvest in organic farming. The QTLs 4025-300 explained 16% of phenotypic variation and had a higher (+4.33 cm) additive effect on plant height than OTL 3718-1026 (+2.61 cm) in the organic farming system. One QTL 4434-804 was found in the organic farming system and explained the highest proportion of phenotypic variation (> 10%) and had

the largest additive effect (- 4.89) on PH in the organic system. The allele G of QTL 4434-804 on chromosome 2H at 68 cM had a positive effect on the plant height at harvest (PH) in the organic farming system only (Additional file 9). QTL 4025-300, previously found associated with CH and LAN, detected for PH in organic and conventional farming systems. This QTL explained a high proportion of phenotypic variation ($> 10\%$), and the additive effect of this QTL was slightly higher in the conventional (ranging from +4.30 to +5.27) than in the organic (4.33) farming systems.

6.4 Discussion

The growing demand for products grown in organic farming system promotes the need for varieties suitable for this type of farming. In developing genotypes for organic farming with high weed suppressive ability, we are interested in detecting QTLs for traits contributing to weed suppressive ability. In addition, we are interested to know whether QTLs identified in conventional farming are in common with the QTLs detected in organic farming systems. We carried out a GWAS to study this.

The main factors for the formation of barley population structure are growth habit, row type, and geographical origin (Pasam et al., 2012). Since in this study only spring barley was included and the countries of origin of the barley accessions were Baltic or Nordic regions, population structure was expected to be weak. Consistently, the first two PCs of our analysis were both related to row type, highlighting the only four six-row (Latvian) accessions in the panel. Since population structure can cause false-positive associations in GWAS, these four six-row Latvian individuals were removed.

The GWAS on barley was conducted using phenotypic data from two organic and two conventional locations. The phenotypic data analysis indicated that grain yield and the traits contributed to weed suppressive ability differ between locations in both farming systems. In addition, the heritability estimates were similar between locations. Using the phenotypic data over the three years, we identified a total of 35 QTLs, of which 7 QTLs were in common between the two farming systems, while 10 QTLs were only found in organic and 18 only in the conventional farming system. Thus, the results of GWAS study revealed that most of the QTL identified were management specific.

Of the nine traits we phenotyped, we found QTLs for only four of them (canopy height at the stem elongation stage, leaf inclination angle, width of flag leaf, plant height at harvest). Unfortunately, no QTL was identified for grain yield, and also none for crop ground cover, and length of the flag leaf, which are essential for weed suppressive ability in organic farming systems. The reason for not finding QTLs for crop ground cover could be the low heritability. However, for other traits (grain yield, plant growth habit, and length of the flag leaf) main reasons could be: i) marker coverage may still be insufficient, especially for some regions with multiple large gaps (Close et al., 2009); ii) the expression of the phenotypic traits may depend on a large number of QTLs with small effects that cannot be detected (lack of adequate statistical power) (Pasam et al., 2012); iii) environmental influences such as diseases, pests, available nitrogen in the soil, humus content, and the fertility management may have decreased the detection power as well.

In the following, we focus on the QTLs with the highest phenotypic variance on the traits of interest. Only the two QTLs 4025-300 and 3718-1026 were in common between the organic and conventional farming systems: they both were located on chromosome 3H and were associated with leaf inclination angle. The QTL 4025-300 was associated with plant height at harvest in both farming systems, while QTL 3718-1026 was related to the plant height only in the organic system. In another barley study, Bai et al. (2021) found that genes linked to plant height may act differently in different environments depending on the environment and experimental management. QTL 4025-300 accounted for 15.4% of phenotypic variation for the plant height in the organic system, while 3718-1026 explained 5.4% of the phenotypic variance in the same system. In addition, the additive effect of the QTL 4025-300 was higher than the additive effect for QTL 3718-1026 (Additional file 7). Both identified QTLs 4025-300 and 3718-1026 under organic farming explained a large amount of phenotypic variation (>19%) for leaf inclination angle, similar to that in conventional farming (>18%) for these QTLs. Thus, we can suppose that both QTLs can be used in the marker-assisted selection for leaf inclination angle for organic farming. In addition, QTL 4025-300 was associated with canopy height at the stem elongation stage in the conventional farming system only and the proportion of phenotypic variation of this marker was high (17.8% - 20.8%). Canopy height at the stem elongation stage is a highly

relevant trait in organic farming because it helps the crop to compete with weeds at this stage of development (Piliksere et al., 2013; Kissing Kucek et al., 2021a and b). QTL 4025-300 was not found for canopy height at the stem elongation stage in the organic farming system, probably due to management practice (dominance of cereals and perennial grasses in the crop rotation, no weed control, the incidence of pests and because this was estimated visually which is quite difficult at this development stage). In our study, canopy height at the stem elongation stage positively correlated to plant height at harvest within each farming system. Our results indicate that the QTL detected for canopy height at the stem elongation stage is management-specific (identified in the conventional farming system only). Therefore we presume that the QTL 4025-300 may have practical importance for conventional breeding purposes to select genotypes competitive against weeds to reduce the use of herbicides. In marker-assisted selection for organic purposes, the better solution could be QTL ABC38781-pHv2346-01. Most the QTLs found for the width of flag leaf were identified in either the conventional or the organic system only, with one in common between the systems.

Plant height at harvest is an essential trait contributing to weed suppressive ability (Murphy et al., 2008; Andrew et al., 2015; Mahajan et al., 2020). QTL 4434-804 was significant for the plant height at harvest in the organic farming system only and it mapped on chromosome 2H (68.2 cM) close to the semi-dwarfing gene *sdw3* region (~71cM) (Gottwald et al., 2004). In our study, QTL 4434-804 explained 13.2 to 15.4% of the phenotypic variance and the allele A was associated with taller plants at harvest by approximately 8 cm compared to allele G. The GWAS panel included varieties registered over the course of more than a century, with some as early as 1901. During this period of time, barley breeding experienced the so-called "green – revolution" – the introduction of dwarfing and semi-dwarfing genes (Kuczynska et al., 2013). In our study the canopy height at the stem elongation stage and plant height at maturity determining QTLs 4025-300 and 3718-1026 were mapped on 3H at 117 cM and 132 cM respectively, near to the genome region harboring the semi-dwarfing gene *sdw1/denso* (127 cM) (Sharma, 2012; Xu et al., 2018). As mentioned above QTL 4434-804 co-localized with *sdw3* gene region on 2H (71 cM). Plants carrying these dwarfing and semi-dwarfing alleles are shorter, have shorter and more erect leaves, and are more resistant to lodging.

In addition, they have late heading and maturity and are higher yielding under high-input agricultural conditions (Kuczyńska et al., 2013).

For three traits (canopy height at the stem elongation stage, leaf inclination angle, width of flag leaf) that contribute to weed suppressive ability, identified QTLs co-localized with previously found genes for flowering time QTL (7H 38-43 cM): *HvFT1/Vrn-H3*, QTL (7H 78-88 cM): *HvCOI* (Pasam et al., 2012; Maurer et al., 2015). In addition, the previous study by Mason et al. (2007) proposed that early maturity is associated with the expression of weed suppression, positively affecting traits as canopy height and soil ground cover. Thus, the early genotypes, having early rapid development, allow achieving good crop ground cover in the first part of the growing period until heading. That suggested that early flowering genotypes could be beneficial under organic farming, as they can outcompete weeds and finally produce higher yields in organic farming than later ripening genotypes.

6.5 Conclusions

In this study, no QTLs were found for grain yield. QTLs for four traits contributing to weed suppression ability (canopy height at stem elongation stage, leaf inclination angle, the width of the flag leaf, and plant height at harvest) were identified using GWAS under organic and conventional growing conditions. Most QTLs detected were management specific, which suggests that it is necessary to do QTL discovery studies under a specific farming system. The QTL ABC38781-pHv2346-01 associated with canopy height at stem elongation stage, QTL 5128-146 associated with the width of the flag leaf and QTL 4434-804 for the plant height at harvest found in the organic farming systems could be used for marker-assisted selection under organic growing conditions (direct selection) and might also be useful within conventional breeding programmes (indirect selection) to select the genotypes for organic farming. QTLs found in common under organic and conventional growing conditions could be applied similarly to organic-specific QTLs either in direct and indirect selection. In a conventional breeding programme they would allow selection of genotypes for organic farming as well as for an integrated management system reducing the use of herbicides.

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Supplementary material

Additional file 1. Description of the barley accessions included in the association mapping panel in Priekuli 2010-2012.

Accession name	Pedigree	Grain type	Country of origin	Row type	Year of variety registration ¹	Particular traits of interest
12811	Austris/Danuta	Hulled	Latvia	2-row	2010	TCAP ² , CGC, TGW
12819	Madelon/Abava	Hulled	Latvia	2-row	2010	CGC, LOD
12820	Auriga/Kristaps	Hulled	Latvia	2-row	2010	TCAP, CGC
12825	Riviera/Comatry//Austris	Hulled	Latvia	2-row	2010	CGC, TGW, DR _{PMW}
1012786-41	814280-15/754277-27	Hulled	Latvia	2-row	1994	LFL _T
1079488-45	75615-73/827580-15//8993	Hulled	Latvia	2-row	1996	DEV_SPEED _R , LAN _D
1163691-34	Kvant/9024	Hulled	Latvia	2-row	1998	DEV_SPEED _R , CGC, LAN _D
1263098-13	1096689-33/1187292-26	Hulled	Latvia	2-row	2005	-
1267199-30	Candice/1181092-12//Candice	Hulled	Latvia	2-row	2006	GWH _E
1271100-26	SB 90201/Bor 94149	Hulled	Latvia	2-row	2007	DEV_SPEED _R , WFL
1272500-36	SV 86107/Manič 459	Hulled	Latvia	2-row	2007	LAN _D
1273300-50	Margit/9089	Hulled	Latvia	2-row	2007	DR _F
250 (PI436150)		Hulled	Chile	2-row	1979	LAN _D , PH _T
3280.14.14		Hulled	Estonia	2-row	2001	GWH _P , DEV_SPEED _{SL} , PH _{SH}
69 (Clhol1319)		Hulled	United Kingdom	2-row	1979	DEV_SPEED _R , WFL
718676-19	602969-5/Mirena	Hulled	Latvia	2-row	1983	LAN _D
743/09	Maaren/Justina	Hulled	Latvia	2-row	2012	TCAP, LAN _D , DR _{PMW}
754277-27	Rupal/k-21874//Ofir	Hulled	Latvia	2-row	1984	DEV_SPEED _R , LAN _D
768678-28	76-34/Ofir//Nadja	Hulled	Latvia	2-row	1985	-
797877-39	76-34/k-21874//Ofir	Hulled	Latvia	2-row	1986	DR _F
813380-13	641572-1/Romana	Hulled	Latvia	2-row	1987	-
827580-15	662573-7/Keg	Hulled	Latvia	2-row	1987	-
Abava	Mari/Elsa//Domen	Hulled	Latvia	2-row	1980	CGC, LAN _D , GY _{STAB}
Agra	Priekulu 1/Otra	Hulled	Latvia	2-row	1984	GR
Alsa	Mirena/mutant fom Gintariniai//Abava/Emir	Hulled	Lithuania	2-row	1996	DEV_SPEED _R , LAN _D
Annabell	Henni/Krona	Hulled	Germany	2-row	1999	TCAP
Anni	Lola/Liisa	Hulled	Estonia	2-row	1980	GWH _P , DEV_SPEED _{SL}
Ansis	Jarek/Taifun	Hulled	Latvia	2-row	2001	GR

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Aura		Hulled	Lithuania	2-row	1999	recommended in OF in LT
B-93	Rūja/Imula	Hulled	Latvia	2-row	2000	GR
Balga	Gunilla/KM-1192	Hulled	Latvia	2-row	1995	GR
Betzes		Hulled	Germany	2-row	1957	PH _T
Bor 88377		Hulled	Finland	2-row	1999	DEV_SPEED _R , MAT ^o _E , PH _{SH} , TGW
BZ12-63	Primus/Idumeja	Hulled	Latvia	2-row	2008	DEV_SPEED _R , CGC, GY
BZ12-83	Primus/Idumeja	Hulled	Latvia	2-row	2008	CGC, LAN _D
BZ12-86	Primus/Idumeja	Hulled	Latvia	2-row	2008	DEV_SPEED _R , GY, TGW, VOL, DR
BZ12-93	Primus/Idumeja	Hulled	Latvia	2-row	2008	CGC, GY, VOL
BZ14-12	Anni/Dziugiai	Hulled	Latvia	2-row	2008	GWH _E , DEV_SPEED ^o _R
BZ14-90	Anni/Dziugiai	Hulled	Latvia	2-row	2008	DEV_SPEED _R , CGC, LAN _D , LFL _T , WFL, PH _T , MAT _E
BZ14-99	Anni/Dziugiai	Hulled	Latvia	2-row	2008	GWH _P , DEV_SPEED ^o _R
Camila		Hulled	Germany	2-row	1974	CGC, DR _{PMW}
Daghesta nicum		Hulless	United Kingdom	2-row	1960	WFL, TGW
Danuta		Hulled	Germany	2-row	1996	GWH _P
Divosnoje		Hulled	Belorus	2-row	2001	LAN _E
Druvis	Dobrij/HVS 115440	Hulled	Latvia	6-row	1999	GWH _E
Dzintars	Selection from Latvian local (Vidzeme)	Hulled	Latvia	6-row	1930	DEV_SPEED _R , WFL, MAT _E
Dziugiai		Hulled	Lithuania	2-row	1947	GWH _E , MAT _E , DEV_SPEED ^o _R , PRO
Eunova		Hulled	Austria	2-row	2000	DR _{PMW} , recom. for OF
G-131 (Austri is)	Ansis/WW8208	Hulled	Latvia	2-row	2012	LAN _E
Gāte	Emir/2*Nadja//HE-497/Hadmersleben 70197/70	Hulled	Latvia	2-row	2000	GR
Golf	Armelle/Lud//Luke	Hulled	United Kingdom	2-row	1986	DEV_SPEED _R , LAN ^o _D
H 130	Filippa/Idumeja	Hulled	Latvia	2-row	1999	
Heils Hanna		Hulled	Czech Republic	2-row	1909	DEV_SPEED _R , LAN ^o _P , PH _T
Hellana		Hulled	Austria	2-row	1995	LAN _D
Heris		Hulled	Czech Republic	2-row	1998	GWH _P , WFL
Iakub		Hulled	Belorus	2-row	2001	-
Idumeja	Imula/Ida	Hulled	Latvia	2-row	2003	TCAP, DEV_SPEED ^o _R , CGC
Ilga	KM-1192/Hadmersleben 70197/70	Hulled	Latvia	2-row	1983	GR
Imula	Abava/2*Akka	Hulled	Latvia	2-row	1990	GR
Inari		Hulled	Finland	2-row	1994	-
Justina		Hulled	Germany	2-row	1999	GWH _P , LAN _D , DEV_SPEED ^o _{SL}
Klinta	Torkel/CF-42	Hulled	Latvia	2-row	1998	GR
Kombaini eris	Maja/Talsu local	Hulled	Latvia	2-row	1955	GR
Kristaps	CF 79502/902383-48	Hulled	Latvia	2-row	2006	GR
L-2295	Gāte/KM-1192	Hulled	Latvia	2-row	1996	GWH _P
L-2544	Nancy/Dina	Hulled	Latvia	2-row	2000	GR
L-2630	Luna/Rasa	Hulled	Latvia	2-row	1997	GWH _P , DEV_SPEED ^o _R
L-2735	Ida/Sv.8329	Hulled	Latvia	2-row	1997	GR

