GENETIC AND PHYSIOLOGICAL RELATIONSHIPS BETWEEN OAT GRAIN QUALITY

COMPONENTS

A Dissertation Submitted to the Graduate Faculty of the North Dakota State University of Agriculture and Applied Science

By

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In Partial Fulfillment for the Degree of DOCTOR OF PHILOSOPHY

Major Department: Plant Sciences

December 2013

Fargo, North Dakota

North Dakota State University Graduate School

Title

Genetic and Physiological Relationships between Oat Grain Quality Components

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The Supervisory Committee certifies that this *disquisition* complies with

North Dakota State University's regulations and meets the accepted standards

for the degree of

DOCTOR OF PHILOSOPHY

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ABSTRACT

The use of oats for human consumption is increasing every day due to the health benefits of oat products. With the objective to study relationships among factors affecting oat grain quality, two Recombinant Inbred Lines (RIL) mapping populations ('ND030299' x 'ND991151' and 'ND030299' x 'Souris') have been used in this study. The two populations with their parents and three check cultivars were evaluated in a square lattice design in 2008 and 2009 at two North Dakota locations. Data were recorded on the following agronomic traits: grain yield, test weight, 1000 kernel weight, thin kernels, heading date, and plant height. Chemical and grain physical analysis were performed for β-glucan, oil, and groat percentage. A total of 4975 SNP markers were assessed on the two populations using a 32-bead chip platform developed by Illumina. QTLs for agronomic and grain physical traits were mapped and characterized in the two populations using Windows QTL Cartographer. Grain yield was positively correlated with test weight, thin kernels, plant height, β -glucan content, and associated negatively with 1000 kernel weight. Thirty linkage groups using 1168 polymorphic markers were formed for population 05021, whereas population 05026 comprised 33 linkage groups using 1024 polymorphic markers. The 30 linkage groups of population 05021 contained from 3 to 62 markers, and varied in size from 15.8 to 225.3 cM for a total map size of 2601.7 cM. The 33 linkage groups of population 05026 comprised from 2 to 42 markers, and varied in size from 2.3 to 143.2 cM for a total map size of 1174.2 cM. Nineteen genomic regions on 14 linkage groups were significantly associated with agronomic and grain chemical traits in the population 05021. Fourteen genomic regions on 12 linkage groups were identified for agronomic traits in the population 05026. The same genomic region on LG 05021-16 was associated with thin kernels, test weight, 1000 kernel weight, and oil content. LG 05026-19 loci, from position 23.7 to 47 cM, had strong effects on

heading date, plant height, and grain yield. The QTLs consistently detected across environments and between the two populations could serve as starting points for marker-assisted selection.

ACKNOWLEDGEMENTS

I would like to express my deepest gratitude to my major advisor Dr Michael McMullen, who has continually and convincingly conveyed a spirit of adventure in regard to research. Without his guidance, help and patience this dissertation would not have been possible.

I would like to thank my committee members: Drs. Elias Elias, Juan M. Osorno, and John Bitzan for their contributions and suggestions to this research, and for taking time to review the final manuscript. Special thanks go to Robert Baumann and the NDSU oat team for organizing and taking care of all the field and greenhouse experiments.

I would also like to thank my friends Raphael Colbert and family, Angela Linares, Renata Jung, Ana Maria Correa, and Roberto Luciano. They were always supporting me and encouraging me with their best wishes.

Finally, I would like to thank my parents and my family in Haiti and Florida.

I dedicate this research to my grandparents: Monsieur and Madame Georges Jean Philippe.

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DISSERTATION ORGANIZATION

The present dissertation contains five chapters including a general introduction (Chapter 1), three body chapters (Chapters 2 - 4), and a general conclusion. Each of the 3 body chapters is a separate manuscript that will be submitted for publication. Chapter 1 includes a general introduction along with a literature review portion and the objectives of the dissertation research. Chapter 2 addresses objective 1 of the dissertation and consists essentially in the evaluation of genotypic and phenotypic relationship among agronomic and grain chemical traits affecting oat grain quality. Chapter 3 covers objective 2 of the dissertation and presents the construction of genetic linkage maps from two spring oat populations followed by the comparison of homologous linkage groups between the two populations and the recently published oat consensus map. Chapter 4 identifies genomic regions associated with genes underlying the expression of agronomic and grain chemical traits in two oat populations by addressing objective 3 of the dissertation. Chapter 5 is the general conclusion and gives a brief outline of the research.

CHAPTER 1

1.1. General introduction

North Dakota is one of the states that leads grain production in the North Central plains region and is a considerable oat (*Avena sativa L.*) producer. During the past decade, North Dakota has been consistently among the five states leading oat production in the United States with acreage of about 176,000 ha. In 2012, the amount of oat produced in North Dakota was 100,000 tonnes (t) and the planted area was 80,937 ha. The average price for oat in 2012 was approximately US \$46.5 kg⁻¹ and oat production represents a value of \$US 21.9 million in the global economy of the state (USDA-NASS, 2013).

Oat has many uses worldwide: food industry and animal feed. The use of oats for human consumption is increasing every day due to the health benefits of oat products. Oat products reduce cholesterol and arthrosclerosis, decrease the risk of diabetes, and provide antioxidant protection and supply a number of important nutrients (Schrickel, 1986; Murphy and Hoffman, 1992). Oat is a good source of calcium (0.25 - 0.32% dry weight) (Broadley et al., 2003; Tiwari et al., 2006), iron (300 mg kg⁻¹), magnesium (0.6% DW) and potassium (2% DW) (Peterson et al., 1974) and it is rich in total dietary fiber and high in essential fatty acids. The protein content of oat is high relative to other cereals and of excellent quality for human and monogastric animal nutrition (Casey, 2008).

During the past 4 years in ND, a reduction in the oat production has been observed. For example, the production in 2012 was 60% smaller than 2009 and the estimated yield (2.2 t/ha) was down 0.2 t/ha (USDA, 2013). The principal cause of this decrease could be disease pressure, shifting farm practices, and bad weather conditions including rotation of drought and flood conditions. Oat is subject to a large number of diseases that can cause severe damage to quality

and reduce yield. The most common oat diseases, crown rust caused by *Puccinia coronata* Corda var. *avenae* W.P. Fraser & Ledingham, stem rust caused by *Puccinia graminis* Pers:Pers. f. sp. *avenae* and barley yellow dwarf virus (BYDV) can reduce significantly the grain quality. Milling markets require high quality standards such as high test weight, bright color, high groat percentage, low oil content, high protein and high β -glucan content (Ransom et al., 2007). Development of new oat varieties with enhanced grain quality and resistance to diseases would be desirable for the future of oat production in North Dakota.

Cultivated oats are an allohexaploid species having 21 pairs of chromosomes with basic chromosome number of 7 (2n = 6x = 42) (Zhu and Kaeppler, 2003) and a relatively large genome of 11315 Mbp (Pal et al., 2002). Molecular mapping research in oat was initiated in the 1990s with the publication of the first molecular map of hexaploid oat based primarily on restriction fragment length polymorphism (RFLP) markers (O'Donoughue et al., 1995). Other mapping studies based on amplified fragment length polymorphism (AFLP) markers, simple sequence repeat (SSR) markers, and diversity array technology (DArT) markers were also reported (Jin et al., 2000; Portyanko et al., 2001; Zhu and Kaeppler, 2003; Tinker et al., 2009). An important advance, in the study of oat genome, was the recent publication of a linkage map containing 21 linkage groups, anchoring the 21 oat chromosomes, using mainly single nucleotide polymorphism (SNP) markers (Oliver et al., 2013). Nevertheless despite more than 20 years of genomic research, oat has lagged behind and has not kept pace with other small grains such as wheat and barley. As a result, quantitative trait loci (QTL) analysis and marker assisted selection has not been used at a large scale in oat cultivar development.

Today the world population is increasing quickly at a rate ever observed, and by the year 2050 the world population will reach approximately 12 billion people. At such rate of increase,

the world will need to produce more than twice the amount of food we are producing now. As a consequence, the continuous development of genotypes with improved qualities is of great importance (Todorovska et al., 2005). Detection of important QTLs and marker-assisted selection (MAS) are potential tools with the objective to increase efficiency by allowing earlier selection and reducing plant population size used during selection. The presence of tight linkage, less than 10 cM, between an important economic trait and a genetic marker can be useful in MAS to increase gain from selection (Todorovska et al., 2005).

1.2. Objectives

The objectives of this research are:

- Evaluate through a two year multi-environment experiment, genotypic and phenotypic relationships among factors such as grain yield, test weight, 1000 kernel weight, thin kernels, and β-glucan content affecting oat grain quality
- Develop a genetic linkage map in two oat populations using SNP and DArT markers.
- Identify quantitative trait loci (QTLs) associated with agronomic traits in two oat populations.

1.3. Literature review

Oat is an important cereal crop used for both human consumption and animal feed (Zhu and Kaeppler, 2003; Peterson et al., 2005) and has long been recognized as a high-quality food and feed (Rines et al., 2006). The beneficial effects and the positive impact of the total dietary fiber on some of the risk factors of cardiovascular diseases have stimulated great interest in oat (Redaelli et al., 2009). The objective of oat breeders is to improve yield while maintaining good grain quality. Some traits important for growers and millers and related to oat grain quality

include agronomic traits (yield, plant height, and lodging, heading date and pest resistance), grain physical traits (groat and kernel weight, test weight, groat percentage) and grain chemical composition traits (protein, oil, β -glucan) (Peterson et al., 2005). Doehlert et al., 2001 indicated that test weight, groat percentage, groat weight and groat composition were the characteristics most commonly used to describe oat quality. When considering the widespread use of oat in human food and animal feed, modern oat breeders utilize biochemical traits to measure grain quality and thus protein content, oil content, and β -glucan content constitute the major groat compositional characteristics related to oat grain quality (Doehlert et al., 2001). Oat grain quality also is related to grain yield production and conditions that lead to production of high yields generally also lead to improved grain quality.

1.3.1. Physical traits affecting oat grain quality

1.3.1.1. Test weight

Among the physical traits that affect grain quality, test weight has been the traditional way to measure oat grain quality and high test weight in oat generally is associated with high grain quality (Forsberg and Reeves, 1992; Doehlert and McMullen, 2008). Test weight, also known as bulk density, can be defined as the weight of grain that fits into a specific volume (Forsberg and Reeves, 1992) and represents the bulk density of oat grain i.e. the measure of the density of oat grain as they are packed into a given volume (Doehlert et al., 2001). Test weight is a volumetric measure and it is commonly expressed in kg m⁻³ or g L⁻¹. Forsberg and Reeves (1992) reported that the standard test weight of oat grain is 412 kg m⁻³ and modern oat cultivars have test weight values ranging from 463 to 515 kg m⁻³. Several factors influence oat test weight such as kernel size and shape, groat density, groat percentage, presence of awns and disease (Forsberg and Reeves, 1992; Doehlert et al., 2001). Doehlert et al., 2006 found a negative relationship between

test weight and grain length and a positive relationship between test weight and grain width. They concluded that long kernels with unstable hulls or awns have more space between them and pack less well than shorter kernels. Jianzhong, 2005 concluded that high test weight is dominant over low test weight by crossing two lines, one with low test weight (496 kg m⁻³) and the other with high test weight (553 kg m⁻³), with all the progeny exhibiting high test weight ranging from 534 to 577 kg m⁻³ with a mean value of 555 kg m⁻³.

1.3.1.2 Groat percentage

The groat or caryopsis refers to the kernel after the hulls (lemma and palea) have been removed (Ransom et al., 2007) and is a primary determining factor in grain quality, whether for feed or milling purposes (Rines et al., 2006). The groat percentage, also known as groat proportion and caryopsis percentage, is the measure of the proportion of the whole oat that is recovered as groat after dehulling (Doehlert et al., 2001) and represents the economic yield that a given lot of oat grain can produce (Doehlert et al., 1999). Depending on genotype and growth conditions, groat percentage in oat grain varied from 70 to 75% in non-stress environments, but can be much lower in environments under biotic and abiotic stress (Rines et al., 2006). Some oat cultivars are hulless (naked oat), but grain with the hull present is often preferred for production and processing. The principal reason is that naked oats, due to the soft texture of the kernels, are more susceptible to weathering and discoloration, saprophytic fungal invasion, and harvest damage (Rines et al., 2006). Bartley and Weiss (1951) indicated that groat percentage was highly influenced by environmental effects whereas Doehlert et al., 2001 found that groat percentage was equally affected by environment and genotype. Ronald et al., 1999 and Wesenberg and Shands, 1973 found that groat percentage was a quantitatively inherited trait with a broad sense heritability of 0.36 to 0.92.

1.3.2. Biochemical traits affecting oat grain quality

1.3.2.1. Oil content

Oat kernels contain a high concentration of lipid which is uniformly distributed throughout the grain and a favorable ratio of unsaturated to saturated fatty acids (Forsberg and Reeves, 1992; Casey, 2008). The major content of oil in oat grain is found in the endosperm (Peterson, 1992) and range between 40 and 110 g kg⁻¹ (Rines et al., 2006). However, Holland et al., 2001 reported oil content of 180 g kg⁻¹ in an experimental line developed by recurrent selection. High oil content is advantageous for animal feeding because of its high content of energy, but in food applications, high oil concentration is deleterious because of its potential to produce enzymatic rancidity reactions that give rise to bitter, grassy or other undesirable flavors (Kianian et al., 1999; Doehlert et al., 2001). Rines et al., 2006 indicated that shelf life of oat products can be severely affected by high oil content with high proportion of unsaturated fatty acids. Youngs et al., 1982 in a survey including studies conducted in various countries reported lipid concentration on oat groat ranging from 20 to 118 g kg⁻¹. Luby and Stuthman, 1983 reported lipid concentration in A. sativa ranged from 56 to 76 g kg⁻¹. In a study involving 12 genotypes of oat grown at four different locations in ND, Doehlert et al., 2001 found that groat lipid concentration varied from 46.4 to 78.1 g kg⁻¹ with a mean of 62.9 g kg⁻¹.

Youngs and Foster, 1979 and Gullord, 1986 showed that fatty acid composition for oat genotypes is relatively stable over a wide range of environments, which indicated that environment does not affect fatty acid composition. Doehlert et al., 2001 demonstrated also the high stability of lipid concentration in oat by reporting a mean square 10 fold greater for genotype than for environment. Broad sense heritability estimated and reported by Karow and Forsberg, 1984 for the different components of oat oil have been high: 0.63 to 0.91 for palmitic

acid, 0.66 to 0.99 for oleic acid and 0.64 to 0.94 for linoleic acid. Because of the stability of oil in oat kernel and the fact that it is highly heritable, breeders can select for both high and low oil with little confounding from environment.

<u>1.3.2.2</u>. β-glucan content

Oat is particularly high in dietary fiber which can be defined as plant polysaccharides and lignin that are resistant to hydrolysis by human digestive enzymes (Peterson, 1992; Casey, 2008). Depending on genotype and environmental growth conditions, β-glucan content in oat varied from 30 to 60 g kg⁻¹ (Rines et al., 2006). However, an experimental line with 71 g kg⁻¹ of β glucan had been reported by Cervantes-Martinez et al., 2001. The dietary fiber of oat is a mixture of soluble and insoluble fractions. The presence of a high concentration of $[(1\rightarrow 3), (1\rightarrow 4) -\beta$ -Dglucans] known, as β -glucan, in the soluble fraction is high relative to other cereals (Peterson, 1992). β-glucan resides in the cell walls of the subaleurone regions throughout the bran and the endosperm (Bacic and Stone, 1981); it is also present in the inner region of the aleurone cell walls (Peterson, 1992). β -glucan lowers the serum cholesterol levels of blood, balances the glucose and insulin content of serum after meals and reduces the risk of cardiovascular diseases (Anderson and Chen, 1986). On the other hand, feed rations high in β -glucan can be detrimental to the weight gain of animals due to the energy differential in metabolism (Anderson et al., 1978). High β-glucan and low oil content are suitable in the food market whereas low β-glucan and high oil-content are more favorable for the feed industry. Doehlert et al., 2001 found that βglucan content for genotypes adapted to oat production in ND varied from 43.3 to 57.6 g kg⁻¹ with a mean of 50.3 g kg⁻¹. Kianan et al., 2000 evaluated recombinant inbred lines from two populations: Kanota x Ogle and Kanota x Marion. They found that the β-glucan content in the population Kanota x Ogle varied from 37.2 to 57.6 g kg⁻¹ with a mean of 45.3 g kg⁻¹ while in the

population Kanota x Marion the β-glucan content ranged from 42.8 to 59.6 g kg⁻¹ with an average of 50.6 g kg⁻¹. Lee et al., 1997, in a study conducted in ND with 10 oats cultivars grown at two locations, reported values of total β-glucan content ranged from 44.4 to 60.5 g kg⁻¹. Levels of β-glucan in oat groat are influenced by both genetic and environmental factors (Doehlert et al., 2001; Lee et al., 1997). Holthaus et al., 1996 and Kianian et al., 2000 reported that β-glucan content in oat is controlled by multiple genetic loci with primarily additive effects and its broad sense heritability is estimated at 0.55. Kianian et al., 2000 using Restriction Fragment Length Polymorphism (RFLP) analysis in 137 Recombinant Inbred Lines (RIL) developed from the cross Kanota x Ogle showed that regions on linkage groups 3, 6, 11, 13, 14, 17, 20 contributed significantly to the β-glucan content. Kanota contributed three alleles and Ogle contributed four alleles for the loci that had a positive influence on this trait.

1.3.3. Cytogenetics of oats

Oats belong to the family Gramineae (Poaceae) and to the genus *Avena*, which consist of diploid (2n = 2x = 14), tetraploid (2n = 4x = 28) and hexaploid (2n = 6x = 42) species (Rines et al., 2006). The diploid species contain A or C genome (AA or CC), tetraploid species have either AB (AABB) or AC (AACC) genomes, and the hexaploid such as cultivated species *A. sativa* and *A. byzantina* have ACD (AACCDD) genomes (Drossou et al., 2004; Morikawa and Nishihara, 2009). It has been proposed that the evolution of cultivated oats from diploid and tetraploid species occurred in two steps (Li et al., 2000; Nikoloudakis et al., 2008). First, an interspecific hybridization of two diploid species occurred to produce a tetraploid (AACC) after chromosome doubling. After which, this tetraploid species intercrossed with a diploid to form the hexaploid, *A. sativa*, by doubling of the chromosomes. The diploid and tetraploid progenitors of the cultivated oats have been subject of many discussions.

Initially, A. strigosa was considered the A genome donor of hexaploids (Rajhathy and Thomas, 1974). However, southern hybridization of a satellite DNA sequence isolated from A. strigosa showed dissimilarities among the As and the A/D genome chromosomes (Linares et al., 1998). Li et al. 2000 using satellite DNA sequence reported that Ac genome of A. canariensis is the progenitor and A genome donor of hexaploids. Recent molecular study of Nikoloudakis et al., 2008 using restriction fragment length polymorphism (RFLP) analysis to screen 54 accessions containing 35 diploid, 12 tetraploid and seven hexaploid suggested that the diploid A. longiglumis is the donor of the A genome. In contrast, Fu and Williams, 2008 found out that A. maroccana (AACC) is the tetraploid progenitor of the hexaploid oats, A. wiestii is the diploid donor of the A genome, and the diploid A. eriantha appears to be the C genome donor. Sequence Characterized Amplified Region (SCAR) markers analysis among 29 accessions done by Nikoloudakis and Katsiotis, 2008 provided molecular evidence that the diploid A. ventricosa is the C genome donor. Moreover, they observed major dissimilarities to A. eriantha and A. clauda species (CpCp) lacking support of their possible participation in the Avena allopolyploid formation. A previous report confirmed that A. ventricosa is the C genome donor for both tetraploid and hexaploid species (Loskutov, 2001). For the D genome donor, the molecular study of Linares et al., 1998 revealed that some chromosome pairs in the A genome have been identified in the D genome, suggesting that the A and the D genomes are closely related. Leggett, 1996 proposed that the D genome did not exist and could be derived from the A genome. Li et al., 2000 used satellite DNA sequence from a microsatellite library of A. sativa to sequence 12 wild oat species. They suggested that the wild hexaploid, A. fatua, is the direct ancestor of the cultivated hexaploids, A. sativa and A. byzantina. Based on complex studies, the identification of the two basic genomes (A and C) was confirmed, however B and D genomes seemed to be derivatives of the A genome (Loskutov, 2008).

1.3.4. Development of molecular linkage maps in oat

1.3.4.1. Mapping in diploid species

Cultivated oats are allohexaploid species having 21 pairs of chromosomes with basic chromosome number of 7 (2n = 6x = 42) (Zhu and Kaeppler, 2003) with a relatively large genome of 11315 Mbp (Pal et al., 2002). Genomic research in oats began in 1992 by the group of O'Donoughue. In order to reduce the complexity of map construction relative to the hexaploid cultivated oats, and to facilitate the future construction of hexaploid maps, the first linkage maps for the oat genome were constructed using diploid species. O'Donoughue et al., 1992 were able to publish the first molecular map for diploid species based on 44 F₂ families derived from a cross between two nondomesticated species *A. atlantica x A. hirtula* using RFLP markers. According to Moore et al., 1995, and Devros and Gale, 2000 this map turned into a pillar in comparative mapping among grasses because the same RFLP markers were mapped in several other grass species. The second diploid oat *A. strigosa* and nondomesticated *A. wiestii*. That map was corrected and enhanced later by Yu and Wise, 2000 using AFLP markers, and by Kremer et al., 2001 using RFLP markers.

1.3.4.2. Mapping in hexaploid oats

In 1995, O'Donoughue published the first hexaploid map (KO map) based on 71 recombinant inbred lines (RILs) from two parents *A. byzantina* cv 'Kanota' and *A. sativa* cv 'Ogle' using RFLP markers. They reported 38 linkage groups that covered genetic distance of

1482 cM with estimated the complete map size of oats to be 2932 cM (O'Donoughue et al., 1995). The KO map is considered as the primary base map in cultivated oat.

Others researchers such as Jin et al., 2000, and Portyanko et al., 2001, independently worked on the enhancement of the original map using AFLP markers. The original map coverage was expanded from 1482 cM to 2049 cM (Portyanko et al., 2001) and 2351 cM (Jin et al., 2000), respectively. However, some problems were encountered such as the number of linkage groups. The map had 38 linkage groups which represented an excess of 17 on the basis of the expected 21chromosome of hexaploid oats. The failure to assign the 38 linkage groups with the 21 oat chromosomes, despite the use of a complete set of 21 monosomics lines from Kanota (Morikawa, 1975), has been associated to the relative small population (71 RILs) used (Rines et al., 2006). The presence of translocations and other chromosomal rearrangements (Jellen et al., 1993) is another difficulty pointed out in mapping in the KO population. These translocations can cause segregation distortions and make it difficult the ordering of markers within the linkage groups.