L-2985.1	L-2025/L-2233 (Imula/Ida, St12128/Athos2/Ida)	Hulled	Latvia	2-row	1998	DEV_SPEED _R , WFL
L-3101	Linga/Run8/453//Linga	Hulled	Latvia	2-row	2001	LAN _E
L24		Hulless	Germany	2-row	2002	DEV_SPEED _R , LAN ^o _D , PH _T
Latvijas Vietējie	Latvian local landrace	Hulled	Latvia	2-row	1900	CGC, PH _T , MAT _L , PH ^o _T
Lawina		Hulless	Germany	2-row	2000	CGC, LAN _D , CH
Leeni		Hulled	Estonia	2-row	2006	DEV-SPEED _{SL} , recommended. for OF in EST
Linga	Gunilla/KM-1192	Hulled	Latvia	2-row	1990	GR
Lysiba	Lamba/SJ 900691	Hulled	Denmark	2-row	1997	GWH _P , PH _{SH}
M9		Hulled	Latvia	2-row	2012	GWH _P , TCAP, DR ^o _{PMW}
Maaren		Hulled	Sweden	2-row	2005	GWH _P , TCAP
Malva	STN8142/STN7542	Hulled	Latvia	2-row	2001	GR
Mik 1		Hulled	Russia	2-row	1990	DR _U NUDA
No.51		Hulless	Germany	2-row	2000	CGC, recom. for OF in Germany
No.79		Hulless	Germany	2-row	2000	WFL, DR _{PMW}
Nuevo		Hulled	Denmark	2-row	2007	GWH _E
Otira	Bartok/SJ 930331	Hulled	Denmark	2-row	1996	GWH _P , LAN _D
Pallas	Mutation selected from X-ray treated Bonus	Hulled	Sweden	2-row	1958	DEV_SPEED _R
Peggy		Hulled	Germany	2-row	1995	GWH _P , LAN _E
Pervonez		Hulled	Ukraine	2-row	1981	DEV_SPEED _R
Jumara (PR3005)	Baronesse/L-2380 (Ww7291/Dina)	Hulled	Latvia	2-row	2010	GY
PR-3134	Rūja/Baronesse//Baronesse/Ida	Hulled	Latvia	2-row	2002	-
PR-3223	Alexis/Sencis//Sencis	Hulled	Latvia	2-row	2004	WFL _N , GY
PR-3245	97B741sex msg6/Thuringa//Gāte	Hulled	Latvia	2-row	2004	-
PR-3282	Ivana/Idumeja	Hulled	Latvia	2-row	2004	-
PR-3297	Lysimax//Linga/#112	Hulled	Latvia	2-row	2004	CGC, LAN _D
PR-3300	Tolat//Linga/#112	Hulled	Latvia	2-row	2004	GWH _P , CGC
PR-3351	Lysimax/Linga	Hulled	Latvia	2-row	2004	GWH _P , CGC
PR-3474	Abava/Sw1290//L-2421	Hulless	Latvia	2-row	2005	TCAP
PR-3475	Abava/Sw1290//L-2421	Hulless	Latvia	2-row	2005	GWH _P , DEV_SPEED ^o _{SL}
PR-3512	Linus/Annabell//L-2421 (Rūja/Bingo)	Hulled	Latvia	2-row	2005	DEV_SPEED _{SL} , WFL
PR-3515	Mette/Tolar	Hulled	Latvia	2-row	2005	GWH _P , DEV_SPEED ^o _{SL} , CGC
PR-3518	L-2905/L-2503 (Dina/Run,Gastinec/Imula)	Hulled	Latvia	2-row	2005	TCAP, CGC
PR-3520	Ivana/Idumeja	Hulled	Latvia	2-row	2005	DEV_SPEED _R
PR-3522	Sencis//P3 645 C/L-2233	Hulled	Latvia	2-row	2005	DEV_SPEED _R
PR-3527	Abava/Sw1290//L-2421	Hulless	Latvia	2-row	2005	TCAP, GWH _P , DEV_SPEED _{SL}
PR-3528	Filippa/McGwire//Kristaps (Irbe)	Hulless	Latvia	2-row	2011	GR
PR-3537	Merlin/Linga//Sencis	Hulless	Latvia	2-row	2005	GWH _P , DEV_SPEED ^o _{SL}
PR-3605	Rūja/Prestige/3/L-2233//Linus/Annabell	Hulled	Latvia	2-row	2006	DEV_SPEED _R , CGC, CH, LAN _D , GY
PR-3636	Rubiola/L-2735 Un8	Hulled	Latvia	2-row	2006	CH, GY
PR-3749	CIMMYT48/Richard	Hulless	Latvia	2-row	2006	CGC, LAN _D , LFL _T
PR-3885	Danuta/L-3105 (Latv.viet/Ww) mlo11	Hulled	Latvia	2-row	2007	CGC, LAN _D

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PR-4115	Tunika/L-3118 mlo11	Hulled	Latvia	2-row	2007	GWH _P , LAN _D , DR ^o PMD/mlo
PR-4121	Tunika/L-3118 mlo11	Hulled	Latvia	2-row	2007	GWH _P , LAN _D , DEV_SPEED ^o R, CGC, TGW, DR ^o PMD/mlo
PR-4144	Rubiola/L-3118 mlo11	Hulled	Latvia	2-row	2007	WFL, LAN _D , DR ^o PMD/mlo
PR-4181	Hydrogen/H-155	Hulled	Latvia	2-row	2007	CGC, LAN _D
PR-4362	Abava/Gainer//Nordus mlo11	Hulless	Latvia	2-row	2007	DEV_SPEED _R , LAN ^o D, DR ^o PMD/mlo
PR-4368	Abava/Gainer//Nordus mlo11	Hulless	Latvia	2-row	2007	DEV_SPEED _R , LAN ^o D, DR ^o PMD/mlo
PR-4760	Richard/Peggy	Hulless	Latvia	2-row	2008	DEV_SPEED _R , WFL ^o N, LAN _E , PH _T , DR ^o PMW
PR-4803	Ansis/Dziugiai B	Hulled	Latvia	2-row	2008	GWH _E , CH, DEV_SPEED ^o R, PH _T , VOL
PR-4810	Rubiola/L-3118BB	Hulled	Latvia	2-row	2008	DEV_SPEED _R , CH, MAT _E
PR-4812	Rubiola/L-3118 BB mlo11	Hulled	Latvia	2-row	2008	DEV_SPEED _R , CGC, CH, DR ^o PMD/mlo, GY
PR-4814	Danuta/L-3008//Rubiola BB	Hulled	Latvia	2-row	2008	DEV_SPEED _H , CGC, GY, TGW, VOL
PR-4822	Abava/Annabell BZ	Hulled	Latvia	2-row	2008	DEV_SPEED _R , CGC, LAN _D
PR-4832	Latvijas vietējie/Inari BB	Hulled	Latvia	2-row	2008	CGC, LAN _D , MAT _L
PR-4835	Rubiola/L-2735	Hulled	Latvia	2-row	2008	DEV_SPEED _R
PR-5105	Rubiola/L-3118//Millena/L-2901 B	Hulled	Latvia	2-row	2009	LFL, TGW
PR-5108	Rubiola/L-3118//L-91 mlo11	Hulless	Latvia	2-row	2009	DEV_SPEED _R , LAN ^o D, VOL, DR ^o PMD/mlo
PR-5109	Roxane/Danuta//Idumeja/3 /L-47	Hulless	Latvia	2-row	2009	CGC, CH
PR-5112	Lawina/3/Silky/CIMMYT-120//Milton	Hulless	Latvia	2-row	2009	CH, VOL
PR-5117	Rubiola/L-3118//L-2985 B	Hulled	Latvia	2-row	2009	CH
PR-5127	Rubiola/L-3118//Australian Early B	Hulled	Latvia	2-row	2009	CGC, LAN _D , PH _T , TGW
PR-5131	Rubiola/L-3101//L-3005 B	Hulled	Latvia	2-row	2009	CH, TGW, VOL
PR-5135	Abava/Annabell//Rubiola inf.F3	Hulled	Latvia	2-row	2009	CGC, LAN _D , TGW, VOL
PR-5137	Abava/Annabell//Rubiola	Hulled	Latvia	2-row	2009	CH, TGW, VOL
PR-5145	Peggy/L-3118//Rubiola B	Hulled	Latvia	2-row	2009	LAN _E , DR ^o PMW
Priekuļu 1	Selection from Norwegian local varieties	Hulled	Latvia	6-row	1959	GR
Priekuļu 60	Tammi/2*(Talsu local/2*Maja)	Hulled	Latvia	2-row	1972	GR
Primus	Selection from Plumage	Hulled	Sweden	2-row	1901	GWH _P , LAN _D
Rasa	Frankengold/KM-R-54/72	Hulled	Latvia	2-row	1996	DEV_SPEED _R , LAN ^o D
Roxana		Hulled	Germany	2-row	2000	DR ^o PMD/mlo
Rubiola	Rūja/Run8/458	Hulled	Latvia	2-row	2011	GR
Rūja	Abava//Kombainieris/Tru mph	Hulled	Latvia	2-row	1996	GR
Sencis	Rupal/Ofir//Torkel	Hulled	Latvia	2-row	2000	GR
Steffi		Hulled	Germany	2-row	1989	LAN _D
Stendes	Drost/Maja	Hulled	Latvia	2-row	1972	GR
SZD 4748		Hulled	Austria	2-row	2001	WFL
Thuringia		Hulled	Germany	2-row	1995	LAN _D
Tocada		Hulled	Germany	2-row	2004	LAN _D
Ula	Roland/CA33787	Hulled	Lithuania	2-row	1996	DEV_SPEED _R , LAN ^o D

V-08-83		Hulless	Germany	2-row	2001	PH _T , GY
Vada		Hulled	Netherland	2-row	1958	LAN _D
s						
Vairogs	Selection from Priekulu local	Hulled	Latvia	6-row	1930	GR
Verena		Hulled	Germany	2-row	2000	CGC, DR _{PMW}
Vienna		Hulled	Austria	2-row	2007	GWH _P , DEV SPEED ² _{SL} , CH
ZNCF		Hulless	Germany	2-row	2001	CH, CGC, LAN _D

¹ Year of variety registration/ line included in yield trials

² trait and trait index abbreviation, what highlights the expression of the respective trait : GWH growth habit (E – erect, P prostrate); TCAP – good tillering capacity; DEV_SPEED early developments speed (SL – low, R – rapid), CH – tall canopy; CGC – good crop ground cover; LAN leaf angle (D – declined, E – erect); LFL – tall flag leaf (SH – short); WFL – wide flag leaf (N – narrow leave); PH plant height (SH – short plants, T – tall plants); LOD – high lodging resistance; MAT – length of growth period from sowing to maturity (E – early, L – late); GY – high grain yield (STAB - stable yield); DR – diseases resistance (F – *Fusarium* subspecies, PMW – *Powdery mildew*, PMW/mlo - *Powdery mildew* on mlo based resistance, U_NUDA – *Ustilago nuda*); TGW – high thousand grain weight; VOL – high volume weight; PRO – high protein content, GR – genetic resource stored at the Latvian Gene Bank

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Additional file 2.Characterisation of soil and crop management practices in two organic (O1 and O2) and two conventional (C1 and C2) growing sites

Location	Year, sowing data	Soil type	Soil texture	pH _{KCl}	P ₂ O ₅ (mg kg ⁻¹)	K ₂ O (mg kg ⁻¹)	Humus content (g kg ⁻¹)	Available N content in soil in spring (kg ha ⁻¹ according to the humus content)	Fertility management		Management of	
									Amount of N, P, K	Type	Diseases, pests	Weeds
O1	2010 21.04	Sod	Loamy podzolic sand	5.7	111	144	28	75.6	Peas for green manure	Green manure, approx. 20 t ha ⁻¹ (precrop)	No	1 × harrowing at tillering stage
	2011, 21.04	Sod	Loamy podzolic sand	5.4	116	98	21	56.7	Peas for green manure	Green manure, approx. 20 t ha ⁻¹ (precrop)	No	1 × harrowing at tillering stage
	2012 28.04	Sod	Loamy podzolic sand	5.7	160	93	23	62.1	Peas for green manure	Green manure, approx. 20 t ha ⁻¹ (precrop)	No	1 × harrowing at tillering stage
O2	2010, 19.04	Sod	Loamy podzolic sand	6.5	265	173	35	94.5	Perennial grasses	Stable manure 40 t ha ⁻¹	No	No
	2011 20.04	Sod	Loamy podzolic sand	6.6	394	167	30	81.0	Spring wheat	- -	No	No
	2012 28.04	Sod	Loamy podzolic sand	5.6	171	129	23	62.1	Perennial grasses	- -	No	No
C1	2010 30.04	Sod	Loamy podzolic sand	5.5	100	132	26	70.2	Potatoes	Inorganic N 81 kg ha ⁻¹ P 48 kg ha ⁻¹ K 84 kg ha ⁻¹	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹

2011 27.04	Sod podzolic sand	Loamy	5.4	187	165	30	81.0	Potatoes	N 83 kg ha ⁻¹ P 45 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
2012 2.05	Sod podzolic loam	Sandy	5.7	264	155	27	72.9	Potatoes	N 80 kg ha ⁻¹ P 10 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic	Insecticide Decis 2.5 e.c. 0.15 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
2010 3.05	Sod podzolic sand	Loamy	5.6	115	159	28	84.3	Potatoes	N 120 kg ha ⁻¹ P 48 kg ha ⁻¹ K 84 kg ha ⁻¹	Inorganic	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
2011 29.04	Sod podzolic loam	Sandy	4.5	200	155	23	62.1	Potatoes	N 120 kg ha ⁻¹ P 45 kg ha ⁻¹ K 75 kg ha ⁻¹	Inorganic	Insecticide Karate 0.2 l ha ⁻¹	Herbicide Secator 0.15 l ha ⁻¹
2012 11.05	Sod podzolic loam	Sandy	5.4	166	175	17	45.9	Potatoes	N 120 kg ha ⁻¹ P 30 kg ha ⁻¹ K 45 kg ha ⁻¹	Inorganic	Herbicide Secator 0.15 l ha ⁻¹ and Estets 600 e.k. 0.5 l ha ⁻¹	

C2

Genome wide association mapping for grain yield and weed suppressive ability in barley (Hordeum vulgare L.) under conventional and organic conditions

Additional file 3. Traits evaluated during the experiment at two organic O1 and O2 and two conventional C1 and C2 growing sites, in 2010 – 2012.

Trait	Abbreviation	Growing stage according to Zadoks et al. (1974)	Unit	Remarks
Grain yield	GY	after harvest	t ha ⁻¹	Measured per full plot (3.7 m ²), the yield was expressed in tonnes ha ⁻¹ after drying and cleaning with 1.8 mm sieve
Traits contributing to weed suppressive ability				
Plant growth habit	GWH	25 - 29	scores	with 1 = erect to 9 = prostrate
Canopy height at stem elongation stage canopy	CH	31-32	cm	measured in cm for five plants per plot
Crop ground cover	CGC	31 - 32	%	visually estimated percentage of plot area covered by plants
Length of flag leaf	LFL	47 – 51	cm	measured in cm for five plants per plot
Width of flag leaf	WFL	47 – 51	cm	measured in cm for five plants per plot
Leaf angle	LAN	47 – 51	scores	flag leaf inclination angle was scored: 1= stature to 9 = declined
Plant height before harvest	PH	90	cm	measured in cm based on five plants per plot
Number of tillers	NT	90		The number of productive tillers was counted in 0.05 m ² plot area

Additional file 4. *P* -values (from four locations ANOVA) for farming system, genotype, year and location main effects and their interaction (significant effects at a 95% confidence level are marked in bold).

Traits	Abbreviation	Genotype (G)	Year (Y)	Four locations (LF)	Genotype × Year (G×Y)	Genotype × four Locations (G×LF)	Year × four Locations (Y×LF)
Grain yield	GY	<.001	<.001	<.001	0.003	0.006	<.001
Plant growth habit ¹	GWH	<.001	<.001	<.001	0.001	0.329	<.001
Canopy height ¹	CH	<.001	<.001	<.001	<.001	0.002	<.001
Crop ground cover ¹	CGC	<.001	<.001	<.001	<.001	0.167	<.001
Leaf angle ¹	LAN	<.001	<.001	<.001	<.001	0.901	<.001
Length of flag leaf ¹	LFL	<.001	<.001	<.001	<.001	0.100	<.001
Width of flag leaf ¹	WFL	<.001	<.001	<.001	<.001	0.008	<.001
Plant height ¹	PH	<.001	<.001	<.001	0.006	0.551	<.001
Number of tillers ¹	NT	<.001	<.001	<.001	0.015	0.650	<.001

¹ Traits contributing to weed suppressive ability (WSA)

Genome wide association mapping for grain yield and weed suppressive ability in barley (Hordeum vulgare L.) under conventional and organic conditions

Additional file 5. Correlations between all traits under organic and conventional farming systems. Traits: grain yield (GY), plant growth habit (GWH), canopy height at stem elongation stage (CH), crop ground cover (CGC), leaf inclination angle (LAN), length of flag leaf (LFL), the width of flag leaf (WFL), plant height at harvest (PH), number of productive tillers (NT), weed suppressive ability(WSA)

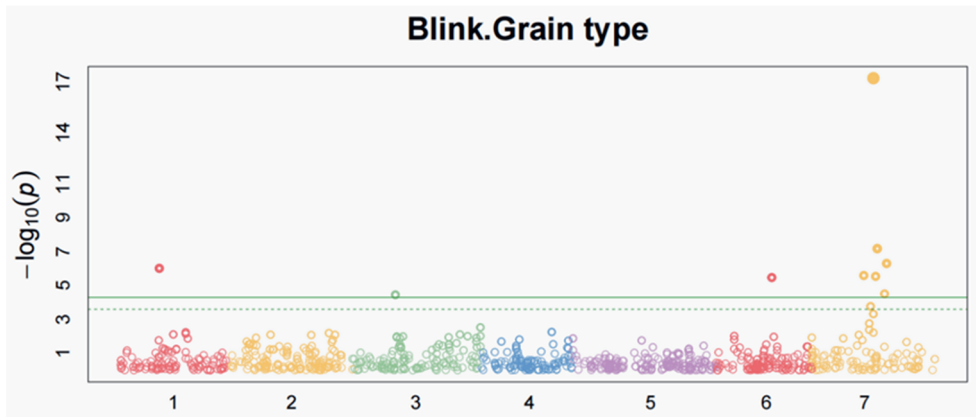
Organic condition

	GY	GWH	CH	CGC	LAN	LFL	WFL	PH	NT	WSA
GY	1.00									
GWH	-0.16	1.00								
CH	0.38	-0.75	1.00							
CGC	0.53	0.19	0.24	1.00						
LAN	0.14	-0.17	0.49	0.21	1.00					
LFL	0.03	-0.30	0.37	-0.03	0.24	1.00				
WFL	-0.05	-0.49	0.41	-0.08	0.03	0.60	1.00			
PH	-0.01	-0.24	0.54	0.09	0.47	0.37	0.35	1.00		
NT	0.32	0.27	-0.08	0.30	-0.05	-0.23	-0.35	-0.25	1.00	
WSA	0.28	-0.50	0.83	0.35	0.67	0.65	0.56	0.74	-0.16	1.00

Conventional condition

	GY	GWH	CH	CGC	LAN	LFL	WFL	PH	NT	WSA
GY	1.00									
GWH	-0.01	1.00								
CH	0.19	-0.69	1.00							
CGC	0.32	0.18	0.35	1.00						
LAN	-0.01	-0.16	0.53	0.32	1.00					
LFL	-0.05	-0.33	0.51	0.26	0.30	1.00				
WFL	-0.16	-0.43	0.45	0.19	0.14	0.62	1.00			
PH	-0.13	-0.29	0.60	0.33	0.48	0.41	0.43	1.00		
NT	0.36	0.20	-0.26	0.00	-0.22	-0.37	-0.40	-0.41	1.00	
WSA	0.05	-0.39	0.81	0.60	0.66	0.72	0.66	0.76	-0.38	1.00

Additional file 6. Manhattan plot of hullessness trait under BLINK model. The X-axis is the position of SNPs in cM and the Y-axis is $-\log_{10}(P\text{-value})$. The solid line in the graph is the BC threshold, and the dashed line is the FDR threshold ($\alpha=0.05$). Peaks mean SNPs have strong associations with the trait. 1 to 7 are the chromosomes and a transition in colour is a transition to another chromosome.



Additional file 7. All significant SNPs summary table for GWAS of the traits contributing to weed suppressive ability under BLINK model at four locations (O1, O2, C1 and C2).

Trait	Location	SNP	Chromosome	Position (cM)	MA F	-log ₁₀ (P value) ¹	FDR_Adjusted P-values ²	Effect	R ² estimate phenotypic variation explained by SNP (compared with the full model)	R ² estimate phenotypic variation explained by SNP (with only one marker) ³
CH ⁴										
		4025-300	3	117.09954	0.32	9.70	1.97E-07	2.73	15.8%	20.8%
		3997-796	5	35.68661	0.18	5.81	0.0008	-1.46	3.9%	4.1%
		111-499	2	112.90614	0.37	5.15	0.0023	1.01	2.5%	3.1%
	C1	1213-1959	7	40.18189	0.44	4.75	0.0043	-1.87	7.2%	11.0%
		4611-178	6	28.3866	0.38	4.64	0.0045	-1.55	6.2%	7.3%
		7729-565	2	70.539	0.42	4.40	0.0065	-0.60	5.9%	1.1%
		3164-1386	6	34.40329	0.32	3.67	0.0298	2.04	7.4%	11.4%
	C2	4025-300	3	117.09954	0.32	7.01	9.49E-05	2.34	17.8%	17.8%
		5385-722	3	68.32246	0.24	7.63	2.31E-05	-0.71	4.8%	2.2%
		5088-59	2	113.47607	0.47	5.48	0.0016	0.78	4.9%	3.6%
	O1	ABC38781-pHv2346-01	3	159.458	0.40	4.63	0.0076	1.45	15.9%	11.8%
LAN-exclude 2010										
		4025-300	3	117.09954	0.32	5.92	0.0012	0.76	9.8%	23.2%
		3436-354	6	72.54215	0.30	5.47	0.0017	-0.57	9.4%	12.6%
	C1	1250-923	2	130.01072	0.22	5.03	0.0031	0.34	2.6%	3.5%
		3718-1026	3	131.59305	0.43	3.94	0.0278	0.64	7.4%	18.2%

	4025-300	3	117.09954	0.32	7.36	4.32E-05	0.72	23.3%	21.0%
C2	1250-923	2	130.01072	0.22	4.74	0.0076	0.40	4.8%	5.0%
	3164-1386	6	34.40329	0.32	4.64	0.0076	0.54	14.9%	11.3%
	7729-565	2	70.539	0.42	4.23	0.0144	-0.36	7.8%	5.8%
	4025-300	3	117.09954	0.32	6.11	0.0008	0.64	17.8%	19.5%
O1	2585-2901	7	42.60231	0.40	4.31	0.0238	-0.51	8.3%	13.9%
	482-1423	4	63.55991	0.33	4.06	0.0288	-0.04	5.2%	0.1%
	1250-923	2	130.01072	0.22	3.75	0.0440	0.28	5.0%	3.0%
	4025-300	3	117.09954	0.32	6.53	0.0003	0.71	12.6%	23.2%
O2	3608-2133	2	137.51261	0.33	5.39	0.0020	0.20	2.6%	1.8%
	3718-1026	3	131.59305	0.43	4.95	0.0037	0.65	9.6%	21.7%
WFL									
	3271-1422	2	125.46452	0.40	6.52	0.0003	0.42	6.4%	10.1%
	5202-1199	7	77.85	0.22	5.19	0.0032	-0.63	13.1%	16.3%
C1	963-386	3	93.43063	0.14	4.52	0.0083	-0.41	4.4%	4.7%
	1906-429	1	23.86327	0.39	4.47	0.0083	0.12	3.6%	0.8%
	5251-184	6	64.36243	0.09	4.17	0.0133	-0.76	6.7%	10.8%
	4403-885	3	162.14889	0.35	4.02	0.0155	-0.24	7.7%	3.1%
	963-386	3	93.43063	0.14	8.56	2.68E-06	-0.32	10.4%	3.2%
	1578-552	7	83.43973	0.23	6.89	6.34E-05	-0.59	8.4%	15.5%
	5286-486	5	142.19946	0.23	5.23	0.0019	-0.55	11.1%	14.3%
C2	11603-445	1	65.96373	0.38	5.01	0.0024	0.34	5.3%	7.1%
	ABC17314-1-1-226	2	145.02776	0.36	4.78	0.0032	-0.20	0.9%	2.4%
	1038-754	3	111.42159	0.45	3.58	0.0428	-0.15	6.0%	1.4%
	5251-184	6	64.36243	0.09	6.60	0.0002	-0.82	13.5%	14.4%
O2	2029-1143	2	26.52834	0.36	4.62	0.0119	0.13	3.5%	1.0%
	5428-146	5	166.63311	0.45	4.33	0.0152	-0.44	9.8%	12.8%

		1038-754	3	111.42159	0.45	4.05	0.0219	-0.20	5.4%	2.7%
		6215-539	4	48.5	0.44	3.92	0.0235	-0.24	5.3%	3.9%
PH										
	C1	4025-300	3	117.09954	0.32	7.51	3.03E-05	5.27	16.5%	16.5%
		4025-300	3	117.09954	0.32	6.92	0.0001	4.30	14.2%	13.9%
	C2	1250-923	2	130.01072	0.22	4.56	0.0134	2.30	3.4%	3.1%
		4434-804	2	68.23939	0.22	4.51	0.0282	-4.89	11.6%	13.2%
	O1	3718-1026	3	131.59305	0.43	4.24	0.0282	2.61	3.8%	5.4%
		4434-804	2	68.23939	0.22	8.06	8.57E-06	-4.89	10.8%	15.4%
	O2	4025-300	3	117.09954	0.32	4.61	0.0120	4.33	10.8%	15.4%

¹ marked bold are significant SNPs under Bonferroni correction (BC) (4.29)

² all significant SNPs under false discovery rate (FDR) adjusted p-value < 0.05 as threshold

³ r^2 (with only one marker) estimated phenotypic variation higher than r^2 (compared with the full model) in most cases, this is because the correlations between markers lead to part of the phenotypic variation of r^2 (with only one marker) also explained by other QTLs.

⁴ Traits: canopy height at stem elongation stage (CH), leaf inclination angle (LAN), the width of flag leaf (WFL), plant height at harvest (PH)

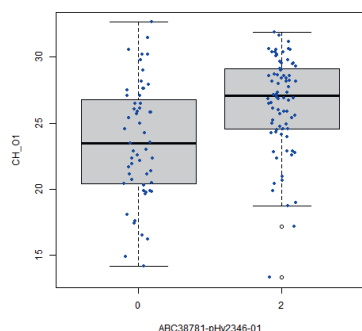
Additional file 8. Significant SNPs summary table for GWAS of grain yield (GY) under BLINK model at four locations (O1, O2, C1 and C2) (FDR adjusted p-value < 0.05 as threshold).

C1									
BLINK	SNP	Chromosome	Position	P value	MAF	nobs	R square of Model without SNP	R square of Model with SNP	FDR Adjusted P-values effect
1	ConsensusGBS 0132-4	7	84.91648	5.41E-14	0.116788321	137	NA	NA	5.31E-11 NA
2	4665-882	1	71.42785	5.07E-05	0.167883212	137	NA	NA	0.024870447 NA
C2									
BLINK	SNP	Chromosome	Position	P value	MAF	nobs	R square of Model without SNP	R square of Model with SNP	FDR Adjusted P-values effect
1	4594-971	7	83.43973	4.75E-19	0.102189781	137	NA	NA	4.66E-16 NA
2	1381-547	2	121.50150	5.76E-05	0.315907652	137	NA	NA	0.028262287 NA
O1									
BLINK	SNP	Chromosome	Position	P value	MAF	nobs	R square of Model without SNP	R square of Model with SNP	FDR Adjusted P-values effect
	4594-971	7	83.43973	7.50E-12	0.102189781	137	NA	NA	7.36E-09 NA
O2									
BLINK	SNP	Chromosome	Position	P value	MAF	nobs	R square of Model without SNP	R square of Model with SNP	FDR Adjusted P-values effect
1	ConsensusGBS 0132-4	7	84.91648	5.07E-10	0.116788321	137	NA	NA	4.97E-07 NA
2	2401-1028	1	55.49319	6.10E-08	0.204379562	137	NA	NA	2.99E-05 NA
3	1493-839	3	173.17375	5.79E-06	0.104418734	137	NA	NA	0.001892074 NA
4	9638-619	1	64.91457	0.00020267	0.233576642	137	NA	NA	0.049704915 NA

Additional file 9. Boxplots of trait BLUEs at each organic location (O1, O2) grouped by the SNP dosages for the most promising SNPs. Traits: canopy height at stem elongation stage (CH), leaf inclination angle (LAN), the width of flag leaf (WFL), plant height at harvest (PH).

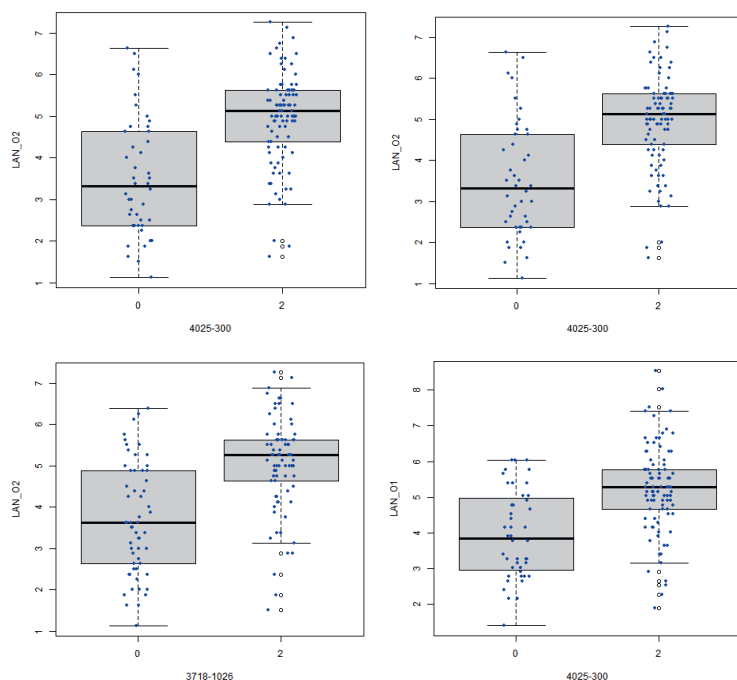
CH

QTL ABC38781-pHv2346-01 dose 0 corresponds to genotype AA, 2 corresponds to genotype GG,



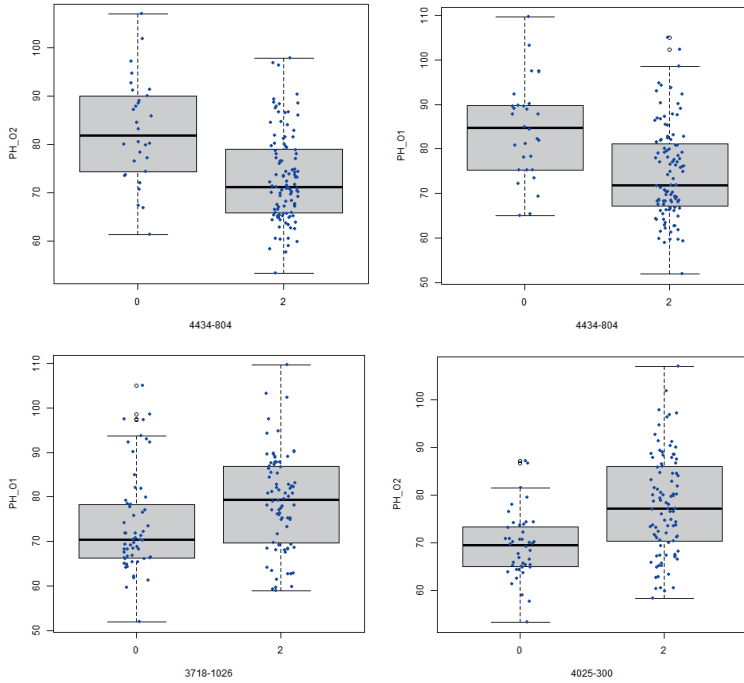
LAN

QTL 4025 300 dose 0 corresponds to genotype TT, 2 corresponds to genotype AA, for QTL 3718-1026 dose 0 corresponds to genotype AA, dose 2 corresponds to genotype CC



PH

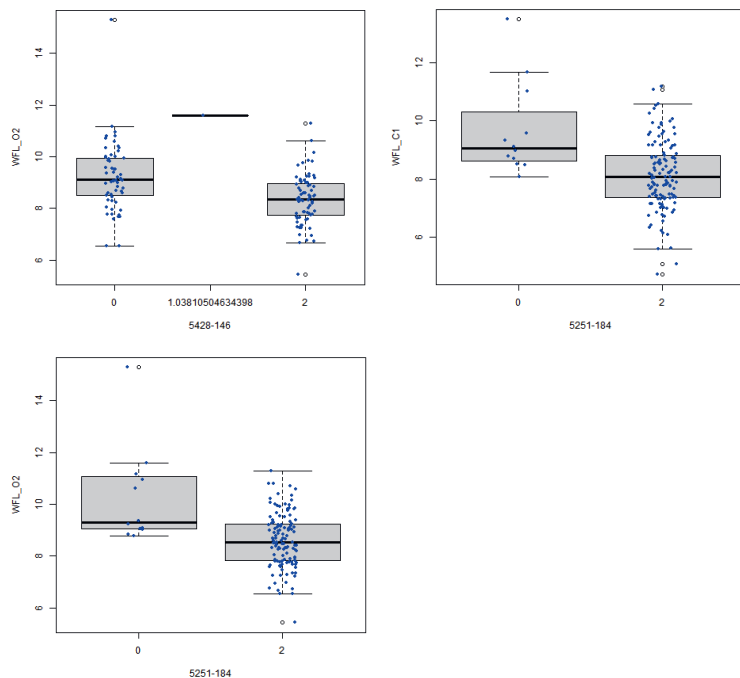
For QTL 4434-804 dose 0 corresponds to genotype AA, dose 2 corresponds to genotype GG, QTL 4025 300 dose 0 corresponds to genotype TT, dose 2 corresponds to genotype AA,



*Genome wide association mapping for grain yield and weed suppressive ability in barley (*Hordeum vulgare* L.) under conventional and organic conditions*

WFL

For QTL 5428-146 dose 0 corresponds to genotype CC, dose 2 corresponds to genotype AA, for QTL 5251-184 dose 0 corresponds to genotype GG, dose 2 corresponds to genotype AA



Chapter 7

General Discussion

The objective of this thesis was to optimise a spring barley breeding strategy for organic farming including all the main steps of the breeding process: choice of crossing parents, selection criteria and selection environment. In **Chapter 1** the research questions are described and justified i) How do varieties differ in yield, yield stability and weed suppressive ability under conventional and organic conditions? ii) Is selection for barley varieties adapted to organic farming systems more effective under organic conditions than under conventional conditions? iii) How does direct selection under organic and indirect selection under conventional conditions of barley genotypes affect traits relevant for organic farming systems? and iv) Can specific markers associated with yield and traits contributing to weed suppressive ability for organic farming be identified in an association mapping population?