In 2003, Zhu and Kaeppler developed a new map using more RILs (152 instead of 71) with combination of RFLP, AFLP and SSR markers to address the problem on linkage groups. They used two hexaploid cultivated genotypes: 'MAM17-5' from the spring oat breeding program of the University of Wisconsin-Madison and 'Ogle' from the oat breeding program of the University of Illinois. These two cultivars showed different characteristics relative to crown rust resistance, plant height, days to heading, barley yellow dwarf virus resistance, groat oil content, and groat protein content. The 151 RILs were produced by the single-seed descent method. They also produced 159 RFLP probes and a combination of 64 AFLP primers by

digesting DNA with four restriction enzymes: DraI, EcoRI, EcoRV, and HindIII. The final map contained 476 marker loci, 28 linkage groups of 5 cM or longer and represented 1396.7 cM.

Tinker et al. 2009 developed a new map using the new Diversity Array Technology (DArT). DArT is based on microarray technology that enables whole-genome profiling even without a need for sequence information (Wenzl et al., 2004). The markers developed are biallelic, which can either be dominant or co-dominant (Kilian et al., 2005; Gupta et al. 2008). DArT array as discussed by Kilian et al. 2005 is an assembly of genomic DNA from a pool of individuals representing the genetic diversity of a species. The DNA mixture is then subjected to complexity reduction to lower the level of repetitive DNA. Complexity reduction involves combination restriction enzymes, adapter ligation, and amplification of adapter-ligated fragments (Wenzl et al., 2004; Kilian et al., 2005). The genomic fragments, known as genomic representation, are amplified and cloned to a vector into Escherichia coli to construct a library. Gupta et al. 2008 described that the labeled genomic representations are hybridized. Those polymorphic clones referred as DArT markers are assembled into a genotyping array for routine assay. DArTsoft software is used for analysis of hybridization intensities. Tinker el al., 2009 used a population of 80 recombinant inbred lines (RILs) from a cross between the genotypes 'Kanota' x 'Ogle' (KxO) including most of the 71 RILs from the map developed by O'Donoughue et al. 1995 to develop the map. A clone library was constructed from 60 cultivated and 14 non-cultivated accessions using PstI/TaqI method. The molecular mapping was done using 1010 DArT markers and 287 markers from previous map. The final map had 30 linkage groups for 2028 cM.

The discovery of single nucleotide polymorphism (SNP) markers was an important step in genetic studies. SNP markers are the most common type of DNA-based markers, represent the smallest unit of genetic variation, and can provide a rich source of useful molecular markers (Cho et al., 1999). For example, 90% of the human genome are SNPs (Kwok and Gu, 1999). Tenaillon et al., 2001 reported 1 SNP every 104 base pair (bp) in the maize genome. Among 15 soybean genotypes, Van et al., 2005 found out that SNPs occurred at a frequency of 1 per 2038 bp in 16302 bp of coding sequence, and 1 per 191 bp in 16960 bp of noncoding regions. As a result, SNPs are useful in the construction of high-density genetic maps because they can be analyzed using high-throughput systems (Van et al., 2005).

The most recent map of hexaploid oat which conciliates the number of linkage groups with the number of chromosomes was developed by Oliver et al., 2013 using a new chromosome anchoring strategy and SNP markers. They developed six mapping populations and derived monosomics lines from the cultivars 'Kanota' and 'Sun II' to anchor the linkage groups to the 21 oat chromosomes. The molecular map was constructed using 1054 SNP markers and contained 21 linkage groups, anchored to the 21 chromosomes, with a total length of 1838 cM. Based on an iterative mapping approach to remove problematic loci and multiple crossovers, the total genetic length of the oat genome is estimated to be closer to 2000 cM (Oliver et al., 2013).

1.4. References

- Anderson, M.A., J.A. Cook, and B.A. Stone. 1978. Enzymatic determination of 1,3:1,4-βglucans in barley grain and other cereals. J. Inst. Brew. 84:233.
- Anderson, J.W., and W.L. Chen. 1986. Cholesterol-lowering properties of oat products. p 309-333. *In* F.W. Webster (ed.). Oats: chemistry and technology. Am. Assoc. Cereal Chem. St Paul, MN.
- Bacic, A., B.A. Stone. 1981. Chemistry and organization of aleurone cell wall components from wheat and barley. Australian Journal of Plant Physiology 8:475-495.
- Bartley, B.G., and M.G. Weiss. 1951. Evaluation of physical factors affecting quality of oat varieties from bond parentage. Agron. J. 43:22-25.

- Broadley, M.R., H.C. Bowen, H.L. Cotterill, J.P. Hammond, M.C. Meacham, A. Mead, P.J. White. 2003. Variation in the shoot calcium content of angiosperms. Journal of Experimental Botany 54:1431–1446.
- Casey, A. 2008. A new healthier oat. Available at http://www.ceresorganic.com/High%20Beta%20Glucan%20Oats%20article%20vDC2.pd f.
- Cervantes-Martinez, C.T., K.J. Frey, P.J. White, D.M. Wesenberg, J.B. Holland. 2001. Selection for greater β-glucan content in oat grain. Crop Sci 41:1085-1091.
- Cho, R.J., M. Mindrinos, D.R. Richards, R.J. Sapolsky, M. Anderson, E. Drenkard, J. Dewdney, T.L. Reuber, M. Stammers, N. Federspiel, A. Theologis, W.H. Yang, E. Hubbell, M. Au, E.Y. Chung, D. Lashkari, B. Lemieux, C. Dean, R.J. Lipshutz, F.M. Ausubel, R.W. Davis, and P.J. Oefner. 1999. Genome-wide mapping with biallelic markers in Arabidopsis thaliana. Nat. Genet. 23:203-207.
- Devos, K.M., and M.D. Gale. 2000. Genome relationships: the grass model in current research. Plant Cell 12:637-646.
- Doehlert, C.D., and M.S. McMullen. 2008. Oat grain density measurement by sand displacement and analysis of physical components of test weight. Cereal Chem. 85: 654-659.
- Doehlert, C.D., M.S. McMullen, and R.R. Baumann. 1999. Factors affecting groat percentage in oat. Crop Sci. 39:1858-1865.
- Doehlert, C.D., M.S. McMullen, and J.J. Hammond. 2001. Genotypic and environmental effects on grain yield and quality of oat grown in North Dakota. Crop Sci. 41:1066-1072.
- Doehlert, C.D., M.S. McMullen, and J.L. Jannink. 2006. Oat grain/groat size ratios: A physical basis for test weight. Cereal Chem. 83:114-118.
- Drossou, A., A. Katsiotis, J.M. Leggett, M. Loukas, and S. Tsakas. 2004. Genome and species relationships in genus Avena based on RAPD and AFLP molecular markers. Theor. Appl. Genet. 109:48-54.
- Forsberg, R.A., and D.L. Reeves. 1992. Breeding oats cultivars for improved grain quality. p. 751-775. In H. G. Marshall and M. E. Sorrells (ed.) Oat science and technology. ASA, Madison, WI.
- Fu, Y.B., and D.J. Williams. 2008. AFLP variation in 25 Avena species. Theor. Appl. Genet. 117:333-342.
- Gullord, M. 1986. Oil and protein content in oats (Avena sativa L.). p. 210-213. *In* D. A. Lawes and H. Thomas (ed.) Proc. 2nd Int. oats conf., Aberstwyth, Wales. 15-18 july 1985. Martinus Nijhoff Publ., Dordrecht, Netherlands.

- Gupta, P.K., S. Rustgi, and R.R. Mir. 2008. Array-based high-throughput DNA markers for crop improvement. Heredity 101:5-18.
- Holland, J.B., K.J. Frey, and E.G. Hammond. 2001. Correlated responses of fatty acid composition, grain quality and agronomic traits to nine cycle of recurrent selection for increased oil content in oat. Euphytica 122:69-79.
- Holthaus, J.F., J.B. Holland, P.J. White, and K.J. Frey. 1996. Inheritance of β-glucan content of oat grain. Crop Sci. 36:567-572.
- Jellen, E.W., W.L. Rooney, R.L. Phillips, and H.W. Rines. 1993. Characterization of the hexaploid oat Avena byzantina cv. Kanota monosomic series using C-banding and RFLPs. Genome 36:962-970.
- Jianzhong, Y. 2005. Advanced backcross QTL analysis and genetic study of an introgressed powdery-mildew resistance gene derived from Avena macrostachya in oat (*Avena sativa*). Ph. D. diss. Martin-Luther Universitat. Wittenberg, Germany. 94p.
- Jin H., L.L. Domier, X. Shen, and F.L. Kolb. 2000. Combined AFLP and RFLP mapping in two hexaploid oat recombinant inbred populations. Genome 43:94-101.
- Karow, R.S., and R.A. Forsberg. 1984. Oil composition in parental F1 and F2 populations of two oat crosses. Crop Sci. 24:629-632.
- Kianian, S.F., M.A. Egli, R.L. Philips, H.W. Rines, D.A. somers, B.G. Gengenbach, F.H. Webster, S.M. Livingston, S. Groh, L.S. O'Donoughue, M.E. Sorrells, D.M. Wesenberg, D.D. Stuthman, and R.G. Fulcher. 1999. Association of groat oil content and acetyl-coA carboxylase in oat. Theor. Appl. Genet. 98:884-894.
- Kianian, S.F., R.L. Phillips, H.W. Rines, R.G. Fulcher, F.H. Webster, and D.D. Stuthmann. 2000. Quantitative trait loci influencing β -glucan content in oat (Avena sativa, 2n = 6x = 42). Theor. Appl. Genet. 101:1039-1048.
- Kilian, A., E. Huttner, P. Wenzl, D. Jaccoud, J. Carling, V. Caig, M. Evers, K. Heller-Uszynska, G. Uszynski, C. Cayla, S. Patarapuwadol, L. Xia, S. Yang, and B. Thomson. 2005. The fast and the cheap: SNP and DArT-based whole genome profiling for crop improvement. p. 443-461. In R. Tuberosa et al. (ed.) Proc. Intl. Congr. "In the Wake of the Double Helix: from the Green Revolution to the Gene Revolution", 27-31 May 2003. Bologna, Italy.
- Kremer, C.A., M. Lee, and J.B. Holland. 2001. A restriction fragment length polymorphism based linkage map of a diploid Avena recombinant inbred line population. Genome 44:192-204.
- Kwok, P.Y., and Z. Gu. 1999. Single nucleotide polymorphism libraries: why and how are we building them? Mol Med Today 5:538-543.

- Lee, C.J., R.D. Horsley, F.A.Manthey, and P.B. Schwarz. 1997. Comparisons of β-glucan of barley and oat. Cereal Chem. 74:571–575.
- Leggett, J.M. 1996. Using and conserving Avena genetic resources. p. 128-132. In G.J. Scoles and B.G. Rossnagel (ed.) Int. Oat Conf., 5th & Int. Barley Genet. Symp., 7th Vol. 1. Univ. of Saskatchewan Ext. Press, Saskatoon, SK, Canada.
- Li, C.D., B.G. Rossnagel, and G.J. Scoles. 2000. Tracing the phylogeny of the hexaploid oat Avena sativa with satellite DNAs. Crop Sci. 40:1755-1763.
- Linares, C., E. Ferrer, and A. Fominaya. 1998. Discrimination of the closely related A and D genomes of the hexaploid oat Avena sativa L. Proc. Natl. Acad. Sci. USA 95:12450-12455.
- Loskutov, I.G. 2001. Interspecific crosses in Avena L. genera. Russ. J. Genet. 37:467-475.
- Loskutov, I.G. 2008. On evolutionary pathways of Avena species. Genet. Resour. Crop Evol. 55:211-220.
- Luby, J.J., and D.D. Stuthman. 1983. Evaluation of *Avena sativa L./A. fatua L.* progenies for agronomic and grain quality characters. Crop Sci. 23:1047-1052.
- Moore, G., K.M. Devos, Z. Wang, and M.D. Gale. 1995. Cereal genome evolution: grasses line up and form a circle. Curr Biol 5:737-739.
- Morikawa, T. 1975. Identification of the 21 monosomic lines in *Avena byzantina* c. Koch cv. 'Kanota'. Theor Appl Genet 70:271-278.
- Morikawa, T., and M. Nishihara. 2009. Genomic and polyploid evolution in genus Avena as revealed by RFLPs of repeated DNA sequences. Genes Genet. Syst. 84:199-208.
- Murphy, J.P., and L.A. Hoffman. 1992. The origin, history and production of oat. p. 1-29. *In*H. G. Marshall and M. E. Sorrells (ed) Oat science and technology. American Society of Agronomy and Crop Science Society of America, Madison, Wisconsin.
- Nikoloudakis, N., and A. Katsiotis. 2008. The origin of the C-genome and cytoplasm of Avena polyploids. Theor. Appl. Genet. 117:273-281.
- Nikoloudakis, N., G. Skaracis, and A. Katsiotis. 2008. Evolutionary insights inferred by molecular analysis of the ITS1-5.8S-ITS2 and IGS Avena sp. sequences. Mol. Phylogenetics Evol. 46:102-115.
- O'Donoughue, L.S., Z. Wang, M. Roder, B. Kneen, M. Leggett, M.E. Sorrells, and S.D. Tanksley. 1992. An RFLP-based linkage map of oats on a cross between two diploid taxa (Avena atlantica x A. hirtula). Genome 35:765-771.
- O'Donoughue L.S., M.E. Sorrells, S.D. Tanksley, E. Autrique, A. Van Deynze, S.F. Kianian, R.L. Phillips, B. Wu, H.W. Rines, P.J. Rayapati, M. Lee, G.A. Penner, G. Fedak, S.J.

Molnar, D. Hoffman, and C.A. Salas. 1995. A molecular linkage map of cultivated oat. Genome 38:368-380.

- Oliver, R.E., N.A. Tinker, G.R. Lazo, S. Chao, E.N. Jellen, M.L. Carson, H.W. Rines, D.E. Obert, J.D. Lutz, I. Shackelford, A.B. Korol, C.P. Wight, K.M. Gardner, J. Hattori, A.D. Beattie, A. Bjornstad, J.M. Bonman, J.L. Jannink, M.E. Sorrells, G.L. Brown-Guedira, J.W.M. Fetch, S.A. Harrison, C.J. Howarth, A. Ibrahim, F.L. Kolb, M.S. McMullen, J.P. Murphy, H.W. Ohm, B.G. Rossnagel, W. Yan, K.J. Miclaus, J. Hiller, P.J. Maughan, R.R.R. Hulse, J.M. Anderson, E. Islamovic, and E.W. Jackson. 2013. SNP discovery and chromosome anchoring provide the first physically-anchored hexaploid oat map and reveal synteny with model species. PLoS ONE 8:e58068. doi:10.1371/journal.pone.0058068.
- Pal, N., J.S. Sandhu, L.L. Domier, and F.L. Kolb. 2002. Development and characterization of microsatellite and RFLP-derived PCR markers in oat. Crop Sci. 42:912-918.
- Peterson, D.M. 1992. Composition and nutritional characteristics of oat grain products. p. 265-292. *In* H. G. Marshall and M. E. Sorrells (ed) Oat science and technology. American Society of Agronomy and Crop Science Society of America. Madison, Wisconsin.
- Peterson, D.M., L.E. Schrader, and V.L. Youngs. 1974. Elemental composition of developing oat plants. Crop Sci. 14:735-739.
- Peterson, D.M., D.M. Wesenberg, D.E. Burrup, and C.A. Erickson. 2005. Relationships among agronomic traits and grain composition in oat genotypes grown in different environments. Crop Sci. 45:1249–1255.
- Portyanko V.A., D.L. Hoffman, M. Lee, J.B. Holland. 2001. A linkage map of hexaploid oat based on grass anchor DNA clones and its relationship to other oat maps. Genome 44:249-265.
- Rajhathy, T., and H. Thomas. 1974. Cytogenetics of oats (Avena L.). Misc. Publ. Genet. Soc. Can. 2:1-90.
- Ransom, J., M.S. McMullen, and D. Meyer. 2007. Oat production in North Dakota. Bull. A-891. North Dakota State Univ., Ext. Serv., Fargo.
- Rayapati, P.J., J.W. Gregory, M. Lee, and R.P. Wise. 1994. A linkage map of diploid Avena based on RFLP loci and a locus conferring resistance to nine isolates of *Puccinia coronata* var. avenae. Theor Appl Genet 89:831-837.
- Redaelli R., D. Sgouletta, G. Scalfati, E. De Stefanis, and P. Cacciatori. 2009. Naked oats for improving human nutrition : Genetic and agronomic variability of grain bioactive components. Crop Sci. 49:1431-1437.

- Rines, H.W., S.J. Molnar, N.A. Tinker, and R.L. Phillips. 2006. Oat. p 211-242. In C. Kole (ed.) Genome mapping and molecular breeding in plants: cereals and millets. Springer publishers. NY.
- Ronald, P.S., P.D. Brown, G.A. Penner, A. Boule-Babel, and S. Kibite. 1999. Heritability of hull percentage in oat. Crop Sci. 28:618-623
- Schrickel, D.J. 1986. Oats production, value and use. p 1-14. *In* F. H. Webster (ed.) Oats: chemistry and technology. American Association of cereal chemist publishers. St Paul MN.
- Tenaillon, M.I., M.C. Sawkins, A.D. Long, R.L. Gaut, J.F. Doebley, and B.S. Gaut. 2001. Patterns of DNA sequence polymorphism along chromosome 1 of maize (*Zea mays ssp. mays L.*). PNAS 98 :9161-9166.
- Tiwari, M.R., S. Khanal, B. Shrestha, and R.K. Jha. 2006. Nutritional variation of different feed ingredients and compound feed found in different parts of Nepal. Nepal Agric. Res. J. 7:75-81.
- Tinker, N.A., A. Kilian, C.P. Wight, K. Heller-Uszynska, P. Wenzl, H.W. Rines, A.
 Bjornstad, C.J. Howarth, J.L. Jannink, J.M. Anderson, B.G. Rossnagel, D.D. Stuthman, M.E. Sorrells, E.W. Jackson, S. Tuvesson, F.L. Kolb, O. Olsson, L.C. Federizzi, M.L.
 Carson, H.H. Ohm, S.J. Molnar, G.J. Scoles, P.E. Eckstein, J.M. Bonman, A. Ceplitis, and T. Langdon. 2009. New DArT markers for oat provide enhanced map coverage and global germplasm characterization. BMC Genomics 10:39. doi:10.1186/1147-2164-10-39.
- Todorovska, E., N. Abumhadi, K. Kamenarova, D. Zheleva, A. Kostova, N. Christov, N. Alexandrova, J.M. Jacquemin, H. Anzai, C. Nakamura, and A. Atanassov. 2005. Biotechnological approaches for cereal crops improvement part II: Use of molecular markers in cereal breeding. Biotechnol. and Biotechnol. Eq. 19:91-104.
- USDA National Agricultural Statistics Service. 2013. Quick stats. Available at http://www.nass.usda.gov/quickstats/ (verified 28 sept. 2013). USDA-NASS, Washington, DC.
- Van, K., E.Y. Hwang, M.Y. Kim, H.J. Park, S.H. Lee, and P.B. Cregan. 2005. Discovery of SNPs in soybean genotypes frequently used as the parents of mapping populations in the United States and Korea. Journal of Heredity 96:529-535.
- Wenzl P., J. Carling, D. Kudrna, D. Jaccoud, E. Huttner, A. Kleinhofs, and A. Kilian. 2004. Diversity arrays technology (DArT) for whole-genome profiling of barley. Proc. Natl. Acad. Sci. USA 101:9915-9920.
- Wesenberg, D.M., and H.L. Shands. 1973. Heritability of oat caryopsis percentage and other grain quality components. Crop Sci. 13:481-484.

- Youngs, V.L., and R.A. Forsberg. 1979. Protein-oil relationships in oats. Crop Sci. 19:798-802.
- Youngs, V.L., D.M. Peterson, and C.M. Brown. 1982. Oats. p 49-105. *In* Pomeranz Y. (ed) Advances in cereal science and technology, vol 5, Am. Assoc. Cereal Chem., St Paul, MN.
- Yu, G.X., and R.P. Wise. 2000. An anchored AFLP- and retrotransposon-based map of diploid Avena. Genome 43:736-749.
- Zhu, S., and H.F. Kaeppler. 2003. A genetic linkage map for hexaploid, cultivated oat (Avena sativa L.) based on intraspecific cross 'Ogle/MAM17-5'. Theor. Appl. Genet. 107:26-35.

CHAPTER 2. PHENOTYPIC AND GENOTYPIC CHARACTERIZATION OF TWO RECOMBINANT INBRED OAT POPULATIONS FOR GRAIN QUALITY

2.1. Abstract

The use of oats for human consumption is increasing every day due to the health benefits of oat products. Oat products reduce cholesterol and arthrosclerosis, decrease the risk of diabetes, and provide antioxidant protection and supply a number of important nutrients. With the objective to study relationships among factors affecting oat grain quality, two F_{4:6} Recombinant Inbred Lines (RIL) mapping populations developed by Single Seed Descent (SSD) have been used in this study. The first population (05021) consisted in 97 RILs derived from the cross 'ND030299 x ND991151' and the second population (05026) with 93 RILs was derived from the cross 'ND030299 x ND961161'. The 190 RILs from the two populations with the three parents and three check cultivars (HIFI, HIFI-9, and YOUNGS) were evaluated in a square lattice design (14x14) with two replicates in 2008 and 2009 at two North Dakota locations. Data were recorded on a plot basis for all experiments on the following agronomic traits: grain yield, test weight, 1000 kernel weight, thin kernels, heading date, and plant height. Chemical analysis and grain physical analysis were performed for β-glucan, oil content, groat percentage and dehulling efficiency. Broad-sense heritability estimates combined across environments were calculated for each trait. Using covariance parameters, genotypic and phenotypic correlations between traits were also estimated. After looking for normality and homogeneity of variance, a combined ANOVA was performed, using the MIXED procedure from SAS, where environments were considered random and genotypes were considered fixed. Combined across years, grain yield varied from 942 kg/ha to 7744 kg/ha (population 05021), and from 824 kg/ha to 7741 kg/ha

(population 05026). Test weight ranged from 398 kg/m³ to 514 kg/m³ for genotypes of the population 05021, and from 443 kg/m³ to 524 kg/m³ for genotypes of the population 05026. In general, plants were taller in population 05021 ranging from 93 to 125 cm than in population 05026 (89 to 111 cm). Population 05026 had more thin kernels (11%) than population 05021 (7%). Most of the variance observed in both populations was associated with the genotype main effect. Grain yield was positively correlated with test weight, thin kernels, plant height, β -glucan content, and associated negatively with 1000 kernel weight. The wide variation observed is an indication of the capacity to improve both populations with respect to grain quality.