The main results of the research chapters are highlighted in Section 7.1 providing answers to the original research questions. In the Sections 7.2. – 7.5, the results of the different chapters will be discussed in a broader context and recommendations for optimisation of barley breeding for organic farming will be presented. A final outlook is discussed in Section 7.6.

7.1. Overview of the main findings

In **Chapter 2**, I analysed how ten barley varieties differed in yield and yield stability under two conventional and two organic management conditions. The analysis showed that generally, the best performing varieties under conventional conditions also performed the best - with high and stable yields - under organic conditions, but there were also exceptions. For example, the short straw variety 'Annabell', bred for high-input farming showed a notable decrease in the yield rank position in organic farming in comparison to its rank in conventional farming. When comparing the ranks across locations, the rank correlation for yield was higher for one of the organic sites and the conventional site with medium input level than between either of the organic sites with the high-input conventional site or between the two organic sites. Therefore, the conventional medium-input site could better predicted variety differences in grain yield under organic conditions than the conventional high-input environment. Heritabilities for yield and yield components (such as number of tillers, thousand-grain weight and number of

grains per tiller) were lower under organic than under conventional conditions, especially at the lowest-yielding organic site, which was a farmer's field. These results lead to the conclusion that selection of high-yielding and stable barley genotypes for organic farming may take place under well managed organic and also under conventional conditions with medium input levels. Differences in management practices between the organic institute and farmer's fields caused changes in the yield level of the varieties. The genotypic rank correlation for grain yield was low between organic sites, indicating that a final testing should be conducted under several organic conditions to confirm the suitability of the selected varieties for cultivation on other organic farms.

In **Chapter 3**, we analysed the same set of varieties studied in Chapter 2 to assess how different morphological and physiological traits contributing to weed suppression performed in organic and conventional farming sites. How do those traits affect yield, and in which growing conditions could the selection of genotypes for organic farming be done? In this study, the genotypes with an erectophile plant growth habit at tillering developed faster, produced taller plants at the beginning of stem elongation and provided good canopy cover at the beginning of the growth period under organic conditions, compared with the genotypes with a planophile plant growth habit. Therefore, in the selection of genotypes for organic farming the planophile growth habit at tillering should be combined with early vigour. The values of the growth habit at tillering, the early vigour, the plant height at the beginning of stem elongation, the length and width of the flag leaf, and the plant height at harvest were more closely correlated between organic and conventional sites than between the two organic sites. Therefore, the selection for these traits may occur under conventional conditions. Most morphological traits contributing to weed suppressive ability positively correlated to grain yield only under poor organic conditions, while a negative correlation was observed between canopy and leaf parameters and yield under more optimally managed organic conditions. Therefore, in breeding for organic farming, the potential trade-off between yield and weed suppressive ability under better managed organic conditions should be considered and further investigated.

In **Chapter 4**, we analysed whether selection over several generations, starting from two different F3 populations derived from an organic breeding

programme, under conventional conditions (indirect selection) can be as effective as selection under organic conditions (direct selection) to develop varieties suitable for organic farming. An organic ideotype score (OIS) comprising various morphological and physiological traits of barley was developed as a selection tool to compare the results between direct and indirect selection. The results indicated that whether direct or indirect selection is more effective depends on the properties of the parents that are crossed. When selecting in the population with morphologically more contrasting parents (short and tall, medium to high yield potential, but with stable yield under different growing conditions) selection under the stressful weedy organic growing conditions led to genotypes with outstanding weed suppression ability but low yield potential in either of the organic sites. The well-managed organic conditions had an advantage over conventional sites to select for a better balance between weed suppressive ability and yield. For the population with less contrasting parents (medium to tall, with stable but low to medium productivity), no large differences were observed between selection sites in obtaining genotypes suitable for organic farming.

The conclusion from this experiment is that direct and indirect selection in early breeding stages are equally suitable for developing varieties for organic farming, if the following conditions are taken into account: 1) yield and also characteristics of importance for organic farming such as the traits contributing to weed suppressive ability, length of crop cycle, disease resistance and grain quality parameters have to be considered; 2) selection is not performed under too stressful conditions, but rather under well managed organic conditions, or conventional medium input conditions, and 3) additional testing of breeding material at later stages of the breeding programme have to be conducted under various organic farming conditions.

In **Chapter 5**, I analysed in more detail and with a large set of genotypes (n=134) how direct or indirect selection affected grain yield (GY) and traits contributing to weed suppressive ability (WSA) under organic conditions. The results showed that direct selection was the most effective in identifying highly suitable genotypes for organic farming at organically managed sites. The selected genotypes performed rather well for both grain yield and weed suppressive ability. The selection procedure where mild selection for WSA was followed by strict selection for GY gave the best result, rather than the

other way around. In principle, selection of genotypes by combining high GY and high WSA is possible under conventional conditions. However, the genotypes that performed well for both traits were selected less frequently, and the number of selected genotypes was smaller than by direct selection under organic conditions. Selection based on organic ideotype scores (OIS), which in addition to GY and WSA comprise also other essential traits for organic farming, resulted in the highest number of well-performing genotypes for grain yield and weed suppressive ability in both organically and in conventionally managed selection fields and could be applied as an alternative to both previously mentioned selection procedures in breeding for organic farming.

In **Chapter 6**, a genome-wide association study (GWAS) was performed to identify quantitative trait loci (QTL) affecting traits relevant for organic farming in spring barley (yield and weed suppressive ability). The QTLs identified in the organic and conventional management systems were compared. Overall, only for four out of the nine traits contributing to weed suppressive ability QTLs were identified: canopy height, leaf inclination angle, width of the flag leaf and plant height at harvest. In total, 35 QTLs were identified, of which 10 were significantly associated with the traits only under organic farming and 18 only under conventional farming conditions, seven were in common between both systems. This means that most of the detected QTLs (80%) were management specific. Notwithstanding highly variable environmental conditions in organic farming, this study helped identify several loci relevant for organic barley breeding.

7.2. Selection material: the value of old and modern (conventionally bred) varieties for organic agriculture

The key factor for the successful development of any breeding programme lies in identifying existing genetic variation for desired traits, resulting in the opportunity to develop new varieties in which such traits are combined. An evaluation of genetic resources by gathering phenological and morphological data and information about yield, quality parameters and disease resistance, provides a basis for the future development of breeding programmes (Newton et al., 2011).

As organic farming systems refrain from chemical-synthetic inputs, they lack the possibility to compensate for limiting environmental conditions such as

high inputs of mineral fertiliser, herbicides, fungicides and pesticides. Thus, in organic farming, the growing conditions vary more over the years in aspects such as soil nutrient status and weed density, disease and pest pressure, between and within farms (Wolfe et al., 2008; Lammerts van Bueren et al., 2011; Ceccarelli & Grando, 2022). Therefore, varieties are required that can adapt to these variable growing conditions while maintaining productivity. Therefore, yield stability in different environments, rather than yield *per se*, is one of the highest priorities in variety choice for organic farming (Østergård, 2002). In that context, there is much discussion in the organic sector whether modern varieties are adapted to organic growing conditions and might have lost important traits such as deep rooting (De Melo, 2003, for onion; and Newton et al., 2011 for cereals), or lack yield stability (Migliorini et al., 2016), and adaptability to stressful conditions (Dwivedi et al., 2016). Other authors such as El Bassam (1997) stress the benefits of including landraces and old cultivars in special breeding programmes of crop varieties for low nutrient conditions. Bellucci et al. (2013) highlighted the landraces as a very heterogeneous and useful source of germplasm for sustainable agriculture in the context of future climate change.

By analysing the yield stability of old versus modern varieties in this thesis, I found that old varieties showed high stability, but had in all cases low ranking positions for grain yield under both organic and conventional conditions (see Chapter 2). On the other hand, some of the modern varieties had high and stable yields across various conventional and organic conditions, and could thus be considered well suited for organic farming. However, these findings of Chapter 2 were obtained with a limited set of ten varieties.

In Chapters 5 and 6, by evaluating a wide range (153) of barley accessions derived over a period of time covering more than one century (approx. 1900 till 2012), I found an increase in barley productivity and adaptation, and also changes in barley plant architecture (Table 7.1). This considerable increase in barley productivity was observed since the middle of the last century at the beginning of the so-called Green Revolution, when the traditional agricultural methods were replaced by modern approaches with high input of agro-chemical resources and technologies. As a higher responsiveness to favourable growing conditions was needed without increasing risk of lodging, this initiated the development of new high-yielding,

disease-resistant and short-straw varieties. Brancourt-Hulmel et al. (2003) studied the changes in traits of winter wheat varieties cultivated in France during the second half of the 20th century and found similar trends in plant architecture such as: decrease in plant height, higher lodging resistance, increase in harvest index values, higher and more stable yields. The main effort of conventional barley-related breeding programmes has been focused on the use of semi-dwarf genes such as *sdw1* and the *sdw1/denso* gene to reduce lodging under high levels of nitrogen inputs and to improve the harvest index (Chen and Yan, 2015). Plants having the semi-dwarf genes are characterised by a prostrate juvenile growth habit type, with narrow, short and erect leaves, short culms, later heading, as well as by late maturity and increased yield (Kuczyńska et al., 2013; 2014).

Similar observations were made by Herrera et al. (2020), but they also noted that in wheat the progress realised for conventional production systems by breeding was not matched under organic management. On the contrary, in their study, a slightly negative trend was observed in grain yield for wheat under organic management. The differences in change in yield between both management systems may indicate on the one hand substantial weaknesses in organic management such as nutrient supply and weed control, that need to be improved. On the other hand, it also points to the challenges of breeding for other types of varieties to improve productivity and stability under organic and low-input conditions.

High productivity in organic cereal production systems partly depends on high weed suppressive ability and high nutrient uptake efficiency since herbicides and synthetic fertilizers are precluded (Löschnerberger et al., 2008; Osman et al., 2016). Weed competitiveness of cereal varieties is highly beneficial to suppress undesirable weeds and volunteer plants (Wolfe et al., 2008). Therefore, the cereal varieties with morphological and physiological traits such as early plant vigour, large leaves, and tall plants at early growing stages and at harvest are essential genetic resources in breeding programmes to improve weed suppressive ability (Andrew et al., 2015; Kissing Kucek et al., 2021). Despite their low grain yield potential, the old varieties have a set of morphological traits relevant for good weed suppression ability: relatively high early vigour, tall plants, and tall and declining leaves (see Chapters 2, 3 and 4). Some modern varieties also had good yield stability under organic

conditions. However, other modern varieties showed a large decrease in yield and a lower rank position under organic conditions compared to their performance under conventional conditions (see Chapters 2 and 4). Some modern short-straw varieties bred for high-input farming showed weed tolerance and could yield relatively well in the heavily weed-infested farmer's field (O2). However, such weed-tolerant varieties are not a good solution for long-term management of weed population dynamics under organic conditions, because they do not reduce weed growth and weed seed production thus creating the risk of establishing a large seedbank (Cosser et al., 1997).

This thesis shows that varieties developed after the 1980s were more adaptable and proved to be suitable for diverse growing conditions in comparison to varieties grown at the beginning of the last century (Table 7.1). My finding is consistent with that of Carr et al. (2006), who, in their experiment with the varieties representing different development eras, concluded that modern spring wheat varieties are adapted to organic environments.

Cereal breeding programmes in Latvia have never been strictly focused on selection for high-input farming and, with some exceptions, are still working with genetic material with medium (70 – 85 cm) to tall (>85 cm) plants (descriptors for spring barley; www.silava.lv). Some of the genotypes developed for high-input farming were responsive to favourable conditions in organic management. These genotypes also stood out for some morphological traits such as: early vigour, canopy height, and plant height at maturity, which are considered to be contributing to weed suppressive ability. Latvian farmers have recognised some of the varieties released from conventional breeding programmes in the 1980s as a useful option for growing in organic farming.

For example, the variety Abava, developed in Stende's plant breeding station and registered in Latvia and also in Ukraine in 1980 (Holms, 1990), was appreciated for its high productivity, wide adaptation, good tillering ability, resistance to lodging and diseases, as well as for its high quality for malt production. Abava is still included in the Latvian Catalogue of Plant Varieties with a remark "suitable for growing in organic farming". The variety 'Rubiola' selected within a conventional breeding programme of my breeding institute and tested as an advanced line in organic trials is currently one of the most demanded varieties among organic farmers in Latvia.

Based on the results of this thesis, I can conclude that, in principle, modern conventional barley breeding can provide suitable material for organic breeding programmes aiming at combining high yield and high weed suppressive ability. Although conventional plant breeding has put much emphasis on yield improvement and remarkably changed the plant types by reducing plant length, the genetic variation for important traits for organic farming is still present in currently available modern breeding material and can be used in breeding programmes for organic farming.

7.3 Selection criteria

Selection in general aims for a genotype with a certain ideotype, which implies a set of traits that will meet the requirements of growers and processors. In organic and conventional breeding programmes, some breeding goals are similar such as high productivity in combination with high resource use efficiency and resistance to pests and diseases as well as high-end product quality. Next to such traits in common, grain yield stability, weed suppressive ability, adaptation to different growing conditions, or specific adaptation to certain conditions are highly desired for organic farming. Thus, the inclusion of these traits in the selection process allows the selection of genotypes suitable for organic farming.

Table 7.1. Adaptability, yield, morphological and phenological traits of 153 spring barley genotypes, according to the time of their release (arranged by decades) in Latvia. For each group of genotypes, data are averaged over three years (2010-2012) and over two sites of two management systems: O (organic) and C (conventional). Means are calculated over these 12 site \times year combinations.

Period of time		1900- 1929	1930- 1939	1940- 1969 ¹	1970- 1979 ²	1980- 1989	1990- 1999	2000- 2009	2010- 2012
No of genotypes		3	2	7	6	12	40	73	10
Adaptability ³	<i>b</i>	0.85	0.77	0.93	0.97	1.01	0.99	1.01	1.10
Grain yield (t/ha ⁻¹)	Mean	2.66	2.55	2.82	3.10	3.23	3.22	3.16	3.45
	O	1.84	1.60	2.10	2.37	2.31	2.32	2.27	2.42
	C	3.48	3.50	3.54	3.83	4.15	4.12	4.04	4.47
Plant growth habit ⁴ (scores from 1- erect to 9- prostrate)	Mean	6	4	4	5	5	5	5	5
	Min	5	4	3	4	5	2	2	4
	Max	7	4	6	8	8	8	9	8
Canopy height at stem elongation stage (cm)	Mean	22	27	24	21	23	21	22	21
	Min	18	27	21	11	14	10	11	13
	Max	26	27	29	25	28	28	30	25
Leaf angle, scores (from 1-erect to 9- declined)	Mean	6	6	6	5	6	4	4	4
	Min	6	6	5	2	2	1	1	3
	Max	6	6	6	6	8	7	7	5
Length of flag leaf (cm) ⁴	Mean	12	14	12	11	11	11	11	11
	Min	11	13	11	11	10	8	8	9
	Max	13	14	14	11	13	13	14	13
Width of flag leaf (cm)	Mean	8	12	9	8	8	8	8	7
	Min	7	12	7	7	7	6	7	6
	Max	9	13	14	9	13	11	14	9
Length of crop cycle (days from sowing to full ripening)	Mean	99	91	96	99	97	97	97	98
	Min	96	90	91	97	94	93	92	95
	Max	102	93	99	99	100	102	102	102
Plant height at maturity (cm)	Mean	102	89	84	78	78	74	78	74
	Min	96	88	71	68	69	62	55	65
	Max	108	90	98	93	86	88	101	87

Weed suppressive ability (WSA) ⁵	Mean	4.1	8.0	3.0	-0.5	1.0	-1.3	0.0	-2.1
	Min	1.8	7.2	-1.2	-5.4	-4.1	-10.5	-8.2	-7.9
	Max	6.1	8.8	7.4	3.3	7.9	3.9	9.5	2.9

¹ The period covers 29 years, as only a small number of varieties was released in that period.

² Grey shading highlights the start of the Green Revolution in cereal breeding.

³ The slope of the regression line (*b*), calculated according to Finlay and Wilkinson (1963). Genotypes with a slope >1 are more responsive to favourable conditions, genotypes with a high yield across environments and a slope close to 1 would be stable and have wide adaptation.

⁴ For more detailed information on trait evaluation, see the Methodology section of Chapter 3.

⁵ For the calculation of WSA, see Chapter 4; for WSA the following traits were included: canopy height, leaf angle, length and width of the flag leaf, plant height at maturity.

7.3.1. Organic ideotype scores

In this thesis (Chapters 4 and 5), I developed and applied the concept of organic ideotype scores (OIS). OIS was created with the aim to compare the suitability of genotypes for organic farming including several traits of importance. A similar index for the evaluation of variety suitability for organic pea production was used by Annicchiarico & Filippi (2007). The index they applied was equal to the average variety rank based on data previously computed for each trait such as yield, plant height at the onset of flowering, the tolerance to disease, earliness, and winter hardness. In another study, Hansen et al. (2008) developed an index for spring barley varieties based on four growth traits, which helped predict weed suppressive ability under weed-free conditions.

In the OIS development, I primarily took into account the characteristics of spring barley varieties required and prioritised by Latvian organic barley growers (see Chapter 1). Most traits are recognised by other authors addressing traits for organic cereal production systems (Wolfe et al., 2008; Hoad et al., 2008; Osman et al., 2016; Mahajan et al., 2020). In this study, OIS includes different morphological and physiological traits. I grouped various traits into five composite traits such as yield (Chapter 2) and weed suppressive ability (Chapters 4 and 5), disease resistance, length of the crop cycle and grain quality. Each OIS component is composed by underlying traits that can interact with and compensate for each other. Therefore, a weight was given to each trait. Each weight for a trait in the OIS was assigned based on expert views of involved breeders and farmers, depending on the relative importance of such traits in contributing to suitability for organic farming. The two main components, GY and WSA, received the highest weights: 40% and 42%,

respectively. The traits for WSA were chosen in accordance with the expert views of our breeders and farmers during field days. The WSA component combines various traits considered important for adaptation to organic farming systems, such as canopy height at stem elongation, canopy cover, the length and width of the flag leaf, and plant height at maturity. Within the WSA structure the highest weights (20% and 10%, respectively) were assigned to the canopy height at stem elongation stage and canopy cover. In OIS, the remaining 18% comprised a combination of other traits important for organic agriculture, such as grain quality aspects (e.g., volume weight) and length of the crop cycle, and resistance against diseases such as powdery mildew (*Blumeria graminis*) and netblotch (*Pyrenophora teres*); these were included in OIS with lower weights.

The results (see Chapter 4) showed that, depending on genotypes and selection environment, selection could lead to genotypes with differently balanced relationships between WSA and GY under organic conditions. I observed that genotypes with high OIS could have a high value for one component, for example grain yield, but low for another (e.g., WSA), depending on the selection site. In the extended experiment with the set of 134 genotypes (Chapter 5), direct and indirect selection on the basis of OIS led to similar results. In OIS as a multi-trait selection criterion which incorporates GY and WSA, high grain yields may compensate for low WSA, and therefore can result in genotypes that may not be suitable for organic farming in terms of WSA. The opposite situation may be possible as well, when selected genotypes have high WSA but low GY. Overlooking my results, in order to avoid too much emphasis on WSA (42% in OIS) and a possible trade-off with GY (40% in OIS), the weight in OIS given to the traits should be carefully evaluated.

Although the emphasis in my thesis was very much on how to improve the combination of GY and WSA, the benefits of also applying OIS was that this multi-trait index allowed comparing how large OIS of the selected genotypes was for a broader set of traits of importance for organic farming. However, OIS requires many observations and calculations, and attention to changes in other characteristics included in OIS as well as their interaction with each other and with the two main components GY and WSA. Depending on the

wish of food producers, other traits such as protein and β -glucan content could be included in an OIS as well.

7.3.2. Selection criteria: combining grain yield with weed suppressive ability

In this paragraph, I will further discuss the potential trade-off between yield and weed suppressive ability. For most organic farmers, yield potential and weed suppressive ability will remain the primary criteria for the choice of cereal cultivars. Therefore, in the selection for organic farming, breeders are aiming at a greater weed suppressive ability without reducing the yield potential and vice versa. Growing high-yielding varieties with high weed suppressive ability is a very relevant option for long-term weed management strategies, reducing the need for costly labour for mechanical weed control and improving the sustainability of cropping systems (Andrew et al., 2015).

In the experiment with 10 barley varieties, the traits contributing to WSA positively correlated with GY only at the organic site with poor weed management (Chapter 3). Further studies have also shown that the better the practices for site management and the more favourable the growing conditions, the weaker (or even negative) the correlation between the traits contributing to WSA and GY (Chapters 4 and 5). These findings indicate that selecting genotypes for a combination of high grain yield and good weed suppressive ability is possible and even better in a weed-free environment which is consistent with other findings (Huel and Hucl, 1996; Andrew et al., 2015; Mahajan et al., 2020). Andrew et al. (2015) concluded that too much emphasis on WSA could lead to lower GY, especially in more productive environments where weed pressure is low. In the selection experiment (Chapter 4), the rank correlation between genotypes for GY and WSA indicated that generally, ranks of the genotypes for GY and WSA tended to be positively associated, or no correlation was observed under organic conditions. However, in my experiments it depended on the cross combination and site of testing. The ranking of genotypes for GY and WSA for the cross, where an old variety was used as one of the parents, showed that GY and WSA tended to be positively associated in well managed organic conditions. However, some lines mainly derived from the cross where the modern high-yielding parent was involved, ranked high for one criterion but low for the

other. In spite of the negative correlation, some genotypes ranked high for both GY and WSA under organic conditions. This finding reveals that it is possible to select genotypes that combine high GY and high WSA. Other studies on barley also indicated that it is possible to reduce the risk of a trade-off between weed suppression ability and yield potential by careful selection (Christensen, 1995; Bertholdsson, 2011).

To combine high GY with high WSA in genotypes I make the following recommendations: Attention should be paid to highly heritable traits under organic conditions such as canopy height at stem elongation, width of the flag leaf, leaf inclination angle, and plant height. Although canopy density or canopy cover is less heritable, this characteristic should not be ignored and added to the selection criteria.

When performing selection for organic farming within a conventional breeding programme, it should be recommended to focus on the traits which correlate highly between conventional and organic systems and for which the heritabilities are high, to avoid genotype \times environment interaction. Examples of such highly heritable traits from our study are canopy height at stem elongation, width of the flag leaf, plant height, and time to heading.

In this study, I paid attention to weed suppression ability as a composite trait, but did not analyse which of the individual component traits contributing to WSA are more responsible for yield reduction than others. Therefore, in future research attention needs to be given to the underlying traits that confer greater competitive ability without incurring a yield reduction.

7.3.3. Selection procedures

In Chapter 5, next to OIS, I applied different selection pressures for combining grain yield and weed suppression ability. I found that under organic conditions the procedure where mild selection for weed suppressive ability was followed by strong selection for yield, allowed to select for highly suitable genotypes in which both grain yield and weed suppressive ability scored high when tested under organic conditions. Under conventional selection conditions, aiming at genotypes with high grain yield and weed suppressive ability for organic farming, the selection should be performed for grain yield and traits contributing to weed suppressive ability. However, the selection under conventional conditions might be less effective than direct selection under

organic conditions. It could lead to a smaller number of genotypes that perform high in yield and weed suppressive ability than direct selection under organic conditions.

7.4. Selection environments

7.4.1. Effectiveness of direct and indirect selection in breeding for organic farming

Between scientists, there is no consensus on the environment where breeding for organic farming could most efficiently be performed. Some studies on winter and spring wheat have led to the conclusion that direct selection under organic conditions is the most effective approach for breeding for organic farming (Atlin and Frey, 1990; Brancourt-Hulmel et al., 2005; Murphy et al., 2007; Reid et al., 2009, 2011; Baenziger et al., 2011; Kirk et al., 2012). On the other hand, nowadays more and more conventional breeding companies aim to serve both the conventional and organic market and are looking for different breeding models for organic farming. As two complete, separate breeding programmes within one company is considered not economically feasible, the option is to integrate breeding for organic into a conventional breeding programme, as Löschenberger et al. (2008) described based on their practical experience with winter wheat in Austria. For breeding for organic and low-input, conventional farming, they suggest that one can start in early generations with a combined breeding programme for conventional and organic varieties both under conventional growing conditions with focus on highly heritable traits important for both farming systems. Then in later generations (from F5/F6 onwards), the programme could be split and continued under conventional and organic conditions separately with focus on selection for the less heritable traits. Similar recommendations were given by other authors such as Baenziger et al. (2011) and Osman et al. (2016), who proposed a blended conventional and organic wheat breeding programme.

The results of studies on comparison of the selection results on winter and spring wheat under conventional versus organic conditions were mainly based on breeding lines and breeding populations derived from conventional breeding programmes intended for the conventional management system (Murphy et al., 2007; Reid et al., 2009, 2011; Baenziger et al., 2011; Gevrek & Atasoy, 2012; Kirk et al., 2012). Kronberga et al. (2013), in their selection

experiment under organic conditions, used winter triticale breeding lines from a conventional breeding programme.

The results of this selection experiment showed that with respect to grain yield lines selected in the better managed organic conditions (O1) and in both high- and medium-input conventional conditions (C1 and C2) performed equally well when tested in the O1 test site. When also taking into account other traits such as weed suppressive ability, only lines derived from O1 and conventional medium-input C1 showed equal results. Lines from the conventional high input site C2 showed high yields but low weed suppressive ability, whereas lines selected in the poorly managed O2 site led to high weed suppressive ability but low yields in any of the organic sites. So, the conclusion of Chapter 4 (Kokare et al., 2017) was that indirect selection under conventional medium input conditions can potentially provide lines that combine high grain yield and weed suppressive ability for organic growing conditions.

However, with respect to the most efficient selection environment, Chapter 5 with the selection exercise based on 134 genotypes taught me this: a significant increase in grain yield and weed suppressive ability for the genotypes was more frequently achieved when the selection was performed under organic conditions (especially in the well managed organic site rather than in the poorly managed organic site) than under conventional conditions. This suggests that the direct selection for genotypes combining high grain yield and weed suppressive ability is more efficient than indirect selection in providing more frequently suitable selections.

7.4.2. Role of cross combinations in the effectiveness of direct and indirect selection

The set-up of the selection experiment of Chapter 4 with two contrasting segregating populations as starting point for the selection process also provided the opportunity to analyse more in-depth the role of the properties of parents that are crossed. The selection results indicated that whether direct or indirect selection is more effective depends on the properties of the parents that are crossed. It demonstrated genotypic differences between the selected lines of each cross combination for weed competitiveness. The lines derived from the more weed competitive parents Primus and Idumeja showed superior

WSA in the organic testing sites. The optimally managed organic selection site (S O1) and both conventional selection sites (S C1 and S C2), allowed to select suitable lines for the organic, weedy, low-input test conditions (T O2) with positive scores for both traits GY and WSA (Fig. 7.1, red ovals). However, the selection using this cross between Primus and Idumeja at the organic low-input selection site (S O2) with a high abundance of weeds did not meet the expectations in terms of high productivity under either organic test conditions (T O1 and T O2, Fig. 7.1).

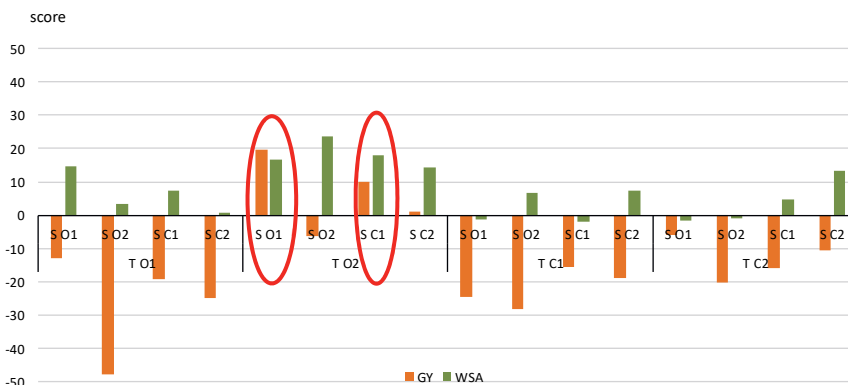


Figure 7.1 Scores for grain yield (GY) and weed suppressive ability (WSA) for the lines of the cross between Primus and Idumeja, at two organic (T O1 and T O2) and two conventional *testing* sites (T C1 and T C2), depending on selections at the *selection* sites (S O1, S O2, S C1, S C2) Scores for GY and WSA are expressed as the difference between the average value of GY and WSA of 5 selected genotypes at each of the selection sites and the average value of the total set of 40 genotypes (which is equal to 0) tested at each of the respective sites. The red ovals mark the selection sites at which selected lines combined high GY and high WSA.

The environment with a high occurrence of weeds allowed to select for a high level of WSA, which was not achieved at any other selection site (Fig. 7.1). At the farm site selected lines from the cross Anni/Dziugiai, had a relatively high WSA and an acceptable (medium) yield when tested under optimally managed organic conditions (Fig. 7.2). Although selection resulted in a superior WSA for the lines of cross Anni/Dziugiai in a farmer's field, the WSA of these lines was lower compared to the lines of the cross Primus/Idumeja in the farmer's field. Considering the possible high weed pressure in the organic site O2, the lines derived from the cross between Primus/Idumeja could be a better approach. The lines derived from the cross between Anni and Dziugiai

exhibited superior GY than the lines from cross Primus/Idumeja, especially if selected in more productive environments (see Fig. 7.2), indicating the ability to tolerate weeds in the offspring of these two parents. The weedy, low-input organic farm site seems to offer a good opportunity to select for specific adaptation ability to less favourable growing conditions with a high abundance of weeds.