2.2. Introduction

Oat is an important cereal crop and has value either as food for humans and feed for livestock (Zhu and Kaeppler, 2003; Peterson et al., 2005; Ozbas et al., 2009). The quality of oat grains can be, in many aspects, superior to that of other cereals. The beneficial effects and the positive impacts of the total dietary fiber on some of the risk factors of cardiovascular diseases have stimulated great interest in oat (Redaelli et al., 2009). The improvement of oat grain quality for human consumption and animal feed has been a primary objective for oat breeders. Important oat agronomic traits include grain yield, plant height, resistance to lodging, heading date; of grain physical traits, the most important are test weight, 1000 kernel weight, groat percentage; and grain chemical composition traits: protein, oil, β -glucan (Peterson et al., 2005). Test weight is an index of grain quality for farmers and millers, and high test weight is generally associated with high grain quality (Forsberg and Reeves, 1992; Klein et al., 1993; Doehlert and McMullen, 2008). Frey and Wiggans, 1956 reported that high test weight is related with high percentage of germination, good seedling stand, and high milling yield. Standard test weight for modern oat cultivars with values ranging between 463 to 515 kg/m³ has been reported by Forsberg and

Reeves, 1992. Test weight can be influenced by several factors including kernel size and shape, groat density, groat percentage, presence of awns and diseases (Doehlert et al., 2001). Groat percentage, also known as groat proportion and caryopsis percentage, represents the economic yield that a given lot of oat grain can produce (Doehlert et al., 1999). One of the major objectives of oat breeders is the reduction of plant height in order to minimize substantially lodging problems that affect grain quality and quantity (Milach et al., 1997). High oil content is advantageous for animal feeding due of its high content of energy, but in food applications high oil content is deleterious because of its potential to produce enzymatic rancidity reactions that give rise to bitter, grassy or other undesirable flavors (Kianian et al., 1999; Doehlert et al., 2001). The presence of a high concentration of β -glucan in the cell walls of the groat (Peterson, 1992) lowers the serum cholesterol levels of blood, balances the glucose and insulin content of serum after meals and reduces the risk of cardiovascular diseases (Anderson and Chen, 1986). Genotypic and phenotypic correlations had been reported between test weight and groat percentage (Doehlert at al., 2004; Peterson et al., 2005), oil and groat percentage (Doehlert et al., 2001; Lyrene and Shands, 1975), yield and test weight (Holland and Munkvold, 2001), heading date and test weight (Pixley and Frey, 1992), and 1000 kernel weight and test weight (Holland and Munkvold, 2001). Genotypic and phenotypic correlations among traits can help in the improvement of selection (direct or indirect), and in the calculation of multiple trait selection indices (Falconer and Mackay, 1996). The objective of this chapter is to evaluate, through a twoyear multi-environment field experiment, genotypic and phenotypic relationships among factors such as grain yield, test weight, kernel weight, heading date, plant height, thin kernels, oil, and β glucan affecting oat grain quality.

2.3. Materials and methods

Two F_{4.6} Recombinant Inbred Lines (RIL) mapping populations developed by Single Seed Descent (SSD) have been used in this study. The first population (05021) consisted in 97 RILs derived from the cross 'ND030299' x 'ND991151' and the second population (05026) with 93 RILs is derived from the cross 'ND030299' x 'Souris'. The two populations had a common parent (ND030299) with the objective to provide a degree of biological replication. 'ND030299' and 'ND991151" are two experimental lines from the oat breeding program of North Dakota State University (NDSU) with high content of β -glucan (6%) and low content of β -glucan (4.3%), respectively. 'Souris' is an oat line released in 2006 by the oat breeding program of NDSU with intermediate content of β -glucan (5.2%). Souris is stable for stem rust resistance and crown rust resistance because it possesses the crown rust resistance gene Pc-91 and the stem rust race NA27 resistance conferred by gene Pg-13. 'ND030299' was derived from a cross between 'HIFI' and 'IAN979-5-1-22'. The pedigree of 'HIFI' included 'ND90141' and 'ND900118'. 'ND90141' was derived from 'R801441', a synthetic hexaploid derived from an A. magna/A. longiglumis hybrid, and 'RL3038'; a breeding line received from R. McKenzie (Winnipeg, Canada) and possessed genes Pc-38, Pc-39, Pg-2, and Pg-13. 'Souris' is a 'HIFI' sister selection. 'ND991151' was derived from a cross between 'Youngs' and 'ND931318' (McMullen, communication personal).

The 190 RILs from the two populations with the three parents and three check cultivars (HIFI, HIFI-9, and YOUNGS) were evaluated in a square lattice design (14x14) with two replicates in 2008 and 2009 at two North Dakota locations: Fargo and Casselton. The randomization within blocks was done by population with the objective that each population stays together in the field. Fargo is located at 46°52' latitude north, and 96°54' longitude west
with an elevation of 275 m above sea level. The average precipitation during the growing season (May to October) is 348 mm and the mean air temperature is 17.9°C. The soil type where the experiments have been conducted is a silty clay (fine smectitic frigid Typic Epiaquert). Casselton is located at a latitude of 46°55' N and a longitude of 97°13' W. The average precipitation in the growing season is 364 mm and the mean temperature is 17.6°C. The soil type is a silty clay loam (fine silty mixed frigid Typic Haploquolls). In 2008, experiments at Fargo and at Casselton were planted on April 22 and April 29, respectively whereas in 2009 experiments were planted on May 11 and May 20, respectively.

A seeding rate of 2.47 x 10⁶ kernels/ha was used for all experiments. Experimental units consisted of four rows spaced 0.3 m apart and 2.4 m long. Data were recorded on a plot basis for all experiments on the following agronomic traits: grain yield (kg/ha), test weight (kg/m³), 1000 kernel weight (g), thin kernels (%), heading date (days after 31st may), and plant height (cm). Heading date, corresponding to the days after May 31st on which the first nodes on 50% of the plants in the plot had emerged completely above the flag leaf, and plant height at maturity, from ground level to the tips of the panicles, were measured on each plot. The two center rows in each plot were harvested with a two-row binder and the plants were bundled together and dried at ambient temperature for 1 week, after which the plants were threshed with a plot thresher and grain yield was measured. Test weight was calculated by weighing a known volume of whole oat and converting to a Winchester bushel. Thin kernels also known as shrunken or damaged kernels are the weight of all matter that passes through a 0.064" x 3/8' oblong-hole sieve after sieving. Percentage of thin kernels was calculated as follows for each plot:

Thin kernels (%) =
$$\frac{\text{thin kernels } (kg/ha)x100}{\text{Grain yield } (kg/ha)}$$

After removing foreign material and broken kernels, 300 kernels were counted and weighted to obtain 1000 kernel weight as follow (measured only in 2008):

$$1000 \ kernel \ weight = \frac{300 \ kernels \ weight \ (g)x1000}{300}$$

Chemical analysis and grain physical analysis were performed for each plot, only in 2008 planting season, for β -glucan content, oil content, groat percentage, and dehulling efficiency. Before performing chemical analysis the oat samples were treated in a vegetable steamer for 20 minutes in order to inactivate enzymes. After, the sample was dehulled with a Codema Laboratory Oat Huller (Codema Inc., Eden Prairie, MN) and milled in a Retsch Model ZM-1 Centrifugal Mill with a 0.5 mm collar screen (Brinkmann Instruments, West-Bury, NY). The milled samples were dried in a convection oven at 130°C for 2h to eliminate interference of the water with the oil signal. The oil analysis was performed using an Oxford 4000 NMR (Abingdon, England). β -glucan concentration was determined by the method of McCleary and Glennie-Holmes, 1985. Groat percentage and dehulling efficiency were assessed using the impact dehuller (Ganssmann and Vorwerck, 1995).

Broad-sense heritability estimates were calculated for the agronomic traits as:

$$h^2 = \frac{\sigma_G^2}{\sigma_G^2 + \sigma_{GE}^2/e + \sigma_e^2/er}$$

Where, h^2 = heritability, σ_G^2 = Genotypic variance, σ_{GE}^2 = Genotype x environment interaction, σ_e^2 = error variance, e= number of environments, and r = replications.

Using genotypic variances and covariance parameters estimates from SAS PROC MIXED, genotypic and phenotypic correlations between traits were estimated as:

$$r_{gij} = \frac{\sigma_{Gij}}{\sigma_{Gi}\sigma_{Gj}}$$

$$r_{pij} = \frac{\sigma_{Pij}}{\sigma_{Pi}\sigma_{Pi}}$$

Where, r_{gij} = genotypic correlation between traits i and j, σ_{Gij} = genotypic covariance between traits i and j, σ_{Gi} = genotypic standard deviation for trait i, σ_{Gj} = genotypic standard deviation for trait j.

 r_{pij} = phenotypic correlation between traits i and j, σ_{Pij} = phenotypic covariance between traits i and j, σ_{Pi} = phenotypic standard deviation for trait i, σ_{Pj} = phenotypic standard deviation for trait j.

Genotypic and phenotypic coefficient of variation were calculated according to Singh and Chaudhary, 1977:

$$GCV = \frac{\sqrt{\sigma_G^2}}{\bar{X}} x100$$
$$PCV = \frac{\sqrt{\sigma_P^2}}{\bar{X}} x100$$

Where, GCV=genotypic coefficient of variation, PCV=phenotypic coefficient of variation, σ^2 G=genotypic variance, σ^2 P=phenotypic variance, \bar{X} = general mean

Transgressive segregation was also estimated for all traits. According to Vega and Frey, 1980 a transgressive genotype had to exceed the parental mean by one "least significant difference (LSD)". Due to the lack of repetitions, Pearson correlations were calculated among biochemical traits, and between biochemical and agronomic traits.

Each environment (a combination of year and location) was analyzed using the MIXED procedure of SAS (SAS Institute, Cary, NC, 2009). Normality of distribution of the traits analyzed was estimated using Shapiro-Wilk test. Homogeneity of variances among environments

was determined by the ratio of the largest to the smallest variances followed by Levene's test and Welch's ANOVA. Except for thin kernels, variances were found to be normal and homogeneous for all variables. In order to perform analysis of variance (ANOVA), the variable thin kernels was log transformed to normalize the variance. After looking for normality and homogeneity of variance, a combined ANOVA by year for all environments was performed, using the MIXED procedure from SAS (SAS Institute, Cary, NC, 2009), where environments were considered random and genotypes were considered fixed.

For the purposes of estimating variance and covariance components, heritability, genotypic and phenotypic correlations, parental and check cultivars were removed from the data set. Variance and covariance parameters were obtained using the COVTEST and ASYCOV options from PROC MIXED of SAS, assuming environments and genotypes to be random. Heritability estimates, genotypic and phenotypic correlations were calculated by multivariate restricted maximum likelihood (REML) implemented in PROC MIXED of SAS as described by Holland et al., 2003 and Holland, 2006. Approximate standard errors for heritability, genotypic and phenotypic correlations were calculated using SAS PROC IML.

2.4. Results

Analysis of variance showed significant (p<0.01) genotype-by-environment interaction for all traits except for 1000 kernel weight. Genotype main effect was highly significant (p<0.01) for all traits. Environment main effect was significant for all traits (p<0.05) but not for grain yield (Table 2.1). 'ND991151' and 'ND030299' showed statistically the same grain yield. 'ND991151' had higher test weight, 1000 kernel weight, plant height, and less percentage of thin kernels than 'ND030299'. 'ND030299' headed later than 'ND991151' (Table 2.3). For population 05026, 'Souris' yielded significantly higher than 'ND030299'. Both parents showed similar 1000 kernel weight, test weight, plant height, and percentage of broken kernels. 'ND030299' headed later than 'Souris' (Table 2.3).

Combined across years and locations, grain yield varied from 942 kg/ha to 7744 kg/ha with an average of 6144 kg/ha (population 05021), and from 824 kg/ha to 7741 kg/ha with an average of 6664 kg/ha (population 05026). Test weight ranged from 398 kg/m³ to 514 kg/m³ (average 488 kg/m³) for genotypes of the population 05021, and from 443 kg/m³ to 524 kg/m³ for genotypes of the population 05026 (average 495 kg/m³). For population 05021, 1000 kernel weight averaged 32 g and ranged from 23 to 40 g, whereas for population 05026, 1000 kernel weight ranged from 25 to 40 g (average 30 g). Heading date ranged from 31 to 41 days after 31st May (population 05021) and from 33 to 45 days after 31st May (population 05026). In general, plant height was taller in population 05021, ranging from 93 to 125 cm (average 109 cm), than in population 05026, ranging from 89 to 111 cm (average 103 cm). Population 05026 had greater percentage of thins kernels, average 11%, than population 05021, average 7% (Table 2.3).

Transgressive genotypes at least in one direction were observed for all traits. In the population 05021, more transgressive segregates were observed for low grain yield, only one genotype yielded significantly more than the high-yielding parent. Transgressive segregation for low 1000 kernel weight, low test weight, and high percentage of thins kernels was observed. Transgressive segregation for heading date occurred more for lateness than for earliness. For plant height, transgressive segregates for tall and short plants occurred at the same frequency (Table 2.3). For the population 05026, no transgressive segregates were observed for high grain yield. Transgressive segregation for high 1000 kernel weight, short plants, high test weight, early

heading, and high percentage of thin kernels were observed (Table 2.3). In conclusion, more transgressive segregants were observed for high 1000 kernel weight, high test weight, earliness, and tall plants in the population 05026. At the other end, more transgressive segregants were observed for high grain yield in the population 05021.

Most of the variance observed in both populations was associated with the genotype main effect, except for heading date that had a large variance associated with the environment main effect due mainly to the difference in planting date between the two locations. The variance associated with the genotype-by-environment interaction was less than 10% of the total variance for all traits (Table 2.2). The variances associated with environment main effect and genotype-by-environment interaction were negligible for 1000 kernel weight in the population 05021 (Table 2.2).

Broad-sense heritability estimates were high for all traits, ranging from 0.93 for grain yield to 0.98 for heading date and percentage of thin kernels. Generally, heritability estimates were slightly higher in population 05021 than in population 05026 for grain yield, test weight, plant height, and percentage of broken kernels. Heritability estimates were statistically similar for heading date for both populations (Table 2.5).

Phenotypic and genotypic coefficients of variation (GCV and PCV) were similar for both populations. For population 05021, GCV and PCV ranged from 3.5% for test weight to 39.5 and 40%, respectively for broken kernels, whereas for population 05026, GCV and PCV ranged from 2.9 and 3.0% for test weight to 20.0 and 20.5% for thin kernels. The highest GCV and PCV were observed for thin kernels followed by grain yield, heading date, plant height, and finally test weight (Table 2.2).

		Grain yield		10	00 kerno weight	el		Fest weight		Н	leading date	•		Height		Tł	nin kernel	s
SOV	Df	MS		Df	MS		Df	MS		Df	MS		Df	MS		Df	MS	
Environment	3	10,482,253	ns	1	128.5	*	3	35,460.0	*	3	10,314.0	*	3	10,793.0	*	3	29.306	**
Rep(environment)	4	9,066,216		2	2.3		4	3,319.7		4	10.1		4	854.2		4	0.947	
Block(rep*environment)	104	1,454,209		52	9.6		104	163.6		104	1.6		104	77.4		104	0.252	
Genotypes	198	3,569,407	**	194	31.3	**	196	1,708.4	**	198	45.4	**	198	313.5	**	198	3.071	**
Genotypes*environment	580	313,646	**	193	1.6	ns	577	111.0	**	582	1.4	**	581	15.5	**	576	0.094	**
Error	644	168,514		327	1.4		653	60.4		671	0.7		659	9.8		651	0.038	
Df=degrees of freedo	m	Μ	S=n	nean s	quare	:	*signi	ficant at 5	5%	**	significan	t at	1%					

Table 2.1. Combined Analysis of Variance (ANOVA) for agronomic traits of two oat populations grown at two ND locations (Fargo, Casselton) and two years (2008, 2009).

Covariance	Grain	1000 kernel	Test	Heading	Heig	Broken
parameter	yield	weight	weight	date	ht	kernels
			Population	05021		
Environment	20,011	0.000	122.3	27.11	35.02	0.072
Rep(environment)	35,136	0.142	25.4	0.12	3.53	0.003
Block(rep*env)	215,764	1.224	9.9	0.17	12.28	0.023
Genotypes	795,093	11.455	285.8	5.26	45.96	0.559
Genotypes*env	102,278	0.000	18.1	0.36	4.14	0.035
Error	172,198	1.562	66.4	0.73	8.70	0.044
CV	6.8	4.0	1.7	2.2	2.7	11.1
GCV	14.5	10.7	3.5	5.8	6.2	39.5
PCV	14.9	10.9	3.5	5.9	6.4	40.0
			Population	05026		
Environment	0	0.950	51.3	26.38	17.82	0.078
Rep(environment)	48,616	0.339	10.1	0.01	4.52	0.020
Block(rep*env)	138,426	0.188	9.2	0.04	4.58	0.011
Genotypes	451,084	4.275	207.7	6.98	26.71	0.224
Genotypes*env	63,255	0.324	30.3	0.36	2.77	0.024
Error	159,417	1.113	58.0	0.63	10.25	0.033
CV	6.0	3.5	1.5	2.0	3.1	7.7
GCV	10.1	6.9	2.9	6.8	5.0	20.0
PCV	10.5	7.2	3.0	6.9	5.2	20.5
			Combin	ed		
Environment	3,272	0.300	83.9	26.75	26.18	0.075
Rep(environment)	32,469	0.000	16.0	0.04	3.47	0.002
Block(rep*env)	192,662	1.228	13.6	0.15	10.01	0.029
Genotypes	689,849	8.656	248.4	6.19	42.79	0.449
Genotypes*env	82,445	0.148	26.4	0.37	3.32	0.030
Error	166,403	1.324	62.3	0.67	9.56	0.038
CV	6.4	3.7	1.6	2.1	2.9	9.2
GCV	13.0	9.6	3.2	6.4	6.2	31.6
PCV	13.4	9.8	3.3	6.5	6.3	32.0

Table 2.2. Estimates of variance component for two oat populations grown at two ND locations (Fargo, Casselton) and two years (2008, 2009).

CV=coefficient of variation, GCV=genotypic coefficient of variation, PCV=phenotypic coefficient of variation

The association among agronomic traits is shown by the genotypic and phenotypic correlations (Table 2.5). Genotypic and phenotypic correlation coefficients were very similar for both populations. Grain yield was genotypically and phenotypically correlated positively with

test weight and thin kernels, and associated negatively with 1000 kernel weight. A significant positive phenotypic correlation was observed between grain yield and plant height, but genotypically that correlation was not significant. A highly significant negative relationship was observed between 1000 kernel weight and thin kernels. Test weight was negatively correlated to heading date.

Relationships between biochemical, grain physical and agronomic traits are shown in Table 2.6 and Table 2.7. Groat percentage was positively correlated (r=0.57, p<0.01) to dehulling efficiency. Dehulling efficiency was negatively correlated (r=-0.20, p<0.05) with oil content, and a positive correlation (r=0.38, p<0.01) was observed between oil and β -glucan content. Grain yield was positively correlated to dehulling efficiency (r=0.40, p<0.01), β -glucan content (r=0.23, p<0.05), and lodging (r=0.25, p<0.05). Percentage of thin kernels was correlated positively with β -glucan (r=0.21, p<0.05) and oil content (r=0.38, p<0.01), whereas 1000 kernel weight was negatively correlated with both as expected. A positive relationship was observed between test weight and groat percentage (r=0.26, p<0.01), and dehulling efficiency (r=0.40, p<0.01), and plant height positively correlated to lodging (r=0.26, p<0.01) as expected.

2.5. Discussion

2.5.1. ANOVA, means and components of variance

The two populations used in this study consisted of genotypes that are adapted to the conditions of eastern North Dakota, which are characterized by warm and dry summers. For all the traits studied, genotype main effect was highly significant and showed considerable amount of variation (Table 2.1).

Population	Par	rents		Populatio	Population statistics				Transgressive segregation			
-	ND991151			-				-				
	Souris	ND030299	Mean	SD	Min	Max	$X \le P_1$ -	SD	$X > P_2$	+SD		
							%	n	%	n		
				Grain yie	eld (kg/ha)							
05021	5998.1 a	6476.4 a	6143.6	953.0	941.9	7743.8	7.2	7	1.0	1		
05026	7439.4 a	6476.4 b	6664.1	740.2	823.8	7741.4	2.2	2	0.0	0		
				1000 kerne	el weight (g)							
05021	37.4 a	30.1 b	31.6	3.5	23.0	39.6	7.2	7	0.0	0		
05026	29.4 a	30.1 a	30.0	2.3	24.6	40.0	4.3	4	14.0	13		
				Test weig	(kg/m^3)							
05021	521.2 a	488.0 b	488.2	17.8	398.5	514.1	13.4	13	0.0	0		
05026	500.3 a	488.0 a	495.0	14.7	442.5	523.9	5.4	5	7.6	7		
				Heading of	date (June)							
05021	37 b	41 a	39.0	2.3	33.7	44.5	1.0	1	6.2	6		
05026	37 b	41 a	38.8	2.7	33.4	45.2	1.1	1	2.2	2		
				Heigh	nt (cm)							
05021	116.8 a	104.2 b	108.7	7.0	93.0	125.0	4.1	4	2.1	2		
05026	99.7 a	104.2 a	103.3	5.4	89.3	110.7	6.5	6	11.9	11		
				Broken k	ernels (%)							
05021	2.1 b	8.3 a	6.6	0.77	1.4	55.4	0.0	0	12.4	12		
05026	8.8 a	8.3 a	10.6	0.48	3.4	33.9	7.6	7	29.2	27		

Table 2.3. Overall means and descriptive statistics of agronomic traits for oat population 05021 (ND991151 x ND030299) and population 05026 (Souris x ND030299) grown at two locations (Fargo, Casselton) and two years (2008, 2009).

Table 2.4. Genotypic and phenotypic correlations of two oat populations combined across two ND locations (Fargo, Casselton) and two years (2008, 2009) – genotypic correlation in the upper diagonal, phenotypic correlation in the lower diagonal, standard errors between parenthesis.

	Grain yield	Kernel weight	Test weight	Heading date	Height	Broken kernels
			Рори	lation 05021		
Grain yield	-	0.01 (0.11)	0.47 (0.10)	0.04 (0.11)	0.24 (0.11)	0.33 (0.10)
Kernel weight	0.00 (0.08)	-	0.16 (0.11)	-0.18 (0.10)	0.07 (0.11)	-0.84 (0.03)
Test weight	0.38 (0.07)	0.23 (0.08)	-	-0.34 (0.10)	0.07 (0.11)	-0.04 (0.11)
Heading date	0.05 (0.08)	-0.15 (0.09)	-0.30 (0.07)	-	-0.01 (0.11)	0.10 (0.10)
Height	0.26 (0.07)	0.01 (0.08)	0.05 (0.08)	0.04 (0.08)	-	-0.05 (0.11)
Broken kernels	0.19 (0.08)	-0.79 (0.03)	-0.11 (0.08)	0.10 (0.09)	-0.03 (0.08)	-
			Рори	lation 05026		
Grain yield	-	-0.30 (0.11)	0.07 (0.19)	0.08 (0.11)	0.22 (0.11)	0.22 (0.11)
Kernel weight	-0.14 (0.07)	-	0.20 (0.12)	-0.09 (0.11)	0.14 (0.12)	-0.76 (0.05)
Test weight	0.06 (0.12)	0.23 (0.08)	-	-0.45 (0.09)	-0.11 (0.11)	-0.26 (0.11)
Heading date	0.06 (0.08)	-0.10 (0.09)	-0.36 (0.07)	-	0.11 (0.11)	0.03 (0.11)
Height	0.27 (0.07)	0.11 (0.08)	-0.05 (0.07)	0.11 (0.08)	-	0.07 (0.11)
Broken kernels	0.12 (0.08)	-0.68 (0.04)	-0.26 (0.07)	0.04 (0.09)	0.02 (0.08)	-
			Combined	across populations		
Grain yield	-	-0.17 (0.08)	0.43 (0.08)	-0.01(0.08)	0.10 (0.08)	0.37 (0.07)
Kernel weight	-0.14 (0.05)	-	0.09 (0.08)	-0.09 (0.08)	0.20 (0.08)	-0.84 (0.02)
Test weight	0.33 (0.06)	0.17 (0.06)	-	-0.42 (0.06)	-0.04 (0.08)	0.03 (0.08)
Heading date	-0.01 (0.08)	-0.10 (0.06)	-0.34 (0.05)	-	0.07 (0.07)	0.02 (0.07)
Height	0.19 (0.05)	0.08 (0.06)	-0.02 (0.05)	0.09 (0.06)	-	-0.15 (0.07)
Broken kernels	0.23 (0.05)	-0.77 (0.02)	-0.10 (0.06)	0.03 (0.06)	-0.12 (0.06)	-

Cells in color mean significant at 5% and 1%

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Population 05021 05026 Combined Traits H^2 H^2 H^2 SE SE SE Grain yield (kg/ha) 0.94 0.01 0.93 0.01 0.94 0.01 Test weight (kg/m^3) 0.96 0.93 0.01 0.95 0.01 0.02 Heading date (June) 0.97 0.00 0.97 0.00 0.01 0.98 Height (cm) 0.96 0.01 0.93 0.01 0.95 0.01 Broken kernels (%) 0.98 0.00 0.96 0.01 0.97 0.00

Table 2.5. Broad-sense heritability estimates and standard error of two oat populations combined across 2 ND locations (Fargo, Casselton) and 2 years (2008, 2009).

H²=heritability estimate SE=standard error

Table 2.6. Pearson correlations between biochemical traits and grain physical traits for oat population 05021 grown in 2008.

Traits	Groat %	Dehulling efficiency	β-glucan	Oil	Lodge
Groat		0.57**	0.007	-0.13	-0.003
Dehulling			-0.06	-0.20*	0.18
β-glucan				0.38**	0.10
Oil					-0.09
*	· · · ·	1.10/			

*significant at 5% **significant at 1%

Table 2.7. Pearson correlations between biochemical, agronomic, and grain physical traits for oat population 05021 grown in 2008.

Traits	Groat %	Dehulling efficiency	β-glucan	Oil	Lodge
Grain yield	0.17	0.40**	0.23*	0.04	0.25*
Broken kernels	0.07	0.003	0.21*	0.38**	0.02
Kernel weight	0.06	0.12	-0.22*	-0.35**	-0.05
Test weight	0.26**	0.40**	-0.07	-0.13	0.16
Heading	-0.38**	-0.12	0.08	0.12	-0.06
Height	-0.17	-0.04	-0.14	-0.05	0.26**

*significant at 5% **significant at 1%

Such wide variation is an indication of the capacity to improve both populations with respect to grain quality. Genotype x environment interaction was also observed for all traits measured, except 1000 kernel weight, although the magnitude of the interactions mean squares were relatively small in comparison to environment main effect and genotype main effect. The most likely important factor contributing to the significant $G \times E$ interaction for grain yield, test

weight, height, and thin kernels can be attributed to the differences in weather conditions especially rainfall. For the 2008 growing season, a total of 671.2 mm of rainfall was reported, whereas during the 2009 growing season only 392.3 mm of rainfall was registered. Similar behavior was reported by Doehlert et al., 2001 for grain yield, and test weight, only the factor contributing to the significant GxE interaction was due to the differential level of crown rust infection among the cultivars. The absence of significant genotype-by-environment interaction for 1000 kernel weight suggests that this trait will remain constant over a range of environmental conditions, and selection for high 1000 kernel weight should be practical.

The magnitude of variance components in both populations suggested that all the traits evaluated were more strongly influenced by genotype than environment. Doehlert et al., 2001 reported that grain yield was more influenced by environment, whereas test weight was equally affected by environment and genotype.

Because the highest yielding parents (Souris and ND030299), with also high percentage of thin kernels were in the population 05026, the genotypes in that population had, on average, higher grain yield, higher test weight, and higher percentage of thin kernels than the genotypes in the population 05021. For the other traits, the genotypes in the population 05021 had higher 1000 kernel weight and plant height than the genotypes in the population 05026. Heading date was similar for genotypes in both populations (Table 2.3 and Table 2.4).

Lower yield than those reported in this experiment, ranging from 3140 kg/ha to 4110 kg/ha (average 3590 kg/ha) for 12 oat cultivars, and from 1510 kg/ha to 2750 kg/ha (average 2010 kg/ha) was reported by Doehlert et al., 2001, and Holland and Munkvold, 2001. In a study conducted at 3 locations in Idaho during 3 consecutive years, Peterson et al., 2005 reported grain yield ranging from 2900 to 8660 kg/ha with an average of 5264 kg/ha.

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Values of test weight reported for oat cultivars adapted to the conditions of North Dakota (ND) are very similar to those found in this study. Doehlert et al., 2001 found that the test weight of 12 genotypes grown at four locations in ND varied from 433 to 601 kg m⁻³ with a mean value of 490 kg m⁻³. In a study conducted at five locations in ND over 3 years and 10 oat cultivars, Doehlert et al., 2004 reported test weight across genotypes ranging from 418 to 517 kg m⁻³ with a mean value of 467 kg m⁻³. Doehlert et al., 2006, analyzed oat grain and groat size from 10 genotypes grown in 10 environments in ND and reported test weight values ranging from 413 to 529 kg m⁻³ with a mean value of 468 kg m⁻³. In another study, Doehlert and McMullen, 2008 reported values of test weight, from six oat cultivars grown at three locations in ND, ranging from 513 to 556 kg m⁻³. Higher test weight ranging from 517 to 585 kg/m³ was reported by Peterson et al., 2005. Lower test weight from 379 kg/m³ to 453 kg/m³ was reported by Holland and Munkvold, 2001. Redaelli et al., 2008 reported values for test weight grown in Italy ranging from 417 to 468 kg/m³.

Values of 1000 kernel weight between 20 and 29 g (average 24 g) were reported by Holland and Munkvold, 2001. Nersting et al., 2006 analyzing Nordic oat material comprising landraces and recent cultivars reported 1000 kernel weight for landraces ranging from 19 to 41 g (average 32 g), and for recent cultivars ranging from 24 to 46 g (average 34 g). Redaelli et al., 2008 found out that 1000 kernel weight for oat cultivated in Italy varied from 30 g to 35 g with an average of 30 g.

A good numbers of RILs, in the population 05026, were identified with superior performance for test weight and plant height that headed early with low percentage of thin kernels. Those transgressive genotypes suggested that those traits are quantitative in nature and

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the favorable additive alleles are brought by both parents as indicated by Ayele, 2011 and Vega and Frey, 1980.

2.5.2. Genotypic and phenotypic correlations among traits

According to Falconer and Mackay, 1996 genotypic correlation among traits is important in the determination of direction and magnitude of response to selection, and the relative efficiency of indirect selection. Genotypic correlation is also a key factor in the calculation of multiple trait selection indices. The absence of strong correlations among traits, except 1000 kernel weight and thin kernels, indicated that improvement of grain yield, test weight, heading date, and plant height should not be difficult in the two populations under study. Grain yield was positively correlated with test weight, plant height, β -glucan content, and dehulling efficiency. This suggests that when quality traits are improved higher yields can be achieved.

Many of the correlations found on this study had been reported previously. Doehlert et al., 2004 (r=0.723), Peterson et al., 2005 (r=0.552) and Doehlert and McMullen, 2008 (r=0.694) found that test weight was positively correlated with groat percentage. Forsberg and Reeves, 1992 concluded that high test weight and high groat percentage generally are associated with well-filled kernels. Peterson et al., 2005 reported a negative relationship between oil and kernel weight (r=-0.38, p<0.05). A negative correlation between oil and groat percentage was reported by Peterson et al., 2005 (r=-0.39, p<0.05), and Doehlert et al., 2001 (r=-0.59, p<0.01). We also found a negative correlation between oil and groat percentage but that correlation was not significant. Negative correlation between groat percentage and groat oil content (r=-0.387) has been reported by Lyrene and Shands, 1975 and Peterson et al., 2005 respectively.

Holland and Munkvold, 2001 reported a positive phenotypic and genotypic correlation between grain yield and test weight ($r_G=0.58\pm0.10$, $r_P=0.47\pm0.08$), and between 1000 kernel

weight and test weight ($r_G=0.38\pm0.09$, $r_P=0.34\pm0.09$). We also found a positive phenotypic and genotypic correlation between grain yield and test weight, but a negative genotypic correlation between 1000 kernel weight and test weight, significant only for population 05026. Doehlert and McMullen, 2000 showed that test weight and grain yield was negatively correlated (r=-0.46, p<0.01). Doehlert and McMullen, 2000 reported a positive correlation between β -glucan, oil (r=0.37, p<0.05), and grain yield (r=0.80, p<0.01).

Kernel weight was negatively correlated with oil content; this result is congruent with Holland et al., 2001 that indicated higher oil oats tended to have thinner kernels. Heading date tended to be negatively correlated with test weight (Pixley and Frey, 1992) and groat percentage (Tanhuanpaa et al., 2012).

2.5.3. Heritabilities

Heritability estimates are important parameters used to determine the role of heredity in the expression of a trait (Allard, 1960). When genotypic effects are substantial, and environmental effects are small, which indicates high heritability, effective selection can be achieved. The high heritabilities observed (Table 2.6) suggested that all the traits measured should respond well to selection on a family-mean basis. Holland and Munkvold, 2001 reported also high values of heritability in a F₆ oat population for grain yield (0.62 ± 0.06), 1000 kernel weight (0.89 ± 0.02), and test weight (0.80 ± 0.04). In a population of 132 oat recombinant inbred lines, heritability estimates reported by Holland, 2006 was high for heading date (0.92 ± 0.01), and moderate for plant height (0.54 ± 0.07). Pixley and Frey, 1992 in 13 oat crosses evaluated at F_{2:3} generation, reported moderate to high heritabilities for grain yield (0.60 to 0.82), test weight (0.63 to 0.91), kernel weight (0.77 to 0.95), plant height (0.61 to 0.87), and heading date (0.57 to 0.95). The lower heritabilities observed on a plot-basis (data not shown) indicated the importance of replication and multiple environment testing to evaluate grain yield, 1000 kernel weight, test

weight, heading date, and plant height.

2.6. References

Allard, R.W. 1960. Principles of plant breeding. John Wiley and Sons, Inc. NY.

- Anderson, J.W., and W.L. Chen. 1986. Cholesterol-lowering properties of oat products. p 309-333. *In* F.W. Webster (ed.). Oats: chemistry and technology. Am. Assoc. Cereal Chem. St Paul, MN.
- Ayele, A.G. 2011. Heritability and genetic advance in recombinant inbred lines for drought tolerance and other related traits in sorghum (Sorghum bicolor). Continental J. Agricultural Science 5:1-9.
- Doehlert, D. C., and M.S. McMullen. 2000. Genotype and environment effects on oat milling characteristics and groat hardness. Cereal Chem. 77:148-154
- Doehlert, C.D., and M.S. McMullen. 2008. Oat grain density measurement by sand displacement and analysis of physical components of test weight. Cereal Chem. 85: 654-659.
- Doehlert, C.D., M.S. McMullen, and R.R. Baumann. 1999. Factors affecting groat percentage in oat. Crop Sci. 39:1858-1865.
- Doehlert, C.D., M.S. McMullen, and J.J. Hammond. 2001. Genotypic and environmental effects on grain yield and quality of oat grown in North Dakota. Crop Sci. 41:1066-1072.
- Doehlert, C.D., M.S. McMullen, and J.L. Jannink. 2006. Oat grain/groat size ratios: A physical basis for test weight. Cereal Chem. 83:114-118.
- Doehlert, C.D., M.S. McMullen, J.L. Jannink, S. Panigrahi, H. Gu, and N. Riveland. 2004. Evaluation of oat kernel size uniformity. Crop Sci. 44: 1178-1186.
- Falconer, D.S., and T.F.C. Mackay. 1996. Introduction to quantitative genetics. 4th ed. Longman Technical, Essex, UK.
- Forsberg, R.A., and D.L. Reeves. 1992. Breeding oats cultivars for improved grain quality. p. 751-775. In H. G. Marshall and M. E. Sorrells (ed.) Oat science and technology. ASA, Madison, WI.
- Frey, K.J., and S.C. Wiggans. 1956. How do test weight affect oat yield. Iowa Agric. Exp. Stn. Farm Sci. 10:199-200.
- Ganssmann, W., and K. Vorwerck. 1995. Oat milling, processing and storage. Pages 369-408. In R. W. Welch, ed. The Oat Crop: Production and Utilization. Chapman and Hall. London.

- Holland, J.B. 2006. Estimating genotypic correlations and their standard errors using multivariate restricted maximum likelihood estimation with SAS Proc MIXED. Crop Sci. 46:642-654.
- Holland, J.B., and G.P. Munkvold. 2001. Genetic relationships of crown rust resistance, grain yield, test weight, and seed weight in oat. Crop Sci. 41:1041-1050.
- Holland, J.B., K.J. Frey, and E.G. Hammond. 2001. Correlated responses of fatty acid composition, grain quality and agronomic traits to nine cycle of recurrent selection for increased oil content in oat. Euphytica 122:69-79.
- Holland, J.B., W.E. Nyquist, and C.T. Cervantes-Martinez. 2003. Estimating and interpreting heritability for plant breeding: An update. Plant Breed. Rev. 22:9–112.
- Kianian, S.F., M.A. Egli, R.L. Philips, H.W. Rines, D.A. somers, B.G. Gengenbach et al. 1999. Association of groat oil content and acetyl-coA carboxylase in oat. Theor. Appl. Genet. 98:884-894.
- Klein, S.J., M.A. Smith, and K.J. Frey. 1993. Recurrent selection for test weight and grain yield of oat. Crop Sci. 33:744-749.
- Lynch, M., and B. Walsh. 1997. Genetics and analysis of quantitative traits. Sinauer Associates, Inc., Sunderland, MA.
- Lyrene, P.M., and H.L. Shands. 1975. Associations among traits in progenies from *Avena* sativa L. x A. sterilis L. crosses. Crop Sci. 15:361-363.
- McCleary, B.V., and M. Glennie-Holmes. 1985. Enzymatic quantification of $(1\rightarrow 3)$, $(1\rightarrow 4)$ - β -glucan in barley and malt. J. Inst. Brew. 91:285-295.
- Milach, S.C.K., H.W. Rines, and R.L. Phillips. 1997. Molecular genetic mapping of dwarfing genes in oat. Theor. Appl. Genet. 95:783-790.
- Nersting L.G., S.B. Andersen, R. Von Bothmer, M. Gullord, and R.B. Jorgensen. 2006. Morphological and molecular diversity of Nordic oat through one hundred years of breeding. Euphytica 150:327-337.
- Ozbas, M.O., A.S. Inan, and M.I. Cagirgan. 2009. Agronomic and quality characterization of oats genotypes selected for winter tolerance. Turkish J. of Field Crops 14:150-158.
- Peterson, D.M. 1992. Composition and nutritional characteristics of oat grain products. p. 265-292. *In* H. G. Marshall and M. E. Sorrells (ed) Oat science and technology. American Society of Agronomy and Crop Science Society of America. Madison, Wisconsin.
- Peterson, D.M., D.M. Wesenberg, D.E. Burrup, and C.A. Erickson. 2005. Relationships among agronomic traits and grain composition in oat genotypes grown in different environments. Crop Sci. 45:1249–1255.

- Pixley, K.V., and K.J. Frey. 1992. Genetic interrelations among grain quality indicators and agronomic traits for oat. Euphytica 60:149-156.
- Redaelli, R., P. Lagana, F. Rizza, O.L.D. Nicosia, and L. Cattivelli. 2008. Genetic progress of oats in Italy. Euphytica 164:679-687.
- Redaelli R., D. Sgouletta, G. Scalfati, E. De Stefanis, and P. Cacciatori. 2009. Naked oats for improving human nutrition: Genetic and agronomic variability of grain bioactive components. Crop Sci. 49:1431-1437.
- SAS Institute. 2009. Proprietary of software, version 9.1.3. SAS Institute, Cary, N.C.
- Tanhuanpaa, P., O. Manninen, A. Beattie, P. Eckstein, G. Scoles, B. Rossnagel, and E. Kiviharju. 2012. An updated doubled haploid oat linkage map and QTL mapping of agronomic and grain quality traits from canadian field trials. Genome 55 :289-301.
- Vega, U., and K.J. Frey. 1980. Transgressive segregation in inter and intraspecific crosses of barley. Euphytica 29:585-594.
- Zhu, S., and H.F. Kaeppler. 2003. A genetic linkage map for hexaploid, cultivated oat (Avena sativa L.) based on intraspecific cross 'Ogle/MAM17-5'. Theor. Appl. Genet. 107:26-35.

CHAPTER 3. GENETIC LINKAGE MAPS FOR HEXAPLOID CULTIVATED OAT (*AVENA SATIVA L.*) FROM TWO SPRING OAT POPULATIONS

3.1. Abstract

The development of linkage maps from diverse breeding populations can be useful in the genetic research of cultivated oat. The objectives of this chapter were to develop genetic linkage maps from two spring oat populations using SNP markers, identify and compare homologous linkage groups between the two populations and the recently published oat consensus map. A total of 4975 SNP markers were assessed on two recombinant inbred populations derived from the crosses 'ND991151/ND030299' (population 05021) and 'Souris/ND030299' (population 05026), using a 32-bead chip platform developed by Illumina. Individual linkage maps were constructed for the two populations using CarthaGene software with a LOD score of 9 and a maximum recombination fraction of 0.30. Thirty linkage groups using 1168 polymorphic markers were formed for population 05021, whereas population 05026 comprised 33 linkage groups using 1024 polymorphic markers. The 30 linkage groups of population 05021 contained from 3 to 62 markers, and varied in size from 15.8 to 225.3 cM for a total map size of 2601.7 cM. The 33 linkage groups of population 05026 comprised from 2 to 42 markers, and varied in size from 2.3 to 143.2 cM for a total map size of 1174.2 cM. Comparison with the recently published oat consensus map indicated that 26 of the 30 linkage groups from population 05021 can be placed on 19 of the 21 oat chromosomes, and that 31 of the 33 linkage groups from population 05026 showed homology with 20 of the 21 oat chromosomes. Further comparison of the homologous regions revealed differences in the ordering of markers between the two populations and the oat consensus map, and are an indication that genomic rearrangements and

intervarietal chromosome interchanges exist in the genome of cultivated oat. Some linkage groups, mostly in the population 05021, were significantly extended compared with the oat consensus map. Since those two linkage maps are from breeding cultivars and provide good coverage of the oat genome, they would be useful tool for identification of qualitative and quantitative trait loci.

3.2. Introduction

Cultivated oats belong to the genus *Avena* and are an allohexaploid species having 21 pairs of chromosomes with a basic chromosome number of 7 (2n=6x=42) (Zhu and Kaeppler, 2003) and a relatively large genome of 11315 Mbp (Pal et al., 2002). Genomic research in oats began in 1992 with the publication of the first molecular map for diploid species based on a cross between *A. atlantica* x *A. hirtula* (O'Donoughue et al., 1992). In 1995, the first hexaploid map based on 71 Recombinant Inbred Lines from *A. byzantina* cv 'Kanota' and *A. sativa* cv 'Ogle' (KO) was published (O'Donoughue et al., 1995). That map contained 561 RFLP markers covering a distance of 1482 cM. The KO map, over the years, had been enhanced and expanded using AFLP and DArT markers (Jin et al., 2000; Wight et al., 2003; Tinker et al., 2009). Subsequently, maps have been published for several hexaploid oat populations including 'OT207/Kanota' (Milach et al., 1997), 'Clintland64/IL86-5698' (Jin et al., 2000), 'Ogle/TAM O-301' (Portyanko et al., 2001), 'Kanota/Marion' (Groh et al., 2001), 'Ogle/MAM17-5' (Zhu and Kaeppler, 2003), 'Terra/Marion' (Dekoeyer et al., 2004), 'MN841801-1/Noble-2' (Portyanko et al., 2012), and 'Aslak/Matilda' (Tanhuanpaa et al., 2012).

The most recent map of hexaploid oat, which conciliates the number of linkage groups with the number of chromosomes, was developed by Oliver et al., 2013 using a new chromosome anchoring strategy and SNP markers. They developed six mapping populations and derived monosomics lines from the cultivars 'Kanota' and 'Sun II' to anchor the linkage groups to the 21 oat chromosomes. The molecular map was constructed using 1054 SNP markers and contained 21 linkage groups, anchored to the 21 chromosomes, with a total length of 1838 cM.

However, the oat consensus map is a useful tool that cannot be used directly in the identification of QTLs affecting important traits in cultivated oats because it is a combination of 6 mapping populations. Specific mapping populations derived from two breeding parents are particularly useful to target genes controlling agronomically important traits in oats.

The oat genome is also subject to several cytogenetics abnormalities such as inversions, translocations, and chromosomal rearrangements that make mapping difficult. If we assume that chromosomal rearrangements are relatively fixed in hexaploid oat populations, high collinearity between two homologous linkage groups for different populations is expected (Wight et al., 2003). Therefore, the oat consensus map can be used as a guide to facilitate mapping and reveal recent genomic rearrangements in the oat genome between two hexaploid oat populations.