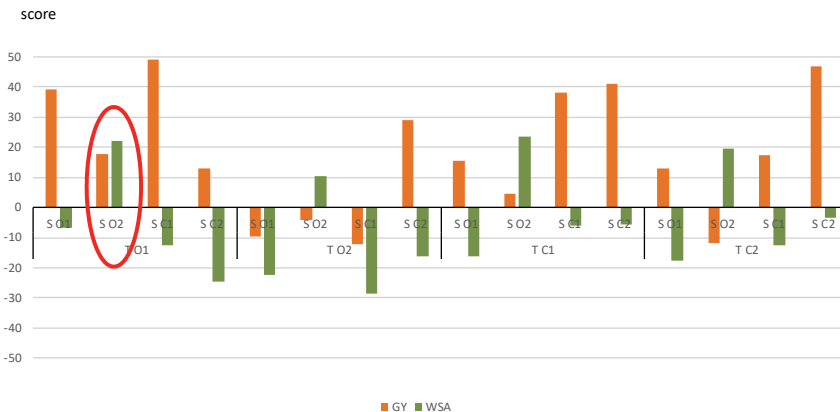


Figure 7.2. Grain yield (GY) and weed suppressive ability (WSA) for the lines of the cross between Anni and Dziugiai, at two organic (T O1 and T O2) and two conventional *testing* sites (T C1 and T C2), depending on selection at four *selection* sites (S O1, S O2, S C1, S C2). Scores for GY and WSA are expressed as the difference between of the average value of GY and WSA of 5 selected genotypes at each of the selection sites and the average value of the total set of 40 genotypes (which is equal to 0) tested at each of the respective sites. The red oval marks the selection site at which selected lines combined high GY and high WSA.

7.4.3. Concluding remarks

The outcomes from the analysis above and from Chapters 4 and 5 confirm that direct selection under well managed organic growing conditions is most effective for organic farming (see also Murphy et al., 2007; Reid et al., 2009, 2011; Baenziger et al., 2011; Kirk et al., 2012; Gevrek & Atasoy, 2012; Kronberga et al., 2013). However, the results showed a low genetic correlation between both organic sites. Therefore, we think that selection results can be less predictable for poorly managed, weedy organic conditions because of variable soil fertility and management conditions. Therefore, testing in

specific organic growing conditions will be needed, as several authors have also argued (Przystalski et al. 2008; Osman et al., 2016).

This thesis also shows that it is possible to select good lines that combine high yield and weed competitiveness through indirect selection (rather under medium-input than under high-input conventional conditions), but less frequently than through direct selection under organic conditions.

What this thesis specifically contributes to the literature is that the choice of the initial breeding material is highly depending on where the breeding programme for organic farming is going to be carried out. When performing selection under organic conditions, more diverse breeding materials can be used. To carry out the selection under conventional (medium-input) conditions for genotypes combining high GY and WSA, the initial breeding material (parents) must be strictly selected for their valuable morphological features that contribute to weed suppression ability and yield stability.

7.5 Application of QTLs in the selection of genotypes

The phenotypic selection and evaluation can be a very time-consuming process. By using DNA markers, this work can be accelerated, and breeding efficiency can be increased. Each individual plant is influenced by its genetic potential as well as its growing environment, which affects the expression of various agronomic and/or morphological traits.

In barley GWAS studies, it was found that genes linked to plant height were located on different chromosomes, and their expression may depend on the environment (e.g., water deficit) (Jabbari et al., 2018; Bai et al., 2021). Locatelli et al. (2013) pointed out that different compensatory mechanisms of complex traits such as grain yield might operate in high- and low-yielding environments. He found some unique QTLs only in low-yielding environments. Asif et al. (2015) and Zou et al. (2017), in studies with spring wheat under organic and conventional conditions, reported on specific QTLs detected in each management system. The results of the barley GWAS study reported in this thesis showed that most QTLs were detected either in organic or conventional management systems and only some in both (Chapter 6).

In this paragraph, I want to assess to what extent the QTLs specifically found under organic growing conditions are associated with component traits of WSA and whether it would be possible to use them in the further selection of genotypes. To gain insight in the presence of beneficial (+) or adverse (-)

alleles of organic-specific QTLs for the traits contributing to weed suppressive ability, I analysed the ten highest yielding genotypes and the ten genotypes with the highest weed suppressive ability scores in the organic and conventional farming system (Tables 7.2 and 7.3).

The results showed that more frequently beneficial (+) alleles of three QTLs: ABC38781-pHv2346-01 (A), 3718-1026 (C), 4025-300 (A) were found to be associated with the traits such as canopy height at stem elongation, leaf inclination angle and plant height at maturity in the organic farming system, for the most productive genotypes in the organic farming system rather than for the most productive accessions in the conventional system. For the genotypes with the highest weed suppressive ability (Table 7.3.) the frequency of traits positive alleles was similar between both farming systems. Slightly fewer positive alleles of two QTLs ABC38781-pHv2346-01 (G) associated with canopy height at stem elongation and QTL 3718-1026 (C) detected for the leaf inclination angle, were present in the top ten genotypes for organic farming. The QTL that was most frequently observed among the top genotypes across both farming systems was QTL 4025-300, which is associated with plant height at harvest.

The highest number of positive alleles of all organic-specific QTLs were found in breeding line PR 5137, variety Klinta, and fo PR 5135. The last was present in the 10 top yielding genotypes in both farming systems. Surprisingly, this line PR 5135 was among the genotypes with the highest weed suppressive ability under conventional conditions, but also among the top 20 genotypes with the highest weed suppressive ability under organic conditions. This would imply that this breeding line could combine genetic factors for high grain yield and good weed suppressive ability. The line PR 5135 was one of the most frequently selected genotypes at the organic site (especially at farmer's O2 field) and at both conventional sites (Chapter 5). PR 5135 had high canopy cover and declining leaves inherited from one of the parents' Abava'. Variety Abava had the highest number of enhancing alleles for the traits contributing to weed suppressive ability, and has already proven to perform with yield stability and high weed suppressive ability in organic practice. It is included in the Latvian Plant Variety Catalogue and is still widely grown in Latvia. Two other parents were high-input variety Annabell and Rubiola, both with high yield potential in environments with different

input levels. Two breeding lines BZ 14-90 (O1-A/DZ-90) and BA 12-63 (O1-P/I-63) selected at organic O1 in the selection experiment (Chapter 4) were among the top genotypes with high weed suppressive ability, but had only three and two (leaf inclination angle, plant height, width of the flag leaf and canopy height) beneficial alleles, respectively (Table 7.3). In the final comparison of the selected lines (Chapter 4) only line BZ 12-63 was included, ranking high (5th place among 20 lines) for grain yield and weed suppressive ability (3rd place among 20 lines) at both organic testing sites. Interestingly, another line B-14-90 was removed from further testing in 2009 due to low yield and tall plants (on average 110 cm), resulting in low lodging resistance among the lines that were grown that year. Line BZ 12-63, unlike line B-14-90, has the canopy height positively influencing allele A of marker ABC38781-pHv2346-01. High canopy is highly valuable for organic farming and it is related to crop ground cover for which no QTL was found. In addition, the line BZ 12-63 was among the most frequently selected genotypes for grain yield and weed suppressive ability in both organic and conventional conditions in the selection experiment (Chapter 4). Canopy height at the stem elongation stage was positively related to early vigour and ensuring high crop ground cover (Chapter 3). In the GWAS study (Chapter 6) the canopy height at stem elongation also positively correlated with crop ground cover. In addition, both these traits had a positive correlation with grain yield. This would imply that the canopy height at stem elongation and crop ground cover are important traits for ensuring high yield under organic conditions. Thus, future research on finding enhancing alleles for canopy height and crop ground cover would be beneficial in varieties for organic farming.

The breeding lines No-79, PR 3282 and the old variety Heils Hanna had the highest number (five) positively influencing alleles of QTLs for the traits contributing to weed suppressive ability found under organic conditions. 'PR 3282' was among the most frequently selected genotypes in organic site O1 (Chapter 5), especially when emphasis in the selection was paid to weed suppressive ability. Compared to 'Hale Hanna', line PR 3282 had a higher grain yield at both organic sites.

Comparing the results obtained in this analysis with the outcome of Chapter 5, it seems that for combining high yield with high weed suppressive ability,

canopy height is an important feature, and therefore the presence of alleles associated with canopy height in the genotypes would be important.

For organic farming, it is important to combine high grain yield with high weed suppressive ability in genotypes. Marker-assisted selection could be an effective tool for the traits for which markers have been identified, making the breeding process more efficient in terms of money and time. For other important traits field evaluation will remain important. Follow-up studies will be needed to validate our results and to develop markers for application in marker-assisted breeding for organic farming.

7.6 Final remarks

The organic sector (and also the organic seed sector) is a growing segment of sustainable agriculture. Also, more and more breeding companies that only focused on conventional agriculture in the past, are interested in serving both organic and conventional markets. These breeders are searching for efficient strategies in combining conventional and organic breeding programmes. This thesis provides in-depth insights into the appropriate selection material, selection criteria and selection environments. This study also explored tools for both field and marker-assisted selection to enhance the breeding of improved organic barley varieties combining yield and weed suppressive ability. In the thesis, I mainly focused on the traits related to grain yield and weed suppressive ability. Future research will be needed to analyse more in-depth which of the individual traits contributing to weed suppressive ability are responsible for potential yield reduction and how the environment (soil characteristics, fertility management, and climate/weather conditions) affects such outcomes. The developed Organic Ideotype Scores index has proven to be an interesting tool for selecting organic varieties with a broad set of characteristics, but can most likely be further optimised. In addition, this thesis will also help serve the Green Deal objectives, e.g. by providing breeding strategies not only for the organic barley sector, but also for the conventional sector searching for ways to reduce the dependency on chemical-synthetic inputs such as herbicides, and making the food system more resilient and more sustainable.

Table 7.2. Top ten yielding accessions in organic and conventional farming systems, their average yield, and the weed suppressive ability (WSA) influencing alleles (positive: +, negative -) at peak markers for QTLs linked to the traits contributing to weed suppressive ability. The genotypes ranged according to grain yield in organic farming system

Accession	Farming system in which accession is among the best ten yielding	Average grain yield t ha ⁻¹				WSA scores ¹		Number of WSA increasin g alleles	ABC38781- PHV2346-01 (G)	Traits contributing to weed suppressive ability				WFL	WFL	5251-184 (G)
		O	C	O	C	CH ²	LAN			LAN, PH	PH					
Jumara	O	3.36	4.98	-1.3	-2.6	3	3	+	+	+	+	+	-	-	-	
Rubiola	O/C ³	3.24	5.00	4.6	-0.9	3	3	·	·	-	+	+	+	+	-	
PR-5105	O	3.10	3.99	3.7	-0.6	2	2	-	-	+	+	+	-	-	-	
PR-5137	O	3.03	4.42	0.1	1.9	5	5	+	+	+	+	+	+	+	-	
PR-3223	O	2.95	4.57	-1.4	-1.4	3	3	+	+	+	+	+	-	-	-	
Sencis	O/C	2.95	5.46	-0.7	0.9	2	2	+	+	+	-	-	-	-	-	
PR-4181	O	2.90	4.65	-1.1	-0.5	3	3	+	+	+	+	+	-	-	-	
PR-4814	O	2.88	4.71	3.0	3.0	2	2	-	-	+	+	+	-	-	-	
Klinta	O	2.87	4.92	3.8	4.3	5	5	+	+	+	+	+	+	+	-	
PR-5135	O/C	2.87	5.01	4.5	5.4	4	4	+	+	+	+	+	-	-	-	
Tocada	C	2.87	5.01	-4.2	-3.8	2	2	+	+	-	-	-	+	+	-	
PR-3518	C	2.75	5.21	-5.0	-5.2	2	2	-	-	-	+	+	-	-	-	
Golf	C	2.46	5.17	2.5	2.3	3	3	-	-	+	+	+	-	+	-	
L-2735	C	2.34	5.01	-1.5	1.6	2	2	+	+	-	+	+	-	-	-	
743-09	C	2.27	5.01	-5.5	-4.8	0	0	-	-	-	-	-	-	-	-	
Leeni	C	2.27	5.17	-7.4	-6.3	0	0	-	-	-	-	-	-	-	-	
1267199-30	C	2.08	5.15	-1.2	1.4	3	3	+	+	+	+	+	-	-	-	
Farming system in which marker was significantly associated with GY																
Frequency of the + allele in set of 17 top yielding accessions																
0.6																
0.6																
O/C																
0																
Frequency of the + allele in set of 10 top yielding accessions in Organic																
0.7																
0.9																
0.9																
0.3																
0.4																
0																
Frequency of the + allele in set of 10 top yielding accessions in Conventional																
0.5																
0.4																
0.6																
0.1																
0.4																
0																

¹ WSA scores are calculated according to the procedure described in Chapter 5, but no weight is given to each trait.

² CH - canopy height; LAN - leaf inclination angle; PH - plant height before harvest; WFL - width of flag leaf;

³ O/C the genotype was among the best ten in the organic and conventional farming system

Table 7.3. The weed suppressive ability (WSA) influencing alleles (positive: +, negative -) at peak markers for QTLs linked to the traits contributing to weed suppressive ability in the top ten accessions with the highest WSA scores in organic (O) and conventional (O) farming systems. The genotypes ranged according to WSA scores in the organic farming system

Accession	Farming system in which accession is among the best ten yielding	WSA scores ¹		Number of increasing alleles	Traits contributing to weed suppressive ability									
		O	C		CH ²	LAN	LAN	PH	PH	WFL	WFL	(A)	(A)	(G)
No-79	O/C ³	8.1	9.7	5	-	+	+	+	+	+	+	4434-804	5428-146	5251-184
AGRA	O/C	7.8	7.6	3	-	+	+	+	-	-	+	-	-	-
Primus	O	7.5	4.5	2	+	-	+	+	-	-	-	-	-	-
Abava	O/C	7.3	7.4	5	+	+	+	+	+	+	+	+	+	-
PR-3282	O	7.2	3.9	5	+	+	+	+	-	-	+	+	+	+
PR-5109	O	6.6	4.5	4	+	+	+	+	-	-	+	+	+	-
BZ14-90	O/C	6.5	9.2	3	-	-	+	+	+	+	+	+	+	-
Dziugiai	O/C	6.2	6.7	3	+	-	+	+	-	-	+	-	+	-
69-Clho11319	O	6.1	3.9	2	+	-	+	+	-	-	-	-	-	-
PR-3605	O	6.1	4.2	2	-	+	+	+	-	-	-	-	-	-
BZ12-63	C	5.0	8.1	2	+	-	+	+	-	-	-	-	-	-
PR-5135	C	4.5	5.4	4	+	+	+	+	-	-	+	-	+	-
Pervonez	C	4.0	5.4	1	+	-	-	-	-	-	-	-	-	-
PR-4368	C	3.7	6.6	3	+	+	+	+	+	+	-	+	-	-
Heils Hanna	C	3.1	6.8	5	+	+	+	+	-	-	+	-	+	+
Farming system in which a marker was significantly associated with the relevant trait														
Frequency of the + allele in the top 15 accessions with high WSA					O/C	O	O	O/C	O	O	O	O	O	O/C
Frequency of the + allele in the top 10 accessions with the highest WSA					0.7	0.6	0.6	0.9	0.3	0.6	0.6	0.2	0.2	0.2
Frequency of the + allele in the set of top 10 accessions with the highest WSA in Organic					0.6	0.5	1.0	0.3	0.3	0.7	0.2	0.2	0.2	0.2
Frequency of the + allele in the set of top 10 accessions with the highest WSA in Conventional					0.7	0.6	0.9	0.4	0.7	0.1	0.1	0.1	0.1	0.1

¹ WSA scores are calculated according to the procedure described in Chapter 5, but no weight is given to each trait.

²CH - canopy height; LAN - leaf inclination angle; PH - plant height before harvest; WFL - width of flag leaf;

³ O/C the genotype was among the best ten in the organic and conventional farming system

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Summary

Organic agriculture combines best practices based on traditional farming methods relying on ecological processes, a high level of biodiversity, and sustains the health of soil, ecosystems and people. The European Commission proposed the Sustainable Europe Investment or Green Deal plan in 2021 to achieve climate neutrality by 2050. The aim for the near future is a 55% reduction of the emission by 2030. Organic farming is considered one of the ways to achieve the Green Deal goals. One of the current EU Green Deal targets is to have at least 25% of the EU's agricultural land under organic farming by 2030. To ensure stable crop yields and competitive product quality under organic farming conditions, the environmental conditions, growing technology, and genetic factors, i.e., crop variety, play an essential role. The studies and breeding activities for organic systems have increased over the last decades. But still, most varieties used in organic farming in Europe are bred for conventional management systems. Conventional varieties are adapted to intensive management with a high nutrient supply. Therefore, these varieties might be less productive and provide less yield stability under organic growing conditions than under conventional farming. In addition, organic farmers give higher priority to additional traits to cope with lower levels of nutrients from organic fertilisation practices, with pests and diseases and weed competition without use of chemical-synthetic inputs. Therefore, **Chapter 1** discusses that a different approach is required in the breeding programmes for organic farming compared to conventional breeding and identifies knowledge gaps on how to optimise plant breeding for organic conditions. The overall objective of the research reported in this thesis is to design a barley breeding approach for organic farming. This research aimed to understand which selection material, selection criteria and selection environments are the most appropriate to select varieties adapted to organic farming conditions. All field experiments in this study were carried out at two organic and two conventional growing sites in Latvia. This thesis contributes to these themes and helps optimize the breeding strategy for organic barley production in Latvia.