The objectives of this chapter were to develop genetic linkage maps from two spring oat populations, identify and compare homologous linkage groups between the two populations and the recently published oat consensus map.

3.3. Materials and methods

3.3.1. DNA extraction and genotyping

F₉ seed from the two populations described earlier were sown in the fall 2012 greenhouse, for 15 days, to provide leaf tissue for DNA extraction. The leaf samples were collected in two 96deep-well plates filled with silica gel. The use of silica gel eliminates the need for liquid nitrogen or freeze drying. Leaf fragment of 2" was cut and inserted in each well, after that the two plates were sealed and sent to Dr. Shioman Chao's USDA-ARS laboratory in Fargo for DNA extraction and genotyping analysis. The DNA was extracted following the procedure of Slotta et al., 2008. Briefly, a preheated (65° C) extraction buffer ($600 \ \mu$ L 0.1M Tris-HCl, pH 7.5, 0.05 M EDTA, pH 8.0, 1.25% SDS) was added to each well. The plates were then heated at 65° C for 15 min and placed at 4° C to cool before the addition of 300 μ L chilled 6M ammonium acetate. In order to pellet proteins and cell debris, the plates were centrifuged for 20 min at 2250 g at 4° C. The supernatant was then transferred to new plates containing 400 μ L 100% isopropanol per well. DNA was pelleted by centrifugation for 20 min at 4°C, and then washed twice with 500 μ L 70% ethanol followed by 10 min centrifugation at 2250g. The quality of the DNA obtained was estimated by gel electrophoresis and quantified using a NanoDrop spectrophotometer.

After DNA extraction, the two populations were genotyped using the oat 6K SNP chip containing 4975 SNPs on an Illumina BeadStation using a 32-bead chip platform developed by Illumina (San Diego, CA). Initially, the oat 6K SNP chip contains 5743 SNPs including 3847 EST markers, 1162 DArT markers, and 734 GBS markers. At the manufacturing stage 768 SNPs failed, and at the end a total of 4975 SNPs can be assayed (Chioman, communication personal). GenomeStudio V.3 software was used to perform allele calls. To avoid mistakes due to the occurrence of cluster compression each allele call was manually edited. After validation of allele calls, the genotype data was exported from GenomeStudio to Excel for linkage mapping analysis.

3.3.2. Map construction

Individual linkage maps were constructed for the two populations using CarthaGene software (www.inra.fr/bia/T/carthagene). Segregation ratios of 1:1 corresponding to a recombinant Inbred Line (RIL) population of all segregating markers on the 190 individuals were checked using a χ^2 test. Markers with highly-distorted segregation ratios (χ^2 >20) and \geq 10% missing genotypes were

removed from the analysis. Due to the large number of markers available, the large size of the oat genome, and some cytogenetic abnormalities such as inversions, translocations, to achieve a robust result and avoid the effects of pseudolinkage, linkage groups were first obtained by the group command using two-point analysis with a LOD (Logarithm of Odds) score of 9 and a maximum recombination fraction of 0.30. The grouping threshold was then relaxed to a LOD score of 6 to validate the joining of groups based on the oat consensus map (Oliver et al., 2013) and other sources of information (Tinker et al., 2009). The ordering of linked markers was determined by maximum likelihood and simulated annealing using the commands build 20, greedy, flip and annealing of the CarthaGene software, with a LOD threshold of 3 and a marker mapping distance no greater than 25 cM. The method of simulated annealing estimates the shortest linear map by simulating different loci orders and keeping only the shortest orders. The Kosambi mapping function was used to convert recombination fractions into map distances in centimorgans (cM). Linkage groups between the two populations studied and the oat consensus map were declared to be homologous if they shared four or more markers as indicated by Hizbai et al., 2012. Graphic presentation of the LGs, QTLs, and homology between Population and oat consensus map was obtained using MapChart version 2.2 (Voorrips, 2002).

3.4. Results

3.4.1. Population 05021

A total of 1284 markers loci were polymorphic on the 97 RILs of the population 05021, 1168 of them can be placed on linkage groups, 51 were discarded for severe distortion, and 65 remained unlinked. Upon analysis, 1168 loci formed 30 linkage groups (LG) of 3 or more markers, 523 of them (46%) cosegregate with mapped loci (Table 3.2), then the final framework map contained 640 markers loci (Appendix 1). Based on homology with the oat consensus map

(Oliver et al., 2013), 26 of the 30 linkage groups (87%) can be placed on 19 of the 21 oat chromosomes (Table 3.1). No polymorphism can be detected for chromosome 10D, and 21D. The remaining 13% (4 LG) cannot be placed on any of the oat chromosomes (Data not shown). The 26 LG, homologs to the oat chromosomes, contained from 3 to 62 markers (average 24 markers) for a total of 627 markers, and varied in size from 15.8 to 225.3 cM for a total map size of 2601.7 cM (Table 3.1). The average distance between markers was 4.1 cM when omitting the cosegregant markers, and 2.2 cM when considering all the markers. A total of 401 markers (64%) of the 627 were Expressed-Sequenced Tags (EST) markers, 159 (25%) were Diversity Array Technology (DArT) markers and the remaining 67 (11%) were Genotyping-By-Sequencing (GBS) markers (Table 3.3). The 4 LG that did not share any markers with the oat chromosomes contained a total of 18 markers ranging from 3 to 6 markers, and varied in size from 8.2 to 15.9 cM, for a total map size of 49.6 cM. Total length and marker density differed between genomes. The C genome had more length (1169.2 cM) and marker density (276) than the A and the D genome. The A genome (759.7 cM - 188 markers) and the D genome (672.7 cM - 163 markers) had approximately the same length and marker density (Table 3.1).

156 marker loci (25%) deviated significantly (χ^2 >10, p<0.001) from the expected 1:1 segregation ratio as determined by χ^2 analysis, 81 of them were extremely distorted (χ^2 >15). All the distorted markers were skewed towards the 'ND030299' allele. Five linkage groups showed regions with high segregation distortion and 4 linkage groups had regions moderately distorted. Seventy-seven percent of LG 05021-1.1 and 51% of LG 05021-6 contained essentially distorted markers (Table 3.5).

After comparative mapping, 26 LG were found to be homologous with 19 of the 21 oat chromosomes. All the chromosomes of the C and A genomes are present in the population.

Chromosome 10D and 21D are missing for the D genome. A total of 178 markers, covering 1028.7 cM in the population 05021 and 773.2 cM in the oat consensus map, were shared between the two. The number of shared markers ranged from 2 (LG 05021-9 and oat chromosome 9D) to 23 (LG 05021-16 and oat chromosome 16A) (Table 3.4).

3.4.2. Population 05026

Analysis of the segregation of the SNP markers in the 93 05026 RILs population identified a total of 1054 polymorphic markers, 1024 of them can be placed into linkage groups, 16 were discarded for severe segregation distortion, and 14 remained unlinked. More than 60% of the polymorphic markers (622) cosegregate with mapped loci (Table 3.7). The final framework map contained 398 markers and formed 33 linkage groups (Appendix 2). Thirty-one (31) of the 33 LG (94%) showed homology with 20 of the 21 oat chromosomes (Oliver et al., 2013). No polymorphism can be detected for oat chromosome 21D. The 31 LG comprised from 2 to 42 markers (average 13) for a total of 392 markers, and varied in size from 2.3 cM to 143.2 cM for a total map size of 1174.2 cM (Table 3.6). When all the markers were taken into account, the average distance between markers was 1.2 cM, and when only the framework markers were considered the average distance was 3.0 cm (Table 3.8). Of the 392 markers of the framework map, a total of 285 markers (72%) were EST markers, 45 (11%) were DArT markers and the remaining 62 (16%) were GBS markers (Table 3.8). As for population 05021, total length and marker density differed between genomes. The C genome, with 211 markers and 638.0 cM, had more length and more density, followed by the A genome with 119 markers and 351.3 cM. Finally the D genome with 62 markers and 184.9 cM had less length and less density (Table 3.6).

Linkage group	Chromoso me	Length	No cosegreg	gating markers	Including co mar	osegregating kers
		сM	Total	Average	Total	Average
			markers	length	markers	length
05021 1 1	10	71.0	C geno	me	<i></i>	1.2
05021-1.1		/1.2	23	3.1	33 27	1.3
05021-1.2		63.4	23	2.8	37	1./
05021-2	20	141.6	28	5.1	59	2.4
05021-3	30	155.3	31	5.0	41	3.8
05021-4	4C	118.1	28	4.2	48	2.5
05021-5	5C	171.7	42	4.1	71	2.4
05021-6	6C	151.1	34	4.4	69	2.2
05021-7.1	7C-17A	168.1	41	4.1	119	1.4
05021-7.2	7C-17A	53.6	12	4.5	17	3.2
05021-7.3	7C-17A	16.4	4	4.1	5	3.3
05021-7.4	7C-17A	34.6	7	4.9	8	4.3
05021-7.5	7C-17A	24.1	3	8.0	4	6.0
12		1169.2	276	4.2	533	2.2
			A geno	me		
05021-8	8A	171.9	51	3.4	98	1.8
05021-11	11A	83.8	18	4.7	18	4.7
05021-13	13A	121.5	21	5.8	31	3.9
05021-15	15A	28.0	3	9.3	3	9.3
05021-16	16A	219.5	62	3.5	125	1.8
05021-19	19A	135.0	33	4.1	72	1.9
6		759.7	188	4.0	347	2.2
			D geno	me		
05021-9	9D	59.7	17	3.5	23	2.6
05021-12	12D	225.3	54	4.2	88	2.6
05021-14.1	14D	90.0	23	3.9	55	1.6
05021-14.2	14D	36.3	9	4.0	13	2.8
05021-14.3	14D	38.7	7	5.5	9	4.3
05021-14.4	14D	15.8	7	2.3	9	1.8
05021-18	18D	146.6	30	4.9	42	3.5
05021-20	20D	60.3	16	3.8	31	1.9
8		672.7	163	4.1	270	2.5
26		2601.6	627	4.1	1150	2.3

Table 3.1. Summary of the molecular linkage map from 'ND991151' x 'ND030299' F_6 recombinant inbred oat population (Population 05021) with length, average marker interval length, number of markers on each linkage group, and corresponding chromosome to the oat consensus map.

Linkage group	Chromosome	Cosegregate markers	% of cosegregation
		C genome	0.0
05021-1.1	1C	32	58
05021-1.2	1C	14	38
05021-2	2C	31	53
05021-3	3C	10	24
05021-4	4C	20	42
05021-5	5C	29	41
05021-6	6C	35	51
05021-7.1	7C-17A	78	66
05021-7.2	7C-17A	5	29
05021-7.3	7C-17A	1	20
05021-7.4	7C-17A	1	13
05021-7.5	7C-17A	1	25
		257	48
		A genome	
05021-8	8A	47	48
05021-11	11A	0	0
05021-13	13A	10	32
05021-15	15A	0	0
05021-16	16A	39	54
05021-19	19A	63	50
		159	46
		D genome	
05021-9	9D	6	26
05021-12	12D	34	37
05021-14.1	14D	32	58
05021-14.2	14D	4	31
05021-14.3	14D	2	22
05021-14.4	14D	2	22
05021-18	18D	12	29
05021-20	20D	15	48
		107	40
		523	46

Table 3.2. Cosegregation by genome observed in the linkage map from 'ND991151' x 'ND030299' F₆ recombinant inbred oat population (Population 05021).

Linkage group	Includ	ing cosegre	gating ma	rkers	No c	osegregat	ing mark	ters
	cDNA	DArT	GBS	Total	cDNA	DarT	GBS	Total
			C ger	nome				
05021-1.1	36	10	9	55	11	10	2	23
05021-1.2	23	9	5	37	13	8	2	23
05021-2	43	9	7	59	13	9	6	28
05021-3	24	10	7	41	19	7	5	31
05021-4	33	12	3	48	17	9	2	28
05021-5	57	8	6	71	34	5	3	42
05021-6	46	13	10	69	25	6	3	34
05021-7.1	72	21	26	119	24	12	5	41
05021-7.2	9	6	2	17	5	5	2	12
05021-7.3	4	1	0	5	3	1	0	4
05021-7.4	5	2	1	8	4	2	1	7
05021-7.5	3	0	1	4	2	0	1	3
	355	101	77	533	170	74	32	276
			A gei	nome				
05021-8	69	25	4	98	31	17	3	51
05021-11	13	4	1	18	13	4	1	18
05021-13	17	7	7	31	14	3	4	21
05021-15	3	0	0	3	3	0	0	3
05021-16	97	17	11	125	42	13	7	62
05021-19	55	9	8	72	23	8	2	33
	254	62	31	347	126	45	17	188
			D gei	nome				
05021-9	13	7	3	23	8	6	3	17
05021-12	59	19	10	88	35	16	3	54
05021-14.1	40	6	9	55	15	5	3	23
05021-14.2	4	6	3	13	3	4	2	9
05021-14.3	9	0	0	9	7	0	0	7
05021-14.4	5	2	2	9	3	2	2	7
05021-18	31	4	7	42	23	4	3	30
05021-20	24	3	4	31	11	3	2	16
	185	47	38	270	105	40	18	163
	794	210	146	1150	401	159	67	627

Table 3.3. Summary of marker type for the molecular linkage map from 'ND991151' x 'ND030299' F_6 recombinant inbred oat population (Population 05021).

GBS = Genotyping-By-Sequencing markers

Po	pulation 05021	1		Oat consensus map				
LG	Homologous	Length	Markers shared	Chromo	Homologous	Length		
	segments	(cM)		some	segments	(cM)		
05021-1.1	4.3-31.6	27.3	14	1C	24.5-43.8	19.3		
					71.5-74.8	3.3		
05021-1.2	17.0-27.7	10.7	5	1C	47.2-63.2	16.0		
05021-2	22.2-63.2	41.0	11	2C	3.0-39.9	36.9		
05021-3	7.2-21.2	14.0	6	3C	11.5-15.7	4.2		
	77.1-131.1	54.0			49.5-86.6	37.1		
05021-4	4.3-11.4	7.1	6	4C	75.5-90.5	15.0		
	100.1-104.3	4.2						
05021-5	1.1-51.2	50.1	17	5C	17.4-76.4	59.0		
	144.7-153.2	8.5			0.0-7.1	7.1		
05021-6	23.7-109.3	85.6	18	6C	15.4-77.9	62.5		
05021-7.1	69.0-167.0	98.0	13	7C-17A	0.0-78.4	78.4		
05021-8	30.2-59.0	28.8	12	8A	17.3-62.0	44.7		
	137.3-139.5	2.2						
05021-9	5.5-47.1	41.6	2	9D	64.2-91.0	26.8		
05021-12	38.6-85.3	46.7	14	12D	25.7-115.0	89.3		
	126.5-225.3	98.8						
05021-13	22.8-61.4	38.6	5	13A	60.1-91.4	31.3		
05021-14.1	52.4-76.0	23.6	6	14D	77.1-102.3	22.0		
05021-14.3	22.9-38.7	15.8	2	14D	0.0-22.0	22.0		
05021-16	1.1-41.3	40.2	23	16A	10.8-80.8	70.0		
	85.7-205.6	119.9						
05021-18	5.3-33.8	28.5	8	18D	0.0-39.9	39.9		
	93.4-146.1	52.7						
05021-19	0.0-3.8	3.8	11	19A	28.6-97.5	68.9		
	68.8-135.0	66.2						
05021-20	24.7-45.5	20.8	5	20D	56.3-75.8	19.5		
Total		1028.7	178			773.2		

Table 3.4. Homologous segments between the molecular linkage map from population 05021 and oat consensus map.

Linkage group	Region (cM)	Length (cM)	% of linkage group	Comment
05021-1.1	15.4-70.0	54.6	77	Highly distorted ($\chi^2 > 15$)
05021-2	44.9-77.7	32.8	23	Highly distorted ($\chi^2 > 15$)
05021-8	29.7-84.5	54.8	32	Highly distorted ($\chi^2 > 15$)
05021-16	27.0-63.6	36.6	17	Highly distorted ($\chi^2 > 15$)
05021-18	50.7-110.6	59.9	41	Highly distorted ($\chi^2 > 15$)
05021-5	43.4-100.1	56.7	33	Moderately distorted ($\chi^2 > 10$)
05021-6	42.6-119.9	77.3	51	Moderately distorted ($\chi^2 > 10$)
05021-14.3	22.4-38.7	16.3	42	Moderately distorted ($\chi^2 > 10$)
05021-19	37.6-85.9	48.3	36	Moderately distorted ($\chi^2 > 10$)

Table 3.5. Region of segregation distortion for the molecular linkage map from 'ND991151' x 'ND030299' F₆ recombinant inbred oat population (Population 05021).

Less segregation distortion, compared to population 05021 was observed. Only 35 markers (9%) showed distortion and deviated significantly ($\chi^2 > 10$, p<0.001) from the expected 1:1 segregation ratio, 24 of them were extremely distorted ($\chi^2 > 15$). Twenty-five (25) of the distorted markers were skewed towards the 'ND030299' allele, the remaining 10 markers were skewed towards the 'Souris' allele. Linkage group 05026-12.2, 05026-18, 05026-13.1 from position 0 to 13.9 cM, and 05026-11 from position 0 to 0.3 contained essentially extremely distorted markers. Linkage groups 05026-13.1, 05026-12.2, and 05026-18 were all skewed towards to 'ND030299' allele whereas linkage group 05026-11 was skewed towards the 'Souris' allele.

647.3 cM, representing 171 markers, in the population 05026, showed homology with 557.5 cM in the oat consensus map. The number of shared markers varied from 2 (LG 05026-14.2 and OC 14D) to 28 (LG 05026-2 and OC 2C) (Table 3.9). 396 markers, covering 886.7 cM in the population 05021 and 540.9 cM in the population 05026, were shared between the two populations. Homologous markers ranged from 3 (LG 05021-13 and LG 05026-13.2) to 53 (LG 05021-16 and LG 05026-16.1) (Table 3.10).

Linkage Chromo-Including cosegregating Length No cosegregating markers markers group some Total Total Average Average сM markers length markers length C genome 05026-1.1 1C 36.8 34 1.1 17 2.2 05026-1.2 1C24.3 28 0.9 11 2.2 2C87 0.8 20 05026-2 71.1 3.6 05026-3 3C 143.2 81 1.8 38 3.8 05026-4 4C23.5 43 0.5 21 1.1 5C 128.7 109 1.2 42 3.1 05026-5.1 05026-5.2 5C 45.3 47 1.0 16 2.8 05026-6 6C 75.0 46 1.6 17 4.4 05026-7.1 7C-17A 71.5 27 2.6 15 4.8 05026-7.2 7C-17A 18.0 18 1.0 12 1.5 05026-7.3 0.2 2 0.3 7C-17A 0.6 3 11 638.0 523 1.2 211 3.0 A genome 05026-8.1 8A 24.1 79 0.3 14 1.7 2.8 7.4 05026-8.2 8A 22.3 8 3 05026-11 11A 35.0 34 1.0 15 2.3 0.8 14 2.7 05026-13.1 13A 38.1 46 13A 2.2 5 5.3 05026-13.2 26.7 12 05026-15 15A 74.4 22 3.4 15 5.0 05026-16.1 16A 60.4 74 0.8 28 2.2 05026-16.2 16A 17.0 14 1.2 5 3.4 0.3 5 0.9 05026-17 17A-7C 4.5 14 19A 05026-19 48.8 49 1.0 15 3.3 10 351.3 352 1.0 119 3.0 **D** genome 05026-9 9D 2.3 0.5 2 5 1.2 05026-10 10D-F-1 26.3 19 1.4 11 2.4 2.7 8 6.7 05026-12.1 12D 53.3 20 2 05026-12.2 12D 2.3 4 0.6 1.2 05026-14.1 14D 29.0 20 1.5 8 3.6 0.7 4 05026-14.2 14D 5.2 8 1.3 05026-14.3 14D 4.3 3 1.4 3 1.4

Table 3.6. Summary of the molecular linkage map from 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) with length, average marker interval length, number of markers on each linkage group, and corresponding chromosome to the oat consensus map.

Table 3.6. Summary of the molecular linkage map from 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) with length, average marker interval length, number of markers on each linkage group, and corresponding chromosome to the oat consensus map (Continued).

Linkage group	Chromoso me	Length	Including cosegregating markers		No cosegregating markers		
		cM	Total markers	Average length	Total markers	Average length	
05026-18	18D	47.2	30	1.6	10	4.7	
05026-20.1	20D	7.5	18	0.4	7	1.1	
05026-20.2	20D	7.5	12	0.6	7	1.1	
10		184.9	139	1.3	62	3.0	
31		1174.2	1014	1.2	392	3.0	

3.5. Discussion

3.5.1. Comparative mapping

3.5.1.1. Population 05021 and oat consensus map

178 markers were shared between population 05021 and oat consensus map representing 40% of the former (1028.7 cM) and 42% of the latter (773.2 cM). The order of markers was very well conserved on linkage groups 05021-3, 05021-7.1, 05021-13, 05021-14, and 05021-20 comparing to the corresponding oat chromosomes 3C, 7C-17A, 13A, 14D, and 20D.

Minor rearrangements and one or several inversions of markers were observed for linkage groups (LG) 05021-1.1, 05021-1.2, 05021-5, 05021-6, 05021-8, 05021-12, 05021-16, 05021-19. On LG 05021-1.1, markers DS cc9481 218, and ES cc13854 225, ES15 c16679 330, and ES cc11019 290 (Figure 3.2, red) mapped at position 23.6 to 29 cM were interchanged with a group of 4 cosegregating markers mapped at position 17 cM and marker ES02 c6368 605 mapped at position 4.3 (Figure 3.1, green). Comparing to oat chromosome 1C, markers ES02 c12621 204, ES02 lrc13446 346, and ES01 c3447 952 located at position 25.4 to 27.7 cM (Figure 3.1, violet) on LG 05021-1.2 switched order with markers ES01 C9472 428 and ES02 lrc13446 328 mapped at position 17 to 18.1 cM (Figure

3.1, dark blue). Marker ES15_C7706_583 positioned at the end of LG 05021-2 (112.2 cM) had been mapped at the beginning of oat chromosome 2C (3 cM) (Figure 3.2).

A group of 4 markers (Figure 3.2, red) placed at the end of LG 05021-5 from position 106.8 to 153.2 cM had been mapped at the very beginning of oat chromosome 5C from 0 to 7.1 cM. Another group of 5 markers (Figure 3.3, green) located at the middle of LG 05021-5 from position 36.3 to 43.9 cM had been mapped at the end of oat chromosome 5C from position 73.2 to 76.4 cM. Two small rearrangements were observed in LG 05021-6. Marker ES01 c5633 477 changed order with marker ES02 c247 241 (Figure 3.4, red and green). Markers ES02 c15952 348 and ES02 c28827 474 that cosegregated at position 53.9 cM had been mapped respectively at position 15.4 to 17.5 cM in oat chromosome 6C (Figure 3.4, violet). Cosegregating markers ES02 c15926 519 and ES17 c11418 547 placed at the beginning of LG 05021-8 at position 30.2 cM, had been mapped at the opposite end of oat chromosome 8A at position 61.6 and 62 cM, respectively (Figure 3.5, red). Two group of markers were inverted in LG 05021-12 comparing to oat chromosome 12D. A first group of 6 markers (Figure 3.6, red) placed at position 126.5 to 225.3 cM had been mapped in the oat chromosome 12D at position 25.6 to 67.9 cM. The second group comprised 8 markers (Figure 3.6, green) placed at position 38.6 to 85.3 cM had been mapped at position 91.3 to 115 cM in oat chromosome 12D. Two groups of markers were also inverted in LG 05021-16. The first group consisted of 9 markers (Figure 3.7, red) had been mapped, in the same order in comparison with oat chromosome 16A, at position 85.7 to 197.3 cM. The second group of 9 markers also (Figure 3.7, green) had been mapped, in reverse order relative to oat chromosome 16A, at position 19.1 to 1.1 cM. Some markers were inverted in LG 05021-18 comparing to oat chromosome 18D. Markers ES15 c10680 325 and ES02 c4268 604, ES17 c3693 421 and ES15 c13674 329, ES17 c2976 706 and ES02 c13415 701 switched order (Figure 3.8).

Linkage group	Chromosome	Cosegregate markers	% of cosegregation							
C genome										
05026-1.1	1C	17	50							
05026-1.2	1C	17	61							
05026-2	2C	67	77							
05026-3	3C	43	55							
05026-4	4C	22	51							
05026-5.1	5C	67	61							
05026-5.2	5C	31	66							
05026-6	6C	29	63							
05026-7.1	7C-17A	12	44							
05026-7.2	7C-17A	6	33							
05026-7.3	7C-17A	1	33							
		312	60							
A genome										
05026-8.1	8A	65	82							
05026-8.2	8A	5	63							
05026-11	11A	19	56							
05026-13.1	13A	32	70							
05026-13.2	13A	7	58							
05026-15	15A	7	32							
05026-16.1	16A	46	62							
05026-16.2	16A	9	64							
05026-17	17A-7C	9	64							
05026-19	19A	34	69							
	233		66							
D genome										
05026-9	9D	3	60							
05026-10	10D-F-1	8	42							
05026-12.1	12D	12	60							
05026-12.2	12D	2	50							
05026-14.1	14D	12	60							
05026-14.2	14D	4	50							
05026-14.3	14D	0	0							
05026-18	18D	20	67							
05026-20.1	20D	11	61							
05026-20.2	20D	5	42							
		77	55							
		622	61							

Table 3.7. Cosegregation by genome observed in the linkage map from 'Souris' x 'ND030299' F_6 recombinant inbred oat population (Population 05026).

Linkage group	Including cosegregating markers				No cosegregating markers			
	cDNA	DArT	GBS	Total	cDNA	DArT	GBS	Total
			C ge	nome				
05026-1.1	27	1	6	34	11	0	6	17
05026-1.2	25	3	0	28	9	2	0	11
05026-2	67	10	10	87	14	3	3	20
05026-3	53	14	14	81	24	7	7	38
05026-4	31	5	7	43	16	3	2	21
05026-5.1	86	6	17	109	33	2	7	42
05026-5.2	38	7	2	47	13	2	1	16
05026-6	30	8	8	46	13	3	1	17
05026-7.1	16	7	4	27	9	4	2	15
05026-7.2	6	5	7	18	3	4	5	12
05026-7.3	3	0	0	3	2	0	0	2
	382	66	75	523	147	30	34	211
			A ge	nome				
05026-8.1	62	15	2	79	11	3	0	14
05026-8.2	8	0	0	8	3	0	0	3
05026-11	29	1	4	34	14	0	1	15
05026-13.1	36	7	3	46	10	3	1	14
05026-13.2	5	4	3	12	2	1	2	5
05026-15	16	2	4	22	11	1	3	15
05026-16.1	59	6	9	74	21	2	5	28
05026-16.2	13	0	1	14	5	0	0	5
05026-17	14	0	0	14	5	0	0	5
05026-19	37	5	7	49	12	1	2	15
	279	40	33	352	94	11	14	119
			D ge	nome				
05026-9	3	1	1	5	0	1	1	2
05026-10	14	2	3	19	9	1	1	11
05026-12.1	14	6	0	20	7	1	0	8
05026-12.2	3	0	1	4	1	0	1	2
05026-14.1	13	1	6	20	4	1	3	8
05026-14.2	5	0	3	8	3	0	1	4
05026-14.3	3	0	0	3	3	0	0	3

Table 3.8. Summary of marker type for the molecular linkage map from 'Souris' x 'ND030299' F_6 recombinant inbred oat population (Population 05026).
0		1 1	< I			,		
Linkage group	Includ	ing cosegre	gating mar	kers	No c	osegregat	ing mark	cers
	cDNA	DArT	GBS	Total	cDNA	DArT	GBS	Total
05026-20.1	16	1	1	18	6	0	1	7
05026-20.2	8	0	4	12	5	0	2	7
	101	12	26	139	44	4	14	62
	762	118	134	1014	285	45	62	392

Table 3.8. Summary of marker type for the molecular linkage map from 'Souris' x 'ND030299' F_6 recombinant inbred oat population (Population 05026) (Continued).

Table 3.9. Homologous segments between the molecular linkage map from population 05026 and oat consensus map.

Po	pulation 05026			Oa	at consensus maj	0
LG	Homologous	Length	Markers shared	Chromo	Homologous	Length
	segments	(cM)		some	segments	(cM)
05026-1.1	0.0-15.8	15.8	3	1C	47.2-63.2	16.0
05026-1.2	0.0-20.3	20.3	4		68.7-74.8	6.1
05026-2	0.0-61.3	61.3	28	2C	3.0-44.3	41.3
05026-3	26.9-143.2	116.3	19	3C	11.5-86.6	75.1
05026-4	5.8-22.4	16.6	6	4C	60.8-90.5	29.7
05026-5.1	66.5-101.0	34.5	17	5C	5.6-30.3	24.7
05026-5.2	0.0-44.7	44.7	16	5C	60.1-108.4	48.3
05026-6	6.6-56.5	49.9	7	6C	26.6-63.9	37.3
05026-8.1	3.5-7.6	4.1	10	8A	17.3-20.6	3.3
05026-10	9.4-26.3	16.9	2	10D-F-1	0.0-4.8	4.8
05026-11	0.0-35.0	35.0	7	11A	11.1-36.0	24.9
05026-12.1	0.6-53.3	52.7	3	12D	43.9-104.7	60.8
05026-13.1	0.0-36.4	36.4	9	13A	83.7-111.2	27.5
05026-14.1	17.6-29.0	11.4	4	14D	96.2-105.8	9.6
05026-14.2	0.0-5.2	5.2	2	14D	22.0-25.5	3.5
05026-15	0.0-48.5	48.5	4	15A	0.0-48.2	48.2
05026-16.1	36.4-60.4	24.0	11	16A	10.8-36.6	25.8
05026-18	37.3-42.1	4.8	2	18D	31.4-41.4	10.0
05026-19	2.2-48.8	46.6	8	19A	0.0-51.9	51.9
05026-20.1	1.7-4.0	2.3	7	20D	72.6-75.8	3.2
Total		647.3	171			552.0

Р	opulation 05021	00021.			Population 05026	
LG	Homologous	Length	Markers	LG	Homologous	Length
20	segments	(cM)	shared	20	segments	(cM)
05021-1 1	0.0	0.0	9	05026-1 2	14 5-15 1	0.6
05021-1.2	17.0-63.4	46.4	18	05026-1.1	0.0-29.9	29.9
05021-2	22.2-35.2	13.0	38	05026-2	0.0-71.1	71.1
	60.0-141.6	81.6				
05021-3	0-131.1	131.1	27	05026-3	20.7-143.2	122.5
05021-4	2 7-17 5	14.8	28	05026-4	4 6-22 4	17.8
	35.9-39.3	3.4				
	100.1-104.3	4.2				
05021-5	0.0-4.9	4.9	23	05026-5.1	66.5-122.4	55.9
	73.0-171.7	98.7	-			
05021-5	35.8-51.2	15.4	10	05026-5.2	24.3-36.9	12.6
05021-6	0.0-6.0	6.0	33	05026-6	73.2-75.0	1.8
	24.2-30.2	6.0			52.5-56.0	3.5
	63.7-64.8	1.1			34.3-36.0	1.7
	114.4-144.4	30.0			0.6-7.2	6.6
05021-7.2	0.0-16.6	16.6	10	05026-7.1	36.2-44.6	8.4
05021-7.3	0.0-14.8	14.8	3	05026-7.3	0.0-0.6	0.6
05021-7.1	0.0	0.0	3	05026-7.2	2.9-12.4	9.5
05021-8	53.5-84.5	31.0	73	05026-8.1	0.0-24.1	24.1
	120.0-138.4	18.4				
05021-9	47.1-49.2	2.1	4	05026-9	0.0-2.3	2.3
05021-11	0.0-2.2	2.2	10	05026-11	19.2-21.6	2.4
	40.4-41.1	0.7			31.6-33.3	1.7
05021-12	38.6-59.6	21.0	18	05026-12.1	0-53.3	53.3
	119.5-126.5	7.0				
	208.6-225.3	16.7				
05021-12	168.0-168.6	0.6	4	05026-12.2	0-2.3	2.3
05021-13	61.4-96.5	35.1	5	05026-13.1	8.9-38.1	29.2
05021-13	0.0-13.0	13.0	8	05026-13.2	23.3-26.7	3.4
05021-14.1	62.8-90.0	27.2	12	05026-14.1	4.0-29.0	25.0
05021-14.3	0.0-10.8	10.8	3	05026-14.3	0.0-4.3	4.3
05021-14.3	37.1-38.7	1.6	3	05026-14.2	0.0-0.7	0.7
05021-14.4	0.8-7.0	6.2	5	05026-14.1	0.0-0.1	0.1
05021-16	98.2-122.0	23.8	65	05026-16.1	0.0-60.4	60.4
	146.3-151.4	5.1				
	188.5-219.5	31.0				
05021-7.1	120.6-124.3	3.7	12	05026-17	2.8-4.5	1.7
05021-18	136.5-146.6	10.1	7	05026-18	42.1-47.2	5.1
05021-19	68.8-135.0	66.2	35	05026-19	16.6-48.8	32.2
05021-20	43.4-58.7	15.3	11	05026-20.1	3.4-7.5	4.1
05021-20	8.6-22.0	13.4	10	05026-20.2	0.0-4.6	4.6
Total		850.2	487			599.4

Table 3.10. Homologous segments between the molecular linkage map from population 05026 and population 05021.

An inversion between two groups of markers was observed in LG 05021-19 in comparison with oat chromosome 19A. A group of 4 markers (Figure 3.9, red) mapped at position 68.8 to 135 cM were inverted with another group of 7 markers (Figure 3.9, green) mapped at position 0 to 3.8 cM.

The longer map observed in population 05021 comparing to oat consensus map might be due to the expansion of some regions of population 05021. Markers ES17_c3418_95, DS_cc8468_91, ES_cc11290_204, DS_cc4033_368 found at position 22.2 to 29 cM on LG 05021-2, and markers ES_cc4978_509, ES01_c8470_599, ES01_c1635_353, ES_cc8700_285, ES01_c24681_389 located at position 60 to 63.2 cM had been mapped together in oat chromosome 2C (Figure 3.4, green and violet). Two groups of markers located at a distance of 90 cM in LG 05021-4 had been mapped together in oat chromosome 4C over 15 cM (Figure 3.10). Two markers (ES_02_c17364_288 and ES02_c13236_178) located respectively at position 1.1 and 4.9 cM in LG 05021-5 and 3 markers (ES17_c3625_404, ES01_c16767_69, ES02_c16344_73) found at position 59 to 60.1 cM had been mapped together in oat chromosome 5C (Figure 3.3, blue and violet). Marker ES02_c16344_816 located at position 59 cM and a group of 9 cosegregating at position 138.4 cM in LG 05021-8 had been mapped together in oat chromosome 8A (Figure 3.5, blue).

05021-1.1



Figure 3.1. Conserved segments between LG 05021-1.1, 05021-1.2, and oat chromosome 1C.