Chapter 2 analyses the yield and yield stability of ten contrasting old and modern varieties under two conventional (medium and high input) and two organic conditions (low and medium input) over three years. Old varieties showed high yield stability but had low grain yield under both organic and

conventional conditions. On the other hand, some modern varieties had high and stable yields across various conventional and organic conditions, and could thus be considered well suited for organic farming. Some modern short-straw varieties bred for high-input farming showed weed tolerance and yielded relatively well at the organic low-input site with high weed pressure, maybe due to their early vigour. However, growing such weed-tolerant varieties does not reduce weed growth and may result in the establishment of a seedbank affecting future crop growth. Some other modern varieties showed a large decrease in yield under organic conditions compared to their high yield under conventional conditions. Heritability estimates for yield and yield components (such as number of tillers, kernels per tiller and thousand-grain weight) were lower under organic than under conventional conditions, especially at the lowest-yielding organic site. High rank correlations for grain yield were found among varieties at both organic sites and at the conventional medium-input site, compared to the yield at the conventional high-input site or between the two organic sites. Therefore, the conventional medium-input site may well predict the ranking in grain yield under organic conditions. These results suggest that the selection under the well-managed organic and also under conventional conditions with medium input levels for varieties with high and stable yield could result in rapid genetic gain. Moreover, because the genotypic rank correlation for grain yield was low between the two organic sites, final testing should be conducted under several organic conditions to confirm the suitability of the selected varieties for cultivation on various organic farms.

Chapter 3 analyses how different morphological and physiological characteristics related to the weed suppressive ability of barley varieties are expressed under organic and conventional farming growing conditions. This study was based on the same set of varieties investigated in Chapter 2. The old varieties had a set of morphological traits relevant for good weed suppressive ability: relatively good early vigour, tall plants, and long and declining leaves. However, these old varieties proved to have poor lodging resistance in unfavourable growing conditions. Medium-old varieties and modern, low-input varieties with good adaptability had medium tall straw, resulting in better lodging resistance. They also had a high tillering rate, early vigour in spring, and reached the same early plant height as old, low-input

varieties. Some of the genotypes developed for high-input farming stood out for some morphological traits, such as early vigour, canopy height at stem elongation, and plant height at maturity; such traits are related to weed suppressive ability. This implies that, in principle, modern conventional barley breeding can provide suitable material for organic breeding programmes if weed suppressive ability is included as selection criterion.

The study also revealed that the traits contributing to weed suppressive ability were more closely correlated between organic and conventional sites than between the two organic sites. Therefore, the selection for these traits may occur under conventional conditions. Most morphological traits positively correlated to the yield only under poor organic conditions, while the negative relation to yield was obtained under optimally managed organic conditions. Therefore, this aspect of potential trade-off between grain yield and weed suppressive ability should be considered in breeding for organic farming.

Chapter 4 analyses whether selection across several generations starting from two different F3 populations derived from an organic breeding programme, under conventional conditions ('indirect selection') can be as effective as selection under organic conditions ('direct selection') to develop varieties suitable for organic farming. The characteristics of the parents of the two populations were first investigated in Chapter 2 and Chapter 3, and contrasted e.g., in maturity date, plant length, early vigour, and yield potential. An organic ideotype score (OIS) comprising various morphological and physiological traits of barley was developed as a selection tool to compare the results between direct and indirect selection. The results indicated that whether direct or indirect selection is more effective depends on the parents' properties involved in the cross combination. The selection in the population with morphologically more contrasting parents (the modern short-straw variety with good yield potential and wide adaptation across various growing conditions was crossed with the tall variety with good early vigour) under the stressful weedy organic growing conditions led to genotypes with outstanding weed suppression ability but low yield potential at both organic sites. In contrast, the lines derived from other selection sites showed high yield but low weed suppressive ability. The opposite trend was found for the population with less contrasting parents (medium to tall, with stable but low to medium productivity). Lines stood out with high weed suppressive ability at organic

sites. For this cross no large differences were observed between selection sites in obtaining genotypes suitable for organic farming.

This Chapter 4 reveals that direct and indirect selection in early breeding stages are equally suitable for developing varieties for organic farming, if the following aspects are considered: the choice of the initial breeding material is very important and should depend on where the breeding programme for organic farming is going to be carry out. When performing selection under organic conditions, more diverse breeding materials can be used. To carry out the selection under conventional (medium-input) conditions for genotypes combining high grain yield and weed suppressive ability, the initial breeding material (parents) must be selected for their valuable morphological features that contribute to weed suppression ability and yield stability. Moreover, the selection should not be performed under too stressful conditions, but rather under well-managed organic conditions, or conventional medium-input conditions. Finally, additional testing of breeding material at later stages of the breeding programme has to be conducted under diverse organic farming conditions.

Chapter 5 analyses in more detail how direct or indirect selection affects grain yield and traits contributing to weed suppression under organic conditions. The aim of this study was to find out which selection procedure provides the best combination of both grain yield and weed suppressive ability for genotypes intended for production in organic farming. We used data on the performance of 134 barley genotypes at two organic and two conventional sites across three years. Grain yield and weed suppressive ability were the two main selection criteria, and were applied with different weight for each. The selection at organically managed sites was the most effective in identifying highly suitable genotypes for organic farming with respect to both grain yield and weed suppressive ability. The selection procedure where mild selection for weed suppressive ability was followed by strong selection for grain yield led to the best results under organic conditions. The selection of genotypes for organic farming that combines high grain yield and high weed suppressive ability can also be performed under conventional conditions if both selection criteria grain yield and weed suppressive ability are considered. However, selection under conventional conditions proved to be less effective than direct selection under organic conditions as it led less frequently to the selection of

genotypes that gave both high yield and good weed suppressive ability than direct selection. Moreover, when such genotypes were selected, the numbers were lower.

In **Chapter 6** a genome-wide association study (GWAS) was performed to identify quantitative trait loci (QTL) affecting grain yield and traits contributing to weed suppression ability in spring barley for organic farming. The expression of the genetic factors contributing to the traits related to yield and weed suppressive ability could be different under organic and conventional conditions, and also their importance could differ under the different conditions. An association mapping population consisting of 153 barley varieties and breeding lines relevant for Latvian farming and breeding was phenotyped for nine traits in two conventionally and two organically managed fields, and genotyped with 1536 SNPs. Overall, of the nine traits contributing to weed suppression ability screened, QTLs were identified only for four of them: canopy height, leaf inclination angle, the width of the flag leaf and plant height at harvest, but no QTLs were found for grain yield. In total, 35 QTLs were identified, of which 10 were significantly associated with traits only under organic and 18 only under conventional farming conditions and only 7 were in common between both systems. This means that most of the detected QTLs (80%) were management specific. The QTLs for canopy height (CH), leaf inclination angle (LAN), width of flag leaf (WFL) and plant height at harvest (PH), were mapped in the organic system on chromosomes 3H, 7H, 6H, 2H, respectively. One QTL (on chromosome 3H) was found associated with several traits (CH, LAN and PH) in both systems. Those identified markers may be combined by breeders to develop barley cultivars with improved weed suppressive ability in organic farming. The results also suggests that it is necessary to continue QTL discovery studies for other important traits under organic farming systems.

Chapter 7 assesses the relevance of the main findings of Chapters 1-6 in the context of this study, its objective and research questions to optimise breeding strategies for organic barley varieties, with respect to selection material, selection criteria, and selection environment (direct versus indirect selection). Some additional analyses were included in this chapter to discuss more in-depth i) the value of old and modern varieties to be used as initial breeding material in breeding programmes for organic varieties, ii) the role of the

characteristics of cross combinations in the effectiveness of direct and indirect selection, and iii) to what extent the QTLs specifically found under organic growing conditions are associated with component traits of weed suppressive ability and whether these QTLs could be used in the further selection of genotypes for organic farming systems.

This thesis will help serve the Green Deal objectives for the European Community, e.g., by providing breeding strategies not only for the organic barley sector, but also for the conventional sector searching for ways to reduce the dependency on chemical-synthetic inputs such as herbicides, and making the food system more resilient and more sustainable.

Kopsavilkums

Bioloģiskā lauksaimniecība apvieno labāko praksi, kuras pamatā ir tradicionālās lauksaimniecības metodes, kas balstās uz ekoloģiskiem procesiem, augstu bioloģiskās daudzveidības līmeni un uztur augšnes, ekosistēmu un cilvēku veselību. 2021. gadā Eiropas Komisija ierosināja Ilgtspējīgas Eiropas investīciju jeb Zaļā kursa plānu, lai līdz 2050. gadam panāktu klimata neitralitāti. Mērķis tuvākajā nākotnē līdz 2030. gadam ir samazināt siltumnīcefekta gāzu emisijas par vismaz 55%. Bioloģiskā lauksaimniecība tiek uzskatīta par vienu no zaļā kursa ieviešanas veidiem. Viens no pašreizējiem ES “Zaļā kursa” mērķiem ir panākt, lai līdz 2030. gadam vismaz 25% no ES lauksaimniecības zemes būtu apsaimniekota ar bioloģiskās lauksaimniecības metodēm. Lai nodrošinātu stabilu ražu un konkurētspējīgu produktu kvalitāti bioloģiskajā saimniekošanas sistēmā, būtiska nozīme ir vides apstākļiem, audzēšanas tehnoloģijai un laukaugu šķirnei. Pēdējo desmitgažu laikā pētījumi saistībā ar bioloģisko saimniekošanas sistēmu un selekciju tās vajadzībām ir paplašinājušies. Tomēr lielākā daļa šķirņu, ko izmanto bioloģiskajā sistēmā Eiropā, ir veidotas konvencionālajai saimniekošanas sistēmai, un tās ir piemērotas intensīvai apsaimniekošanai ar augstu barības vielu nodrošinājumu. Tāpēc šīs šķirnes bioloģiskajā saimniekošanas sistēmā var būt mazāk produktīvas un ar zemāku ražas stabilitāti, salīdzinot ar – tieši bioloģiskajā sistēmā veidotām šķirnēm. Turklāt bioloģiskie lauksaimnieki piešķir lielāku prioritāti pazīmēm, kas palīdz veidot pieņemamas ražas apstākļos ar zemāku barības vielu nodrošinājumu, kaitēkļiem, slimībām un nezālēm, neizmantojot minerālmēslus un ķīmiskos augu aizsardzības līdzekļus.

Ievadā tiek aplūkots, ka selekcijas programmās bioloģiskajai saimniekošanas sistēmai ir nepieciešama atšķirīga pieeja, salīdzinot ar selekciju konvencionālajai lauksaimniecībai. Tiek identificētas zināšanu nepilnības par to, kā optimizēt šķirņu selekciju bioloģiskajā saimniekošanas sistēmā. Promocijas darba galvenais mērķis ir izstrādāt miežu selekcijas stratēģiju bioloģiskajai saimniekošanas sistēmai. Pētījuma mērķis bija saprast, kāds selekcijas izejmateriāls, izlases kritēriji un izlases vide ir vispiemērotākā, lai izveidotu bioloģiskajai saimniekošanas sistēmai piemērotas šķirnes.

2. nodaļā ir analizēta desmit dažādas izcelsmes un dažādos laikos izveidotu miežu šķirņu raža un tās stabilitāte divās vietās bioloģiskajā un

konvencionālajā saimniekošanas sistēmā. Bioloģiskajos audzēšanas apstākļos ar un bez nezāles ierobežojošajiem pasākumiem un konvencionālajos apstākļos ar vidēju un augstu mēslojuma daudzumu un herbicīdu pielietošanu. Pagājušā gadsimta sākumā un senāk veidotām šķirnēm bija augsta ražas stabilitāte, taču zems ražas līmenis gan bioloģiskos, gan konvencionālos audzēšanas apstākļos. Pagājušā gadsimta vidū un beigās izveidotām šķirnēm bija augsta un stabila raža atšķirīgos konvencionālajos un bioloģiskajos audzēšanas apstākļos, un tādējādi tās varētu uzskatīt par labi piemērotām bioloģiskajai saimniekošanas sistēmai. Atsevišķas salīdzinoši nesen izveidotas īsstiebrainās miežu šķirnes uzrādīja labu ražu bioloģiskajos audzēšanas apstākļos ar augstu nezāļu īpatsvaru, kas norādīja uz labu nezāļu toleranci. Tomēr šādu pret nezālēm tolerantu šķirņu audzēšana nesamazina nezāļu īpatsvaru augu sekā, tādējādi ietekmējot turpmāko laukaugu augšanu un attīstību un galarezultātā ražu. Citām īstiebrainajām miežu šķirnēm bioloģiskajos audzēšanas apstākļos bija liels ražas samazinājums, salīdzinot ar ražu konvencionālajos apstākļos. Ražai un tās komponentiem, piemēram, produktīvo stiebru skaitam, graudu skaitam vārpā un tūkstoš graudu masai iedzimstamība bioloģiskajos apstākļos bija zemāka, salīdzinot ar konvencionālajiem, īpaši bioloģiskajos audzēšanas apstākļos ar lielu nezāļu īpatsvaru. Salīdzinoši mazas ražas rangi izmaiņas tika konstatētas šķirnēm starp abām bioloģiskajām audzēšanas vietām un konvencionālo audzēšanas vietu ar vidēji augstu mēslojuma daudzumu. Šie rezultāti liecina, ka selekcija bioloģiskajos apstākļos un arī konvencionālos audzēšanas apstākļos ar vidēji augstu mēslojuma daudzumu var nodrošināt labus izlases rezultātus. Tā kā šķirņu graudu ražas korelācija starp abām bioloģiskajām audzēšanas vietām bija zema, selekcijas procesa noslēgumā genotipu pārbaude būtu jāveic vairākos atšķirīgos bioloģiskos audzēšanas apstākļos, lai apstiprinātu atlasīto šķirņu piemērotību audzēšanai dažādās bioloģiskajās saimniecībās.

3. nodaļā analizēts, kā divos atšķirīgos bioloģiskās un konvencionālās audzēšanas apstākļos izpaužas dažādas augu morfoloģiskās un fizioloģiskās īpašības, kas saistītas ar miežu šķirņu konkurētspēju ar nezālēm. Šis pētījums tika balstīts uz to pašu šķirņu kopumu, kas aprakstīts 2. nodaļā. Pagājušā gadsimta sākumā un senāk veidotām šķirnēm konstatēts morfoloģisko īpašību kopums, kas saistīts uz labu konkurētspēju ar nezālēm: salīdzinoši straujš attīstības temps augšanas sākumposmā, garas un platas un noliekušās lapas.

Tomēr šīm šķirnēm labvēlīgos augšanas apstākļos, kad veidojās garš stiebrs, izrādījās zema izturība pret veldrēšanos. Šķirnēm, kas izveidotas pagājušā gadsimta vidū un beigās (intensīva tipa), bija laba pielāgošanās spēja dažādiem audzēšanas apstākļiem. Tām bija vidēji garš stiebrs, kā rezultātā bija arī augstāka izturība pret veldrēšanos. Šīs šķirnes raksturojās ar strauju attīstības tempu augšanas sākumposmā, un tās sasniedza tādu pašu zelmeņa augstumu stiebrošanas fāzes sākumā kā vecākās šķirnes. Atsevišķas intensīvā tipa šķirnes izcēlās ar dažām morfoloģiskām pazīmēm, kas saistītas ar spēju nomākt nezāles. Tas nozīmē, ka principā modernā konvencionālā miežu selekcija var nodrošināt piemērotu izejmateriālu selekcijas programmām priekš bioloģiskās saimniekošanas sistēmas, ja pazīmes, kas saistītas ar konkurētspēju ar nezālēm, ir iekļautas kā izlases kritēriji. Ciešāka korelācija pazīmēm, kas saistītas ar konkurētspēju ar nezālēm, tika novērota starp abām bioloģiskajām audzēšanas vietām un konvencionālajiem audzēšanas apstākļiem ar vidēju mēslojuma daudzumu. Tāpēc izlase pēc šīm pazīmēm varētu tikt veikta šādos konvencionālos apstākļos. Lielākā daļa morfoloģisko pazīmju pozitīvi korelēja ar ražu tikai nelabvēlīgākos bioloģiskajos audzēšanas apstākļos ar lielu nezāļu īpatsvaru, savukārt negatīvā korelatīvā sakarība ar ražu tika iegūta bioloģiskajos audzēšanas apstākļos, kuros veic nezāļu ierobežošanas pasākumus. Veidojot šķirnes bioloģiskajai saimniekošanas sistēmai, jāņem vērā šī saistība starp graudu ražu un konkurētspēju ar nezālēm.