05021-2

		001	GMI ES CC4334 304
		3.0 -	GMI ES15 c7706 583
		5.4	GMI_ES01_c13403_102
ר 0.0	/ GMI_ES01_c25052_201	5.8 -	GMI_ES15_c690_324
15.8	/ OPt-6104	22.4	IGMI ES CC12360 189 GMI DS CC9187 141
22.2 -	∭GMI_ES14_c19842_74 GMI_ES17_c3418_95	27.1 -	GMI ES17 c3418 95
~~~ \\	IGMI_ES05_c15526_511 GMI_ES14_c7020_89	28.4	GMI_ES17_c3291_859
25.5	GMI_ES05_c8599_963 GMI_DS_LB_4000	31.2 -	GMI ES02 c3181 680
20.0 11	[IGMI_ES03_Irc9679_178	33.0 -	GMI ES01 c13657 337
27.7	GMI_ES14_c6271_602	33.1 -	GMI_ES02_c33014_195
88 1	∦ GMI_ES14_c6885_194 GMI_ES15_c6381_389	35.1 -	GMI ES CC4978 509 GMI ES01 c8470 599
WH	GMI_ES14_c8931_606 GMI_ES05_c101_122	35.3 -	GMI DS CC8468 91
28.2	GMI_DS_CC8468_91 GMI_ES05_c18574_288	35.7 -	GMI ES CC11290 204
	GMI_ES05_c5594_313 GMI_ES15_c7440_275	36.0	GMI ES CC6708 301
	IGMI_ES_CC11290_204	36.4	GMI ES01 c1635 353 GMI ES CC8700 285
29.3	YGMI_ES15_Irc8841_69 GMI_DS_CC4033_368	- 36 8 - P	GMI DS CC4033 368
29.9 - 🎢 🔤	GMI_ES05_c16116_187	37.5	GMI ES17 Irc14708 436 GMI ES02 c17596 199
31.2	WGMI_GBS_73687 GMI_GBS_64994	37.6	GMI ES01 c24681 389
31.8 -	GMI_GBS_108223	37.9	GMI ES02 Irc37378 471 GMI ES02 c38444 270
35.2 -	WGMI_ES05_c8417_343 GMI_ES15_c13909_303	38.3	GMI DS CC6030 255 GMI ES01 c3327 180
43.4 1	P 0Pt-10449	38.5 -	GMI_ES_CC9730_217
44.9	P 0Pt-110/5	38.6 -	GMI DS CC1057 147
53.1 -/	0PT-14513	38.8 -	/ GMI_DS_CC9934_185
	IGMI_ES03_C14095_37_GMI_ES03_C22566_720	39.1 🛁	GMI ES15 c2110 730
59.4 - JE	GMI_ES03_03966_66 GMI_ES15_07005_126	[~] 39.2 -⁄/	GMI_SNP22314_1
	IGMI_ESU2_01435_212	39.9 -//	GMI_ES01_c3302_178
- co o (11)	GWI_ESUS_CISSUI_SOI GWI_ESUI_COSU2_178	40.3	GMI_ES02_c8676_360
60.0	GMI_ES17_C4046_112 GMI_ESU1_C24661_369	40.7	GMI_ES01_c12277_1252 GMI_ES01_c3298_226
	IGMI_ES14_C4827_489 GMI_GBS_7061	41.4	GMI_DS_CC2679_57
62.2	GMI_ES_CC10136_320	42.1	GMI_ES02_c20004_64
62.2	CMI_ES05_C10021_522	42.2	GMI_ES02_c37525_294
02.7	GMLES01_01635_353 GMLES_008700_285	MM	GMI_DS_CC2032_276 GMI_ES01_c14226_61
63.2	GML E S01 c8470 599 GML ES CC4978 509	43.3	GMI_ES01_c11537_64 GMI_ES01_c24758_394
777	GML ES03 c10410_600	10 O M	IGMI_ES01_C9044_416 GMI_ES15_C112_624
78.8	GMI_GBS_70341	43.9	GMI_ES_CC10804_287
	GMI ES22 c1298 437 GMI ES14 c2320 26	44.0	GMI_ES_CC13637_173 GMI_ES17_IIC20311_707
86.0	GMI DS LB 3936 GMI ES17 c899 571	1	IGMI_ESU2_0045_199 GMI_ES_005909_242
100.0	GMI GBS 81139	Ň	CML ES17_C4200_911 GML ES15_C6925_619
107.9	GMI ES05 Irc18863 260	J. J.	GML_ES15_C2996_340 GML_ES15_C7006_322
112.2	GMI ES15 c7706 583	44.3	GML_ES15_C4287_160_GML_ES_CC7315_202
115.1 🖞 👘	GMI_GBS_31405		GMI_DS_CC5529_175_GMI_ES17_C7296_694
141.6	GMI_DS_LB_4073		GML_SNP5252_1
		44.5	GMI ES02 c4655 137
		60.2	TLP-3
		71.3	GMI SNP11164 1
		76.2	TLP-1

Figure 3.2. Conserved segments between LG 05021-2, and oat chromosome 2C.

05021	-5

0.0 1	GMI ES15 c6458 250	0.01	r GMI E \$17 c5197 503
11	GMI E S02 c17364 288 GMI E S05 c11331 441	28-	GML ES02 c10836 31
- "' I	IGML ES03 (13946-240 GML ES05 (26190-676	4.5	GML SN P51_1
22-	GMI_ES02_c2221_320 GMI_ES15_c8606_304	471	GML SN P51 2
2.2	CMI_ES17_011616_252	4.7	CML 5 517 61196 142
<u> </u>	IGWI_ES17_UT1616_203	0.67	GIVILES17_C1186_142
3.5 1	GMI_ES22_08005_394	6.01	GIVII_ESU2_026223_26
4.2	/ GMI_ES03_IC10769_351	6.2	GMI_ES15_C6191_370
4.9 -	GMI_GBS_9614 GMI_ES02_C13236_178	7.1	GMI_E \$02_c15089_19
ן 14.9	GMI_ES05_c17311_95	8.0 1	GMI_ES01_c1223_200
15.4 -	GMI_ES05_c553_469	10.7	GMI_ES15_c3706_138
17.0 -	GMI_ES05_c20981_389	44.0	[GMI_ES15_c6652_253]
ار 18.6 ⁻	GMI_ES05_c2760_657	11.2	GMI ES_CC16529_138
19.1 -	// GMI_ES22_c11452_253	11.4	- GMI ES15 c12818 36
34.2 ٦	🚍 / GMI_ES02_c15552_416	11.5	GMI_ES15_c6914_663
35.3 -	// GMI ⁻ ES05 ⁻ c26873 ⁻ 318	12 4	GMI_ES01_c11975_32
1	/IGMI DS LB 5972 GMI ES02 c11794 636	127-	GMLES17_c12075_35
35.8 ~	GMI DS LB 1766	12.8	IGM ES CC11658 395
36.3 -	GMI ES CC14804 235	12.0	GMI_ES15_08970_315
00.0	GML ES14 (769 500 GML DS LB 10794	12.3	UGML_ES01_09170_468
43.4 -⁄	GMI E 501 Irc8208 413 GMI E 503 c21288 330 -	13.4	GML ES01_012564_21
1	LCML E 502 012844 499 CML E 517 0552 217		IGML ES01_022008_15
43.9 [/] /	EN CIMI E 502 C12644 466 GIMI E 517 C655 217	í 🗖 🚽	CMI_ES01_022390_13
		13.6	GNI_ES01_IIC0457_64
46.1 -1	M GMI_GBS_21414		GMI_ES15_C5451_544
51.2 °j	GMI_DS_CC6822_86 GMI_ES17_c3006_879		IGNI_ES02_C4756_515
	GMI_ES22_c1052_894 GMI_ES15_c10501_398 / /	13.7 -	BM_897D
52 A J	GMI_ES15_Irc9414_222 GMI_ES_CC12161_200 / //	14.0	GMI_ES02_c28204_25
33.4	GMI_ES05_c9138_267 GMI_GBS_90539	14.1 -	GMI_ES01_c6298_257
(	🖃 IGMI ES22 c18205 366 / /	14.3	GMI_DS_CC6107_131
53.9	GMI ES22 c8057 338	17.4	GMI_DS_oPt-1466_323
59.0	GMI E \$17 c3625 404	17.7 -	GMI_ES15_c8238_156
60.1	GMI E S01 c16767 69 GMI E S02 c3374 73	17.9	GMI_ES15_c9085_462
72.5	GMI DS oPt-1466 323	18.4 -	BM 878
73.0	GML E \$01 c3480 411	19.1 -	GMIES01 c3480 411
00.0	M GML ES14 c10034 107	19.3 -	GMI_ES15_c20302_11
90.0	MICMI ES17 04047 260 CMI ES17 04047 520	21.9	GMI_ES02_c11855_52
97.3	CMI_CBS_10607_CMI_ES02_c6656_426	22.4-	GMI_ES01_c25986_120
	CMI_GDS_19097 GMI_E305_00000_400	22.7-	GMI_ES15_c2968_502
98.5	GWI_ES22_L3073_197 GWI_ES17_L4716_700	22.0-	GMI_ES17_c4979_427
100.1	IGMI_ES15_012291_669	24.0-	GMI E 502 c17364 28
100.1	GMI_ES17_C9953_261	24.5	GML ES17 c18582 10
103.4	GMI_GBS_38632	23.4	GMI_ES17_010302_13
106.8 -	GIVILES1/_C1186_142	21.0	AF237553_1
109.6	GMI_DS_LB_7757	20.4	CML E 617 62625 404
120.5	oPt-7110	29.01	GIVIE 317_03625_404
136.1 -	oPt-6125	30.3	GIVILES02_C13236_17
144.7 -	GMI_ES15_c18028_289 GMI_ES17_c5197_503	31.2 -	GMI_ES01_C16767_69
150.0 -	GMI_ES05_c26263_333	31.5-	BA_grs_c5853_310 GM
150.5 -	GMI_ES05_c4270_561	34.8	GMI_ES01_c12952_34
151.0	GMI ES15 c6191 370	37.2 -	GMI_ES17_c5744_689
153.2 -	GMI E S02 c15089 196	38.4 -	GMI_ES02_c14691_63
154.9	GMI ES02 c2714 373	38.9-	GMI_ES01_c1493_96
171.2	GMI ES05 Irc12954 281	39.2 -	GMI_ES01_c12117_56
	GML ES22 c13490 299 GML ES01 c3990 524	41.0	GMI_ES17_c2183 936
171.7	GML GBS 57836	57.5-	GMI_ES02_c3531_401
		60.1-	GMI_DS_CC6822_86
		65.7 -	GMI ES15 c10103 41

ES02_c10836_312 SNP51_1 SN P51_2 **E \$17_c1186_142** E\$02_c26223_268 E \$15_c6191_370 E \$02_c15089_196 ES01_c1223_200 ES15_c3706_138 ES15_c6652_253 GMI_ES02_c15112_271 ES_CC16529_138 ES15_c12818_361 ES15_c12818_361 ES15_c12818_361 ES15_c6914_663 ES01_c11975_322 GMI_ES15_c12600_230 ES17_c12075_354 ES_CC11685_395 GMI_ES17_c1482_118 ES15_c8970_315 ES01_c9170_468 GMI_DS_CC892_260 ES01_c12564_210 GMI_ES17_c5762_827 ES01_c12564_210 GMI_ES15_c12436_395 ES01_inc8457_64 GMI_ES01_c11126_277 ES15_c5451_344 GMI_ES15_c3159_412 ES02_c4756_515 GMI_ES01_c4174_228 897b 97b 397b ES02_c28204_255 ES01_c6298_257 DS_CC6107_131 DS_0Pt-1466_323 ES15_c8238_156 ES15_c9085_462 E \$01_c3480_411 ES15_c20302_110 ES02_c11855_528 ES01_c25986_126 ES15_c2968_502 ES17_c4979_427 ES02_c17364_288 ES17_c18582_193 ES02_lrc23878_108 7553-1 03625 40 E \$02_c13236_178 GMI ES02 c3374 73 Eso1_c16767_69_GMI_ESo2_c3374_73 ps_c5853_310_GMI_ESo2_c22115_682 ESO1_c12952_349 ESO2_c14691_637 ESO1_c1493_96 ESO1_c12117_562 ES17_c2183_936 ESO2_c3531_401 DS_CC6822_86 ES15_c10403_410 GMI_ESU2_G3S31_401 GMI_DS_CC6822_86 GMI_ES15_c10103_419 GMI_ES02_c31937_212 GMI_ES02_c21781_486 GMI_ES17_c653_546 GMI_ES01_c2879_300 GMI_ES01_c13663_212 GMI_ES01_c2879_300 GMI_ES01_c13663_212 GMI_ES01_c2879_300 GMI_ES01_c2971_66 GMI_ES01_c2974 GMI_ES01_c29634_105 GMI_ES15_c3717_245 GMI_ES01_c12849_484 GMI_ES01_c12844_488 GMI_ES01_c10147_214 GMI_ES15_c10756_401 GMI_ES01_c10147_214 GMI_ES15_c10756_401 GMI_ES01_c10147_214 GMI_ES15_c10756_401 GMI_ES01_c3688_477 GMI_ES01_c3688_477 GMI_ES01_c2911_072 71.1 -71.4 -72.0 -72.3-72.6 72.9 73.1 73.2 73.4 73.6 74.3 75.7 76.4 77.1 GMI_ES_CC14804_238 GMI_ES01_c791_1072 GMI_ES01_c15886_573 GMI_ES17_c12067_507 GMI_ES17_c2656_146 GMI_ES02_c2554_426 GMI_ES17_c3370_293 GMI_DS_CC6823_75 GMI_ES01_c2481_1101 78.0 -83.0 -86.4 92.2 108.4 121.9 -126.0 -

5C

Figure 3.3. Conserved segments between LG 05021-5, and oat chromosome 5C.

05021-6

0.0 J GMI ES01 c12222 428	
6.0 - GMI ES22 c13166 513 GMI ES01 c17867 331	
23.7   HGMI ES02 Irc12751 147 GMI DS CC10035 89	GMI_ES01_c3132_376 ر
24.2 - GMI ES02 c4510 183	3.3 GMI_ES17_c11186_555
24.7 - GMI GBS 95496	12.1 GMI_ES15_c8803_430
26.4 M GMI ES LB 9861	15.4 - GMI_ES02_c15952_348
IGMI ES03 Irc23007 427 GMI DS LB 4510	17.5 - GMI_ES02_c28827_474
28.6 GML ES22 c2572 214 GML DS CC 9093 95	22.0 - GMI_DS_oPt-14552_101
GMI DS CC7847 62	22.6 - GMI_SNP5153_1
29.1 JU GML GBS 18001	23.5 - GMI_ES15_c10866_209
IGMI ES02 Irc18844 168 GMI GBS 12492	24.8 GMI_SNP_Irc14030_1
GMI ES02 c34690 199 GML DS LB 1821	25.3 GMI_ES02_c23210_257
GML GBS 720 GML ES17 c5666 258	26.6 GMI_DS_CC1776_314 GMI_DS_CC7847_62
30.2 GMI ES02 c1481 139 GMI GBS 66708	20.0 GMI_ES02_c34690_199 GMI_DS_CC9093_95
GML GBS 109228 GML DS CC1776 314	26.9 GMI_ES15_c1951_371
GML_ES15_c10332_306_GML_ES05_c6476_469	GMI_ES17_c5666_258 GMI_ES17_c2308_1026
42.6 - OPt-11380	27.3 GMI_DS_CC9448_443 GMI_ES01_c8337_431
LIGHLIGBS 6566 GMI ES17 c2308 1026	GMI_ES01_c3899_470
45.7 GML DS CC9448 443	28.4 GMI DS CC10035 89
AUGMIES01 c14139 498 GML GBS 24059	29.1 GMI ES02 c349 407
46.3 GML ES01 08337 431 GML ES01 03899 470	29.4 - EV GMI ES01 c14139 498
MUGMLES01_00007_401 GML_501_00005_470	29.6 - 🗮 - GMI ES02 c17191 397
46.9 46.9 46.9 46.9 46.9 46.9 46.9 46.9	29.8 CM GMI DS CC7795 77
47.5 MICHIES CC12/51_340 GWIES01_C17064_407	30.2 / SGMTES CC9431 260 GMI ES02 c8298 599
47.5 - 47 GMI_ESUS_C2934_220 GMI_ESTS_C5555_651	31.7 GMI ESO1 c17064 407
52 0 JUL 4 CMI E 502 015952 348 CMI E 502 028827 474	46.1 - GMI ES15 c14048 223
53.9 41 11 CML ES CC4628 110 CML CBS 51052	46.5 / GMI ES02 c2118 202
61.0 TH THIGHT ES_CC4636_THEGML_GD5_51955	48.3 GMI ES17 c327 972
65.7 - W GWI ES05_C11465_76 GWI ES02_C2116_202	58.4 GMI ES17 c5637 152
64.8 1/ GMI_ES 15_010645_554 GMI_ES05_01/96_425	
00.2 J	61.1 GMI EST7 c21168 588
90.3 1	61.2 GMI ES01 c2163 409
102.0 M GML ES LB 4976	61.3 - GMI_ES_CC17866_90
100.3 M GML ES14 c453 304 GML ES01 c5633 477	63.4 - GMI_ES17_c4515_297
100.8 1 M GML ES03 c11517 450	63.7 GMI_ES02_Irc34335_153
114 A MENGMEDS LB 5045 GML ES22 c2410 401	63.9 - GMI_ES_CC7312_286 GMI_ES02_c7632_364
1166 JI M GML DS LB 10336	68.5 📲 📲 GMI_DS_CC5652_132
117.7 JL XM GML ES14 (5483 345	72.8 GMI_ES01_c5633_477
1100 M GMLES 22 (5422 320	73.3 GMI_ES15_c3806_357
133.0 - GML ES05 c2253 434	76.4 BM_897a
120.6 M GML ES01_014606_61	77.4 GMI_DS_A3_435_344
140.9 L GML ES02 c310 458	77.9 - GMI_ES02_c247_241
141.4 - GML ES LB 9771	84.2 GMI_ES02_c4066_165
144.4 GMI ES01 Irc9147 285	84.3 GMI_ES17_c10053_867
151.1 J GMI ES01 c1913 998 GMI ES03 c1394 130	84.6 GMI_ES01_c14065_159
101.1 10.MI_E001_01810_880 0.MI_E000_01084_100	88.4 GMI_ES15_c858_507
	88.5 GMI_ES01_c24478_209
	95.4 J GMI_ES15_c1697_192

Figure 3.4. Conserved segments between LG 05021-6, and oat chromosome 6C.

6C

#### 05021-8.1

139.5 -150.8 -152.0 -153.8 -165.4 -170.2 -171.9 -

oPt-12964 oPt-0373 oPt-14526 oPt-9329 oPt-13092

ר 0.0		GMI_ES17_c18602_497	
3.9 -		/ GMI_ES22_lrc15031_170	
11.0 -\\	//	# GMI ES05 c8792 292	
12.1	#1	//₀GMI ES14 c15814 593 0	ו.0 ן GMI_ES15_Irc19149_99
12.6	AW	GMI ES03 c11741 332 10	0.0 GMI_ES01_2320_569 GMI_ES17_c9652_803
14.8	$\square \mathbb{Z}$	GMI GBS 13607 10	.1 GMI_ES_CC15237_134
297	ĦV	JGMI ES02 c16427 394 GMI DS LB 6890 10	.2 📲 🗰 GMI_ES15_11428_323
20.1	H/	IGMLES02 c17321 600 GMLES02 c15926 519 10	.6 M BM_907
30.2 -\	//	/ GMI_ES17_c11418_547 14	.9 GMI DS CC9972 92
212-	Ľ.	- CML ES14 c11707 371	.1 √ CMI DS CC8640 82
242	FK		.2 - GMI ES01 c24051 67
34.2 -	Ц.	~ CML ES14 c19247 456	3 - GMI DS CC8923 203 GMI ES01 8015 409
41.1 -		- GIVII_ES14_010000_440	3 GMI ES02 c16344 816
- 03.0 J	V	COMILES 14 C12990 449	6 GMI DS CC10461 118
57.4 ~	□-	GMI_ESUS_C3002_595 GMI_DS_LB_176	GML ES CC9711 432
500 /		IGNI ES05_CT1765_564	GML ES02 c14642 662
59.0 -//		VIGNI ESUZ C16344 816 GMI ESUZ C16019 318	E CML ES02 C11842 367 CML ES01 c10022 104
60.6		VGMI_ESU1_C10526_328 GMI_DS_LB_1611 V 20	1.5 -1 - 11 <u>GMI_E302_C11042_007</u> GMI_E301_C10035_104
64.5 ⁻ //	= 8	YGMI_ES_LB_12000 GMI_DS_LB_1773	GMI_ES17_C2941_220 GMI_ES02_C2898_314
65.1	18	GMI_DS_LB_1454	16 17 - GIVI ESUT IC28/11 407 GIVI ESUT C16239 145
66.2		GMI_ES15_C11400_361	GMI_ESU1_C8817_242 GMI_ES17_C1115_127
69.2		GMI_GBS_111982 21	.4 - GMI_ES01_C9236_115
70.9	1 108	MGMI_DS_LB_920 GMI_ES13_c3498_934 40	.4 - M GMI_ES02_c38800_418
71.4	0	GMI_DS_LB_919 46	6.6 M GMI_ES17_c5762_827
719		GMI_ES15_c5920_476 GMI_ES17_c20393_596 53	.4 - GMI_ES15_c285_271
-  I.	H١	IGMI_ES01_c24747_317 61	.6 1 H GMI_ES02_c15926_519
74.2		oPt-5427 61	.7 - GMI_ES17_c15132_382
75.3 -	-1	oPt-5729 62	.0 1 GMI_ES17_c11418_547
84.5 -//		GMI_ES03_c695_117 62	.4 GMI_ES15_c5280_161
116.7	=	GMI_ES01_c13259_737 63	.5 GMI_ES01_c10387_303
120.0		GMI_ES17_c29_442 68	.2 GMI_DS_A3_489_351
120.5		GMI_ES01_c8085_365 87	'.1 ' GMI_ES15_c2709_405
120.6		GMI_ES02_c14767_300	
120.7 -		² GMI_ES17_c8136_792	
120.8	N	GMI_ES02_c345_484	
120.9	$\sim$	GMI_ES02_c3243_375	
121.0		GMI_ES_LB_9797	
121.1		GMI_ES01_c4774_155	
121.2		GMI_ES14_c19637_237	
121.3		"GMI_ES14_c2964_319	
ſ		GMI_ES17_C18781_74 GMI_ES14_C8070_421	
Į.		GMI_ES01_Irc17667_357 GMI_ES17_c2595_383	
	- 5	GMI_ES03_c18978_181 GMI_ES22_c9164_352	
		IGMI_ES01_C11895_354 GMI_ES02_C13433_326	
121.8		'GMI_ES02_c4080_192 GMI_ES05_c16865_227	
		GMI_ES14_C1477_192 GMI_DS_CC11622_247	
1		IGMI_ES22_c16986_// GMI_ES17_c528_116	
L L		GMI_ES01_C10641_511_GMI_ES02_C35475_97	
		IGMI_ES01_IC10/25_434	
122.9 -		GMI_ES14_0001_94	
124.0		GMI_ES02_C3872_668	
128.7 -		IGMI_DS_LB_5780 GMI_ES01_018988_143	
129.2		IGMI_ES15_c3025_170 GMI_GBS_20451	
132.6		GMI_ES22_c4593_1054	
137.3 -		<u>GMI_DS_CC8923_203</u>	
		GMI_DS_LB_10677 GMI_ES01_Irc28711_407	
		GMI_ES01_c16239_143 GMI_ES_CC9711_432	
		GMI_ES01_c8817_242 GMI_ES02_c2898_314	
- 1		GMI_ES03_c9962_935 GMI_ES03_c22699_353	
138 /		GMI_ES22_c1771_507 GMI_ES01_c9464_550	
100.4		GMI_ES02_c22898_583 GMI_DS_LB_9592	
	]	GMI_GBS_110184 GMI_ES02_c11842_367	
		GMI_DS_LB_5749 GMI_DS_LB_10540	
I		GMI_ES17_c1033_262 GMI_ES02_c14642_662	
1		IGMI_DS_LB_4619	
139.5 -		GMI_DS_CC10461_118	
150.8 -		oPt-12924	
152.0 -		oPt-12964	
4500		- D1 0070	

**8A** 

Figure 3.5. Conserved segments between LG 05021-8, and oat chromosome 8A.

## 05021-12

12D

			0.0 1	「GMI_DS_oPt-16541_253
0.0 -	0Pt7556		14.4	GMI_ES01_c4452_325
521	oPt-17014		25.4	GMI_ES01_c27024_157
21.4	GMI DS LB 5997		25.7	GMI_DS_0PT-17694_374
32.6 -	GMI_ES02_c25243_77		39.5	IGMLES01_010372_615_GMLES15_08103_525
35.9 -	GMI_GBS_99439 GMI_ES02_c38462_575		40.6	GMLES01_024656_105
37.0 -	GMI_DS_oPt-17670_78 GMI_ES05_c14477_258		41.0	GML ES02 14559 362
38.6 -	GMI_ES05_Irc16479_180 GMI_ES05_c23907_190	)	41.4	GMI ES02 c5884 318 GMI ES15 c15399 80
00.0	I <u>GMI_ES17_C12958_273</u>		43.9	GMI ES CC7433 103
39.1 -	GMI_ES03_C3230_663		44.5	GMI_ES01_c16275_104
20.0	GMI_GBS_52290 GMI_ES14_C14567_105		47.5	GMI_ES_CC11621_230
- 39.0 T	CML CRS 00299 CML ES17 02477 95		47.8	GMI_ES02_c7694_423
45.4	GML E S17 c12536 430		48.1	GMI_DS_oPt-18005_248
46.5	GML DS LB 1859 GML ES02 (8737-267		51.5 1	GMI_ES02_c13596_178
53.6 -	GMI_ES03_c5662_209		53.8 1	GMI_ES17_C20660_396
54.7	GMI E \$02 c8277 506		56.0	CMLES13_0373_311
58.0	GMI GBS 100951		57.2	GMLES17_024091_200
58.5 - 🖓	GMI_ES15_c5831_418		61.1	GMI ES01 c16437 198
59.6 -	GMI_ES03_c2612_410		64.2	GMI ES01 c26749 466
	GMI_ES05_c7724_591 GMI_DS_LB_1354		66.3	GMI ES CC6818 106
ר 61.2	GMI_ES17_c3134_489 GMI_ES02_c22830_111	,	66.5	GMI_ES_CC11253 379
1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 -	(IGMI_ES15_C5554_3/7	/	66.6	GMI_ES02_c16987_268
62.3 y	GMI_ES02_C2959_310 GMI_ES02_C4007_487	s /.	66.9	GMI_DS_oPt-17084_293
GE O L	GML_GDS_00000		67.9	<u>GMI_ES_CC11933_314</u>
65.5	JGMLES LB 12070 GMLDS LB 10630	N //.	68.7	GMI_DS_CC5975_93
66.6	- GML GBS 90968		71.4	/ GMI_DS_0Pt-13898_690
	GMI ES05 Irc12441 760 GMI GBS 72768		72.3	- GMI_ES02_012/37_306
67.1 -/	GMI ES17 c18901 119	$\cdot$	85.9	AGMI_ES_CC6673_460 GMI_ESUT_IC12212_420
68.2 J	GMI_ES02_c15462_413	$\sim \times \infty$	91.3	- GML ES17 (7306 1002
85.3	GMI_ES17_c12269_176	- XXX	92.0	GML_ES15_c3200_563
104.5 -	~ GMI_GBS_78545		93.5	GMI ES CC8499 229
105.0 -/日	GMI_DS_LB_2922		94.0	GMI ES02 c9794 269
107.7 -//	GMI_ES22_c10434_196	1 //	102.9	GMI_ES01_c21239_190
110.4	CML_ES05_C1719_239	- ///	104.0	GMI_ES17_c15573_481
110.5	GMI_ES03_012156_400	////	104.5 (	<pre>\GMI_ES02_c6762_308 GMI_DS_CC5683_441</pre>
113.0	IGMI_ES14_c1133_505_GMI_DS_LB_10723	////	104.7	GMI_ES17_C12958_273
121.7	GMI DS LB 6430 GMI ES14 c1791 574		106.1	GNI ES15 C5004 3/1
	GMI_ES15_c2518_589	1///	106.71	GMI_EST7_C1305_210
122.4	IGMI_ES05_c4098_426 GMI_ES02_c3556_502	7/ 1	107.0	GMI_ES02_c2959_310
123.8	GMI_GBS_94839 GMI_ES05_c8628_73	71	107.5	GML ES01 Jrr 19619 275 GML DS oPt-12215 45
126.0	GMI_ES14_c5917_794	• [	107.6	GMI DS CC11787 243
126.5	GNI_ES_CC7433_103		109.5	GMI DS 0Pt-14907 38
128.7	IGMI_GBS_107339 GMI_ES05_010102_262		109.6	GMI_ES02_c3044_164
129.2	GML_ES17_c24_198	/	112.3	GMI_ES02_c15462_413