4. nodaļā analizēti fenotipiskās izlases rezultāti divām atšķirīgām hibrīdajām populācijām divos dažādos bioloģiskajos (tiešā selekcija) un konvencionālajos audzēšanas apstākļos (netiešā selekcija). Hibrīdo populāciju vecākaugi tika iepriekš pārbaudīti 2. un 3. nodaļā aprakstītajos izmēģinājumos. Iegūto izlases rezultātu salīdzināšanai savā starpā tika izstrādāts “piemērotības rādītājs bioloģiskajai saimniekošanas sistēmai”, kas ietver svarīgas miežu morfoloģiskās un fizioloģiskās pazīmes. Rezultāti parādīja, ka tas, vai efektīvāka ir tiešā (veikta bioloģiskajos audzēšanas apstākļos) vai netiešā (konvencionālajos audzēšanas apstākļos) izlase, ir atkarīgs no hibrīdizācijā izmantoto vecāku morfoloģiskajām pazīmēm. Selekcija populācijā ar morfoloģiski atšķirīgiem vecākiem (intensīva, īstiebraina šķirne ar augstu ražas potenciālu un plašu pielāgošanos dažādiem augšanas apstākļiem tika krustota ar garstiebrainu šķirni ar strauju attīstības

tempu augšanas sākumposmā), bioloģiskās audzēšanas apstākļos ar lielu nezālainību, ļāva izlasīt genotipus ar izcilu konkurētspēju ar nezālēm, bet toties zemu ražas potenciālu. Turpretim iepriekšminētās krustojumu kombinācijas līnijām, kas tika izlasītas trīs pārējās izlases vietās (optimāli bioloģiskie audzēšanas apstākļi un konvencionālās audzēšanas vietas ar vidēju un augstu mēslošanas fonu), bija augsta raža, bet zema konkurētspēja ar nezālēm. Pretēja tendence tika konstatēta populācijā ar morfoloģiski mazāk atšķirīgiem vecākiem (vidēju līdz garu augumu, ar stabilu, bet zemu līdz vidēju produktivitātes līmeni). No šīs kombinācijas atlasītās līnijas izcēlās ar augstu konkurētspēju ar nezālēm abās bioloģiskās audzēšanas vietās un netika novērotas lielas atšķirības starp dažādās selekcijas vietās izlasītām līnijām.

4. nodaļa atklāj, ka tiešā un netiešā selekcija agrīnās selekcijas stadijās ir vienlīdz piemērota bioloģiskajai saimniekošanas sistēmai paredzēto šķirņu veidošanai, ja tiek ņemti vērā šādi aspekti: selekcijas izejmateriāla izvēle ir ļoti svarīga, un jāvadās no tā, kur selekcijas programma tiek īstenota. Veicot selekciju bioloģiskos apstākļos, var izmantot pēc morfoloģiskajām pazīmēm atšķirīgāku selekcijas izejmateriālu. Lai veiktu selekciju konvencionālos apstākļos ar vidēju mēslojuma daudzumu, izejmateriāls (vecākaugi) ir jāizvēlas ar morfoloģiskajām iezīmēm, kas veicina konkurētspēju ar nezālēm un kas ir ar stabilu ražu. Šādas selekcijas programmas vēlākajos posmos ir jāveic perspektīvā materiāla papildus pārbaude dažādos bioloģiskos audzēšanas apstākļos.

5. nodaļā sīkāk analizēts, kā tiešā (bioloģiskajos audzēšanas apstākļos) vai netiešā (konvencionālajos audzēšanas apstākļos) selekcija ietekmē graudu ražu un īpašības, kas veicina konkurētspēju ar nezālēm. Šī pētījuma mērķis bija noskaidrot, kurš izlases veids nodrošina genotipiem gan augstu graudu ražu, gan augstu konkurētspēju ar nezālēm, lai tie būtu piemēroti audzēšanai bioloģiskajā saimniekošanas sistēmā. Pētījumā tika izmantoti 134 miežu genotipu dati divos bioloģiskajos un divos konvencionālajos audzēšanas apstākļos, kas iegūti trīs gadu laikā. Graudu raža un konkurētspēja ar nezālēm bija divi galvenie izlases kritēriji, kuri tika atšķirīgi kombinēti. Izlase bioloģiskajos audzēšanas apstākļos bija visefektīvākā, jo tā genotipos vislabāk apvieno gan ražību, gan konkurētspēju ar nezālēm. Labākie rezultāti tika iegūti, ja izlasei pēc konkurētspējas ar nezālēm sekoja stingra izlase pēc graudu ražas bioloģiskajos audzēšanas apstākļos. Genotipu izlasi

bioloģiskajai saimniekošanas sistēmai var veikt arī konvencionālos apstākļos, ja tiek ņemti vērā gan graudu raža, gan konkurētspēja ar nezālēm. Tomēr selekcija konvencionālos apstākļos izrādījās mazāk efektīva nekā tiešā selekcija bioloģiskos audzēšanas apstākļos, jo tika atlasīts mazāks genotipu skaits, kas uzrādīja gan augstu ražu, gan vienlaicīgi arī augstu konkurētspēju ar nezālēm, pārbaudot bioloģiskajos apstākļos.

6. nodaļā tika veikta asociāciju kartēšana ar mērķi identificēt kvantitatīvo pazīmju lokusus (QTL) bioloģiskajai saimniekošanas sistēmai svarīgākajām morfoloģiskajām pazīmēm. Asociāciju kartēšanas populācija sastāvēja no 153 dažādas izcelsmes miežu šķirnēm un selekcijas līnijām. Pazīmju fenotipēšana tika veikta graudu ražai un deviņām ar konkurētspēju ar nezālēm saistītām pazīmēm divos bioloģiskos un divos konvencionālos audzēšanas apstākļos. Genotipēšanā izmantoti 1536 viena nukleotīda polimorfisma (SNP) marķieri. Kopumā tika identificēti QTL četrām ar konkurētspēju pret nezālēm saistītām augu pazīmēm: zemeņa augstumam, lapu noliekšanās leņķim, karoglapas platumam un auga garumam ražas novākšanas laikā. Graudu ražai QTL netika atrasti. Kopumā tika identificēti 35 QTL, no kuriem desmit bija būtiski saistīti ar pazīmēm tikai bioloģiskajā un 18 tikai konvencionālajā audzēšanas sistēmā, un septiņi QTL bija kopīgi abām audzēšanas sistēmām. Tas nozīmē, ka lielākā daļa atklāto kvantitatīvo pazīmju lokusu (80%) bija specifiski saimniekošanas sistēmai. Bioloģiskajā saimniekošanas sistēmā zemeņa augstumam, lapu noliekšanās leņķim, karoglapas platumam un auga garumam kvantitatīvo pazīmju lokusi tika kartēti attiecīgi 3H, 7H, 6H un 2H hromosomās. Tika konstatēts, ka viens QTL (hromosomā 3H) ir saistīts ar vairākām pazīmēm (zemeņa augstumu, lapu noliekšanās leņķi un auga garumu) abās audzēšanas sistēmās. Identificētie QTL varētu kalpot marķieru izveidošanai, lai atlasītu miežu genotipus ar uzlabotu konkurētspēju ar nezālēm bioloģiskajai saimniekošanas sistēmai.

7. nodaļā ir apkopoti 1.–6. nodaļā aprakstītie pētījuma rezultāti kontekstā ar tā mērķi un izvirzītajiem pētījuma jautājumiem, lai optimizētu miežu šķirņu selekcijas procesu bioloģiskās saimniekošanas sistēmas vajadzībām, kas ietver selekcijas izejmateriālu, izlases kritērijus, kā arī vidi, kurā izlase būtu veicama. Šajā nodaļā iekļautas dažas papildus analīzes, lai padziļināti pētītu i) senāk un nesenāk izveidotu šķirņu vērtību, kā selekcijas izejmateriālam selekcijas programmās priekš bioloģiskās saimniekošanas sistēmas,

ii) krustojumu kombināciju īpašību lomu izlases efektivitātē gan tiešās, gan netiešās selekcijas procesā, un iii) cik lielā mērā bioloģiskās audzēšanas apstākļos identificētie QTL ir saistīti ar konkurētspēju ar nezālēm nodrošinotām pazīmēm un vai šos QTL varētu izmantot turpmākajā genotipu izlasē priekš bioloģiskās saimniekošanas sistēmas.

Šajā promocijas darbā iegūtās atziņas palīdzēs īstenot Eiropas Kopienas “Zaļā kursa” mērķus, un veidot šķirnes ne tikai bioloģiskajai, bet arī konvencionālajai saimniekošanas sistēmai, kā arī meklēt veidus, kā samazināt atkarību no ķīmiskajiem augu aizsardzības līdzekļiem un padarīt pārtikas ražošanas nozari videi draudzīgāku un ilgtspējīgāku.

Biography

Aina Kokare was born on January 29, 1965, in Murmastienes Pagasts in Latvia. After finishing elementary school, she attended Smiltene Agricultural Technical School (college), where she studied agronomy. After graduating from college, she continued her studies at the Latvia University of Life Sciences and Technologies (previously known as Latvia University of Agriculture) (in Jelgava), where she obtained a bachelor's degree and started working at the former Priekuli Breeding Station as an assistant in the Winter Cereals breeding group. At the same time, she started her master's studies and after completion she obtained her master's degree in agriculture. The topic of the master's thesis was the assessment of grain quality of winter rye varieties. After completing her master's studies, she continued working in breeding of winter rye and winter triticale in Priekuli. Later, within a financial contribution of the European Economic Area (EEA) Norwegian Financial Mechanism Project, she worked on the breeding of spring barley for organic farming when she met Prof. dr. Edith Lammerts van Bueren, with whom she established cooperation and she began her PhD study at Wageningen University, next to her full time job as breeder.

She had been involved in multiple project initiatives. For example, she has participated in the Norwegian Financial Mechanism EEA grant EEZ08AP-27 with the projects 'Elaboration of crop breeding strategy for organic farming' and 'Innovative approach to hull-less spring cereals and triticale use from human health perspective ('NFI/R/2014/11'). After that, she participated in the European Social Fund co-financed project 'Development, improvement, and implementation of environmentally friendly and sustainable crop breeding technologies'. She has also been involved in FP7 Research Project EUROLEGUME 'Enhancing of legumes growing in Europe through sustainable cropping for protein supply for food and feed', where her main tasks were: sourcing and evaluating the performance of local genetic resources of pea, and fava bean accessions for potential benefits concerning selective breeding for site-specific abiotic and biotic conditions. She collaborated in the recent European LIVESEED project 'Improve performance of organic agriculture by boosting organic seed and plant breeding efforts across Europe' from 2017-2021. Her main activities included: analysing the general mixing

ability between crop species mixtures (cereals with legumes) in an organic farming system.

She is a co-breeder of the rye variety Kaupo, which is widely grown in Latvia, the triticale variety Ruja that was registered in Estonia, and of pea variety Rebekka PR.

Currently, Aina Kokare is working as a breeder researcher in the Institute of Agricultural Resources and Economics, Department of Plant Breeding and Agroecology. The main research areas are legumes, including breeding for organic farming. She is a team member of the Latvian Council of Sciences and is now working in pea breeding for conventional and organic farming.

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PE&RC Training and Education Statement

With the training and education activities listed below the PhD candidate has complied with the requirements set by the C.T. de Wit Graduate School for Production Ecology and Resource Conservation (PE&RC) which comprises of a minimum total of 32 ECTS (= 22 weeks of activities)



Review of literature (6 ECTS)

- Plant breeding strategies for low-input farming systems

Writing of Project proposal (4.5 ECTS)

- Designing breeding strategies for organic agriculture in spring barley (*Hordeum vulgare* L.)

Post-graduate courses (4.8 ECTS)

- Root Ecology: Drivers of foraging and interactions in a spatial context; PE&RC, KU Copenhagen, DK (2012)
- Use of plant genetic resources for breeding and scientific research; Nordic Genetic Resource Centre (NordGen), University of Latvia, Riga (2014)
- NOVA PhD Course adaptation and resilience in plant breeding; Umeå, Sweden, SLU, Sweden (2017)
- Plant breeding & resilient seed systems, options for stakeholder engagement and benefit sharing; PE&RC, Wageningen University (2020)

Deficiency, refresh, brush-up courses (7.8 ECTS)

- Statistics, multivariate methods; Latvia University of Agriculture, Riga (2009)
- Methodology of agronomic trials; Latvia University of Agriculture, Riga (2010)
- Organic plant breeding & seed production; Wageningen University, (2011)

Competence strengthening / skills courses (3.7 ECTS)

- Scientific writing; Latvia University of Agriculture, Riga (2010)
- Techniques for writing and presenting a scientific paper; Wageningen University (2012)

PE&RC Annual meetings, seminars or weekends (0.6 ECTS)

- PE&RC Last years weekend (2017)

Discussion groups / local seminars or scientific meetings (6 ECTS)

- COST ACTION workshops, meetings; Driebergen, the Netherlands; La Besse, France (2005, 2006)
- Annual scientific seminar for PhD students in training and research farm; Latvia University of Agriculture Vecauce (2009, 2016)
- Trial seminar; Latvian Academy of Sciences, Priekuli, Latvia (2011)
- Diversity in plant breeding and agriculture: strategies for healthy lifestyle; Stende, Latvia (2012)
- Agriculture and processing technologies meeting; Latvian Academy of Sciences, Latvia (2015)

- The scientific and practical conference harmonious agriculture; the Latvia University of Agriculture, Jelgava, Latvia (2018, 2021))
- Challenges in plant breeding: the role of plant breeding in agricultural development in the future; Jelgava, Latvia (2019)
- Annual scientific practical conference; the Latvia University of Agriculture, Latvia (2020)
- Annual scientific meeting of LIVESEED project; Latvia (2021)

International symposia, workshops and conferences (9.5 ECTS)

- EUCARPIA/PE&RC Symposium of working group organic plant breeding, plant breeding for organic and sustainable, low-input agriculture: dealing with genotype-environment interactions; Wageningen, the Netherlands (2007)
- International conference development of plant breeding and crop management in time and space; Priekule, Latvia (2008)
- EUCARPIA/Bioexploit Workshop on the role of marker assisted selection in breeding varieties for organic agriculture; Wageningen, the Netherlands (2009)
- IFOAM 1st World conference on organic animal plant breeding; Santa Fe, New Mexico (2009)
- Eucarpia Section organic low-input breeding conference on breeding for resilience; Paris, France (2010)
- International scientific conference research for rural development agricultural; Latvia University of Agriculture, Jelgava (2010)
- ECO-PB 10 years anniversary conference organic plant breeding; Frankfurt, Germany (2011)
- International conference crop breeding and management for environmentally friendly farming; Priekule, Latvia (2013)
- EUCARPIA Section organic and low input agriculture and EU NUE-CROPS project; Göttingen, Germany (2013)
- 20th Eucarpia congress plant breeding, the art of bringing science to life; Zurich, Switzerland (2016)
- 3rd Meeting of the section of agronomy and physiology of EAPR; Rīga, Latvia (2016)
- NJF Conference, legumes from field to fork a Nordic-Baltic perspective on production, development and marketing of legumes (2017)
- 8th IOBC – WPRS working group meeting landscape management for functional biodiversity; Wageningen, the Netherlands (2018)
- Workshop organic seed in the Baltic States: how to increase the production and use; Riga, Latvia (2019)
- Eucarpia section organic and low input agriculture on breeding and seed sector innovations for organic food systems; Latvia (2021)
- 13th International barley genetics symposium; Riga, Latvia (2022)

Lecturing / supervision of practicals / tutorials (0.6 ECTS)

- Biology, ecology and cultivation characteristics of rye; the Latvia University of Agriculture, Jelgava (2017)
- Biological characteristics and quality indicators of spring pea; the Latvia University of Agriculture, Jelgava (2018)

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