138.1	oPt-0553	4	114.0	·GMI_ES15_c1855_452
146.8	oPt-13898		115.0	<u>GMI_ES17_c12269_176</u>
147.4 -	oPt-0133		119.6	GMI_DS_CC9493_63
159.2 -	oPt-15688		120.1	GMI_DS_CC2783_160
168.0 -	GMI_ES02_c7694_423		121.7	GML ES15 c12355 287
168.6 -	IGMI_ES17_c7973_279 GMI_ES_CC11933_314		124.8	GML ES17 c14126 105
174.9 -	GMI_ES05_c13908_354		133.1	GMI DS oPt-18064 151
175.5 -	GMI_ES_LB_11007 GMI_ES01_lrc19721_107			
177.1	IGNILESUZ_010907_390			
1816	GMI_E303_022043_430 GMI_E302_016367_266			
101.0	IGML ES15 c12065 114 GML ES01 c16437 198			
189.4 -	GML DS LB 10822 GML ES15 c5432 126			
190.5 -	GMI ES17 c19330 272			
208.6 -	GMI_ES14_c347_1030			
209.1	GMI_DS_LB_6423 GMI_DS_LB_9683			
224.2	GMI_DS_LB_6395			
225.3 ^J	<u>GMI_DS_oPt-17694_374</u>			

Figure 3.6. Conserved segments between LG 05021-12, and oat chromosome 12D.

#### 05021-16

	•	
Т	n	Δ
	v.	

		014 50// 0500 57/	0.0 נ		GMI ES15 c4222 543
0.0		GMI_ES14_c2532_5/1	3.3	1	GMI_ES17_c10073_640
1.1		GMI_ES_LB_11151 GMI_ESU1_C235_61	10.8		GMI_ES17_c20215_324
1.0		GMI_ES02_019030_120	14.7		GMI_ES_CC12708_442
J.2		GML ES03 Irc9794 213 GML ES17 c11967 396	15.8		GMI_ES17_c17558_304
5.0 -		GMI E S02 c16349 294	16.1		GMI DS CC4394 195
		GMI ES14 c940 534 GMI ES17 c9625 419	10.01		GMI_ES17_C1779_044
5.6 -		GMI_ES03_c15150_160 GMI_ES17_c5367_259	17.5		GMI_ES02_c6920_143
1		IGMI_ES_LB_4286	18.0		GMI ES02 c20648 208
19.4 -		GMI_ES02_c15898_126	18.1		GMI ES17 c12516 818 GMI ES01 c8716 268
í		GMI_ES_CC8503_89 GMI_DS_LB_8383	18.2 -		GMI_ES15_c7719_163
		GMI_ES02_C23814_94 GMI_ES14_C11471_199	19.2		GMI_ES01_c13907_104
		GMI_ES15_06130_326 GMI_ES01_IC22977_595	19.3		GMI_ES15_c5905_473
19.9		GML_ES11_C397_1001 GML_ES22_C6670_76	20.3		<u>GMI_ES01_c18017_440</u>
- 1		GMI_C322_C4307_370 GMI_C310_C110_C40	20.6		GMI_ES17_0968_903
		GMI E \$15 c2041 605 GMI E \$03 c9245 234	21.6		GMI_ES01_07970_395
		GMI ES02 c12776 148	22.1		GMI_ES02_00055_242 GMI_ES02_020676_946
27.0 ₁	LI	GMI_ES14_c1865_384	23.3		GMI_ES17_c9257_328
27.5	1	GMI_ES05_c11419_658 GMI_ES17_c18708_336	23.6		GMI ES15 Irc19562 699
21.0	1 1	GMI_ES05_c10905_124 GMI_ES_LB_8449	24.5		GMI_ES15_c19227_114
28.6 1	Q 📙	GMI_ES02_012535_326 GMI_ES05_01760_188	24.6		GMI_ES17_c2699_441 GMI_ES17_c2356_160
30.81		GML DS CC4430 239	24.01		IGMI_ES02_c14677_456
35.8 -		GML ES15 c7380 296	24.8 1		<u>GMI_ES17_C5590_702</u>
39.1	1	GMI ES LB 11832	20.21	( <mark> </mark>	GMLES02_03206_293
41.3	1 <mark>1</mark>	GMI E S17 c4241 356	29.01	1 I	GML ES02 c11702 476
45.8 -		GMI_ES01_c13021_254 GMI_ES22_c10495_451	\$ 31.7 ·	(e)	GML SNP2043 1
46.9		GMI_ES22_c7349_315		( <b>_</b> )	GMI ES02 c1538 477 GMI ES CC18060 346
62.0		GMI_GBS_31651 GMI_ES03_c7197_681	35.8	V <b>I</b> V	GMI ES01 c6696 348
62.5 -		GMI_GBS_23580	36.6		GMI_ES_CC2716_392
63.6 T	l	GMI_ES15_C/069_237	. 38.9	N <b>e</b> i	GMI_SNP_Irc40347_1
020-	14	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	42.1-	⋽	- GMI_ES17_c8741_79
85.7 1	A <b>I</b> K	GML E \$17 c12516 818	43.8	Т	- <u>GMI_ES02_C15898_126</u>
98.2 1	N 18	GMI ES17 c20215 324			~ GIVILES17_C3846_396
104.7		GMI ES05 c10300 165 GMI DS CC4394 195	52.5		CML ES CC13348 03
	0-1/	GMI ES02 c6920 143 GMI ES14 c197 108	54.2		MGML ES15 c7722 409 GML ES15 c900 850
		GMI_ES05_c15948_428 GMI_ES15_c5368_259	//////		GMI ES02 c21402 61
ر 105.2 م		GMI_ES05_c9397_421 GMI_ES03_c16835_129	•////	IM I	GMI ES15 c10811 217 GMI ES CC8503 89
	∖ - /	GMI_ES03_c14702_225 GMI_ES15_c1060_702	·/// /// ECO	i i	GMI_ES15_c713_848 GMI_ES15_c2041_605
400.0	X	IGMI_ES05_c20848_84 GMI_ES02_c6920_272	J0.2		GMI_ES01_Irc22977_595 GMI_DS_CC8733_129
106.3		GMI_DS_LB_2849	¥ / // )		<u>GMI_ES17_c5367_259</u>
106.0 -		GMI_CBS_81931_GML_ES17_c15455_93	/// 56.4		GMI_ES17_c4475_475
109.7 ^J	8 1	GMI ES01 c13907 104 GMI ES03 c1096 257	/// 56.5		IGMI_DS_CC4575_55 GMI_ES17_C9625_419
111.8	i <b>- 1</b>	GMI ES14 c1929 383 GMI ES15 c7106 329			GMLES17_C1066_770
112.8	i 🖃	GMI ES02 c1493 323 GMI ES01 c18017 440	57.0		GMLES CC14000 280
114.4	1 - Y	GMI_GBS_57847	// 58.2		GML ES15 c2802 625
115.5-	1 5	GMI_ES05_c23665_336 GMI_DS_LB_4140	// 58.8		GMI ES02 c8034 282
110.0	۲ <mark>س</mark>	GMI_ES02_c8953_242	// 59.0		GMI_ES01_c8899_324
117.1 -	2	GMI_GBS_/1/51	// 59.6		GMI_ES_CC6497_157
118.21		GML_GDS_99936 GML_ES05_C12170_315	// 59.8		GMI_ES02_c2923_535
122.0		GMI_GBS_4508	60.4		GMI_ES02_06507_520
133.9	i E i	oPt-14033	61.0	1 1	CMLES17_03200_273
140.6	. 1	oPt-5564	61.8		GML ES17_01612_641
146.3		GMI_ES02_c20364_439 GMI_ES_LB_11858	65.8		M38721-1
	11	GMI_ES02_c21087_487 GMI_ES03_c2344_498	67.0		AF237553-1-2
148.0 -		GMI_ES03_C9714_720 GMI_ES15_C7254_710	68.6		GMI_ES15_c10509_256
Í	11	IGMI_DS_LB_0041 GMI_ESU1_0401_022	70.9		BM_912a
150.2		GMI_ES22_Irc17882_62_GMI_ES17_c780_489	74.1		GMI_ES17_c5169_555
		GMI E \$17 c5590 702	(4.7		GWI_ESUT_C17040_394 GMI_DS_CC4430_239
151.4 -		GMI_GBS_99493	74.9		GML DS 0Pt-15681 90
156.7 -		oPt-18007	76.2		GML ES01 c1287 580
160.8 -		oPt-0003	76.5		GMI ES02 c12598 260
169.2 -	1 1	oPt-15376	80.8		GMI_ES17_Irc7334_312 GMI_ES17_c4241_356
1/4.9		GML ES03 c12150 403	Q1 C .		GMI_ES01_c284_1036 GMI_ES17_c2122_619
166.0 1		GMI_ES03_C12109_495	01.0		GMI_ES17_c2063_243
195.0		GMI_ES02_c27935_359 GMI_ES02_c41019_526	85.6		^L BM_183a
100.0		IGMI ES17 c9934 427			
107.0		GMI_ES05_c4422_582 GMI_ES02_c20648_208			
197.3		GMI_ES22_c19289_86 GMI_DS_LB_7758			
198.8 -		GMI_ES03_c9203_225			
198.9 -		GMI_ES05_c14456_70			
199.0 -		GMI_GBS_81937			
199.1 -	I	GWI_ESU1_03388_616 GML_ES15_02738_176			
199.2 1		GML ES01 c21179 347			
202.0		GMI ES15 c7233 246			
205.6 -		GMI_ES_CC4005_62 GMI_ES02_c1538_477			
208.6		GMI_ES05_c2682_281			
219.5		^L GMI_ES15_c10847_268			

Figure 3.7. Conserved segments between LG 05021-16, and oat chromosome 16A.



Figure 3.8. Conserved segments between LG 05021-18, and oat chromosome 18D.

	0.0 , (GML DS oPt-13151 665	
	2.0 GMI ES02 c14927 478	
	7.5 GMI_ES01_c14349_357	
	13.1 GMI_ES01_c19245_178	
	19.3 GMI_ES01_c507_760	
	28.6 · GMI_ES15_c12694_287	
	30.0 · GMI_ES17_c2454_883	
	39.5 · GMI_SNP5469_1	
	39.7 GMI_ES15_c6155_291	
	40.8 GMI_ES17_c1912_929 GMI_ES01_c13342_3	01
	43.4 GMI_ES01_C19628_354	
	43.7 GML_ESUT_ICT2090_96	
0.0 3 GMI_ES02_Irc13785_433 GMI_ES_LB_11028	44.2 GMI_ES17_C3195_666	
IGMI_ES_LB_11026	47.2 GML ES CC11076 204	
1.1 1 I GMI_ES05_65960_544 GMI_ES02_62449_552	47.6 GMI ES01 c18839 132	
1GWI_ESU2_IC38264_316	49.4 - GMI DS CC10481 156	
2.2 1 // GMI_ES_LD_0000	49.5 GMI ES02 c7768 318	
GML_ES02_Irc38280_700_GML_ES15_c6243_310	49.7 GMI_ES01_c27869_512	
GMI ES02 c14349 258 GMI ES03 c8567 90	49.9 GMI_ES_CC15389_69	
3.3 \ // GMI_ES01_c12979_488 GMI_ES05_c9412_222	50.7 GMI_ES02_Irc34335_153	
GMI ES17 c10189 970 GMI ES17 c5870 387 4	51.1 GMI_ES02_Irc12182_165	
GMI E S01 c677 223	51.3 BM_130D	
OR / GMI ES LB 11401 GMI ES01 c13113 119	51.4 GMI_ESU1_C12378_433	
^{3.8}	51.51 GMI_ES15_C10020_120	
14.6 - GMI_ES22_c10247_408	51.8 GML ES01_C796_180	
19.3 - OPt-16024	51.9 GMI ES CC15240 111	
37.6 GMI_ES01_c1793_450	52.6 GMI ES01 c10310 366	
54.0 1 V 0Pt-159/1	53.9 GMI ES15 c8191 415	
55.2 T P ( 0Pt-5224	54.2 GMI_ES02_c22225_492	
57.0 1 1 1 0 PE 10979	54.8 AM87-2	
64.1 JU V/ 0Pt-11426	55.2 GMI_ES_CC13848_210	
68.8 GMI ES22 c11596 73 GMI ES15 c12694 287	55.5 GMI_DS_0PT-15595_189 GMI_ES01_01511_1	1015
72.1 VIGMI ES22 c9203 288 GMI ES03 c10715 468	56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56.0 - 56	
74.8 - GMI_ES02_c20190_105	67.3	
76.4 - GMI_ES01_c1107_182	68.9 - GMI DS CC6556 81	
76.9 GML ES01_c19628_354	69.8 GMI EST5 c15790 334	
78.3 - GMI_ES22_C10946_366	70.7 / IGMI_ES15_c6102_442 GMI_ES17_c8412_39	8
80.5 - GMI_ESUS_C4458_502 GMI_GBS_25061	70.9 GMI_ES15_c11012_143	
81.2 GMI GBS 92025	72.8 W BM 692a	77
GMI ES01 c2436 405 GMI ES13 c14470 87	73.5 % GMI_ESU1_C25664_161 GMI_ESU2_C6764_2	
81.7 - GMI_ES21_c6485_431 GMI_GBS_10457	75.8 M GMI ES02 c4352 145	
GMI_ES15_c8569_380	81.4 GML ES02 c26937 168	
82.2 - GMI_ES16_c20195_287 GMI_GBS_100319	82.3 GMI DS CC885 419	
GMI_ES15_C17597_267_GMI_ES17_C7039_546	/ 84.5 GMI_ES02_c6122_167	
83.3 M GMI_DS_LB_9310 GMI_ES_LB_11958	87.7 GMI_DS_oPt-12991_114	
GMI_ES_EB_T1974 GMI_ES02_C1035_400	88.5 GMI ES02 Irc13785 433	
83.8 - GMI_ES22_c18838_446 GMI_ES_CC15240_111	91.6 GMI_ES02_c2449_552	
84.3 - M GMI ES03 c9122 309 GMI DS LB 950	97.4 J GMI_DS_CC6354_109	70
84.8 - GMI_ES02_c16381_584	GML ES02 C14349 258 GML DS CC12000	73
85.0 GMI_ES_LB_11633 GMI_GBS_96826	97.5 GML ES15 (5810, 415 GML ES17, 5870, 38	7
IGMI_ES02_c3016_666	GML_ES15_C394_171	
93.3 1 GMI_ES14_c118_502	IGMLES17 c2598 325 GMLES15 c9521 56	1
109.1 1 IGMI_ES05_C2066_503 GMI_ES22_C4463_275	97.7 GMI_ES15_c2046_330	
114.3 GMI_GBS_3323	98.0 GMI_ES02_c12488_534	
135.0 J GMI ES01 c27869 512 GMI GBS 80511	98.8 GMI_ES_CC15133_247	
100.0 1 <u>0/01_02/000_012</u> 000_000_00011	99.6 GMI_ES17_c8066_237	
	100.7 BM_74	
	104.2 GMI_ES02_C631_591	
	106.3 GMI_DS_CC3275_96	73
	100.0 10001_L302_010002_290 GMI_E302_07238_4	10
	109.4 GMI_ES02_Irc18911_190	
	115.4 GMI_ES_CC11271_65	
	115.5 GMI_ES02_c12761_183	

Figure 3.9. Conserved segments between LG 05021-19, and oat chromosome 19A.



Figure 3.10. Conserved segments between LG 05021-4, and oat chromosome 4C.

## 3.5.1.2. Population 05026 and oat consensus map

171 markers were shared covering 647.3 cM in population 05026 (55% of the total map), and 552 cM in the oat consensus map (30% of the map). Marker order within the syntenic segments was well conserved on linkage groups 05026-3, 05026-4, 05026-5.2, 05026-11, 05026-10, 05026-14.2, 05026-19, and 05026-20.1. The same order was observed, but in reverse, for linkage groups 05026-1.1, 05026-5.1, 05026-6, 05026-12.1, 05026-14.1, and 05026-15. On LG 05026-1.2, marker ES15_c5908_278 switched order with marker ES15_c6153_392 (Figure 3.11, red). Three small inversions were observed in LG 05026-2. Marker ES15_c7706_583 located at the end of LG 05026-2 (position 61.3 cM) had been mapped at the beginning of oat chromosome 2C (Figure 3.12, red). Cosegregating markers DS CC8468 91and ES CC11290 204 changed

order with other cosegregating markers ES01_c1635_353 and ES_CC8700_285 at position 5.8 cM for the former and 6.4 cM for the latter (Figure 3.12). Cosegregating markers ES17_lrc20311_707 and ES_CC3989_242 (Figure 14, blue) at position 0 cM changed order with a group of 10 cosegregating markers at position 0.6 cM (Figure 3.12, violet). Marker ES17_c5784_752 (Figure 3.15, blue) positioned at 45.3 cM in LG 05026-3 changed order with a group of 5 cosegregating markers at position 46.5 cM (Figure 3.13, red). Marker ES02_lrc16798_330 located at the beginning of LG 05026-13.1 had been mapped at the end of oat chromosome 13A (Figure 3.14, red). According to Singh and Kolb, 1991 the differences noted in the ordering of markers between the two populations and the oat consensus map are an indication that genomic rearrangements and intervarietal chromosome interchanges exist in the genome of cultivated oat.

## **3.5.2. Segregation distortion**

Twenty five percent of the markers tested for population 05021, and 9% for population 05026 showed significant distortion from the expected segregation ratio. Similar results to those obtained for population 05026 (Zhu and Kaeppler, 2003; Portyanko et al., 2001; O'Donoughue et al., 1995) and for population 05021 (Hizbai et al., 2012; O'Donoughue et al., 1992) had been previously reported in other mapping studies. Zhu and Kaeppler, 2003 reported 9% of segregation distortion in the Ogle/MAM 17-5 mapping population, Portyanko et al., 2001 reported 13% in the Ogle/TAM O-301 population, and O'Donoughue et al., 1995 reported 8% of segregation distortion in the Kanota/Ogle population. Hizbai et al., 2012 found out that 27% of the markers in the Dal/Exeter mapping population exhibited segregation distortion, and O'Donoughue et al., 1992 reported 19% of distortion in the diploid map *A. atlantica* x *A. hirtula*.



1C

Figure 3.11. Conserved segments between LG 05026-1.1, 05026-1.2, and oat chromosome 1C.



Figure 3.12. Conserved segments between LG 05026-2 and oat chromosome 2C.

05026-3

2	^
J	L

		0.0.1	GML ES17 (3320-786
0.0-	CMI CR8 26225	1.7	GML ES02 c10749 342
70	CMI_GD5_20020	11.5	GMI ES01 c11827 414
16.0	GMI_L303_C12033_243	12.4	GML ES_CC5758_340
20.7	GML_ES15_c1433_87	13.7	GMI ES01 c2827 649 GMI ES17 c125 448
23.0	GMI_ES13_C1435_07	14.4	GMI ES15 c605 638
25.1 -	GML_CBS_69683	15.7	GMI DS CC1149 344
26.3 -	GML_ES03_c3525_308	18.3 -	GMI DS CC10993 70
20.0	IGMI_DS_LB_7523_GMI_DS_LB_8152	18.9	GMI ES CC15057 51
26.9	GMI_ES03_c13250_560 GMI_ES01_c11827_414	19.3 -	GMI DS CC5134 229 GMI ES01 c461 1288
28.8	GML E \$15 c605 638	10.0	GML ES17 c2398 610 GML ES_CC3491 422
45.4	GML ES05 c5558 288 GML ES17 c5784 752	י 19.8	GMI ES17 c10984 690
-0	GMI DS CC10993 70 GMI GBS 84241	19.9	BA grs c8269 158
	GML ES01 c461 1288 GML ES CC15160 63	20.7	GMI ES15 c1806 90
	CMLES CC15057 51 GMLDS LB 2002	20.9	GMI ES17 c5784 752
46.5 r	GML DS_CC5134_229 BA_grs_C8269_158	21.1	GMI ES02 c13608 538
H H	GML ES14 c14040 375 GML ES05 c20438 78	22.2	GMI DS CC8554 381
- N-	IGMI_ES05_c11113_348_GMI_GBS_7855	24.2	GMI DS CC5751 111
40.4	GML GBS 113531	- A	GMI ES01 c10120 303 GMI ES01 c23001 372
58.7	IGMI_600_110001	25.5	IGMIES CC8805 161
67.3	GMI_GBS_13773 GMI_DS_0Pt-18257_376	28.1	GMI ES CC14261 141 GMI ES01 c11741 182
67.0	GML ES01 (8255 502	32.1	GMI ES01 c109 982
69.6	GML ES02 c10035 217	, 39.9 -	- GMI_ES15_c6870_218
70.2	GML_ES02_c24184_389	<b>44.0</b>	- <u>GMI ES02 c1819 259</u>
72.5	W GMI ES14 c8386 549	.49.5	- GMI_ES01_c20367_331
73.1	GMI_ES22_c526_367	51.8	GMI ES01 In: 22461 468
73.7	GMI ES02 c1819 259	53.2	GMI ES CC8697 53
762	GMI GBS 23770	/ / 🗴 53.5 🥼 🛋	GMI ES17 c16539 472
80.8	/ GMI ES05 c10478 455	/ / <b>54.7</b> 📲	GMI_ES17_Inc19617_111
831	/ GMI_GBS_102428 GMI_ES14 c16101_333	/ / 54.9	GMI_DS_CC1800_254
84.2 -	GMI DS LB 6241	57.5	<u>GMI_ES17_c4051_315</u>
87.7 -	GMI ES14 c18975 277	/// •59.2 📜	GMI_ES15_c7272_387
	GMI_ES17_c16539_472 GMI_GBS_101811	// / 59.4	GMI_ES01_c14397_365
00.3 -	\]GMI_DS_LB_10786 ///	// / 59.7	GMI_ES02_c17906_415
92.5	YGMI_ES02_Irc27323_95 GMI_GBS_6872 / //	65.1 <b>(</b>	GMI_ES01_c13233_204
1110	]GMI_ES17_c4051_315 GMI_GBS_79375	/ 66.5	<u>GMI_ES_CC7714_103</u>
····.2	IGMI_ES03_c5837_61	/ <u>66.8</u>	- <u>GMI_ES17_c13962_600</u>
	GMI_ES12_c8736_490 GMI_ES22_c9270_194	/ 68.1	IGMI_ES17_c4821_368 GMI_DS_CC11154_271
114.2	GMI_ES01_c9377_521 GMI_GBS_39204	69.4	GMI_ES02_c12942_675 GMI_ES01_c17319_464
	GMI_DS_LB_7069	71.2	GMI_ES_CC11028_196
114.9 -//	GMI_DS_LB_10721 /	74.2	GMI_ES02_c841_728 GMI_ES01_c22540_100
116.3	GMI_GBS_93278 GMI_GBS_76760	74.8	· <u>GMI_ES01_c16727_290</u>
	GMI_ES05_c24963_586 GMI_ES22_c2904_356 /	75.7	<u>GMI_ES15_c14533_341</u>
1160	GMI_ES17_C13962_600 GMI_ES03_C19505_223 ●	83.2 -	AB_STS_28
110.3	GMI_ES02_c21260_142 GMI_ES_CC7714_103	86.6	BA_grs_c10318_236
	IGMI_DS_CC11154_271 GMI_ES15_c32_761	87.7	GMI_ES17_c8729_764
117.5	GMI_ES05_c1323_286 GMI_ES05_c21329_243	93.5	GMI_ES_CC16445_119
	IGMI_ES02_c23166_443		
120.2	GMI_ES01_c16727_290 GMI_DS_LB_6609		
20.2	GMI_ES02_c12942_675		
, j	GMI_ES05_c9704_189 GMI_ES22_c2352_65		
121.3 -	GMI_ES02_c841_728 GMI_ES14_c982_713		
1	GMI_ES15_c14533_341		
123.0	GMI_ES05_c20576_219		
124.1	GMI_DS_LB_6383		
143.2 ^J	^L BA_grs_c10318_236		

Figure 3.13. Conserved segments between LG 05026-3 and oat chromosome 3C.

0.0-

28

0.0 1

9.2 -9.8 -

GMI_DS_LB_1958

GMI_ES05_c2494_581

GMI_ES15_c1630_786

GMI E S01 c5430 179

GMI_ES22_c408_595 GMI_DS_LB_6022

0.0 GMI_ES01_c12710_289 8.6 M38722-2-2 GMI_DS_A3_227_89 12.8 · 14.9 · M38722-1-2 16.8-AF237553-2 25.4 M38722 28.3 54.3 M38721-3-2 GMI_ES02_c17921_986 GMI_ES01_c27692_191 60.1 GMI_ES17_Irc11962_142 GMI_DS_CC4373_180 60.4-61.8 GMI_ES02_c27723_426 GMI_ES02_lrc20576_405 62.2 GMI_ES15_lrc19345_536 IGMI_ES15_III9345_305 GMI_ES17_III09623_475 GMI_ES15_c17486_204 IGMI_ES_CC9934_55 GMI_ES15_c6756_354 IGMI_DS_CC5684_99 64.6 65.1 65.3 IGMI_DS_CC5684_99 GMI_ES02_c14336_416 GMI_ES17_c19062_191 GMI_ES17_c1415_605 GMI_ES17_c3808_324 GMI_ES_CC7684_273 GMI_DS_A3_37_143 GMI_ES17_c1926_571 GMI_ES17_c1926_571 65.9 66.0 66.1 66.4 66.7 70.2 71.5 GMI_ES01_c8301_93 GMI_ES_CC13970_83 75.2 GMI_DS_CC7582_55 GMI_ES02_c14492_305 GMI_ES01_Irc22746_326 75.3 76.0 -GMI_ES01_c14084_302 GMI_ES01_c14084_302 GMI_ES01_c9768_191 GMI_ES02_c14497_585 GMI_ES17_c822_647 77.2 -77.7 -79.1 ⁻ 79.8 -80.9-GMI DS oPt-2385 166 GMI_ES15_c1630_786 GMI_ES17_Irc11645_145 GMI_ES01_Irc7807_649 837-85.1 86.0 .



Figure 3.14. Conserved segments between LG 05026-13.1 and oat chromosome 13C.

Specific regions of segregation distortion had been reported in the Kanota/Ogle population KO 6, KO 11, KO 24 (O'Donoughue et al., 1995), the Ogle/MAM 17-5 population OM 1, OM 3 (Zhu and Kaeppler, 2003), and the Dal/Exeter population (Hizbai et al., 2012).

13A

Those linkage groups showed homology with distorted regions reported in population 05021 and population 05026. KO 6 is homologous to 05026-13; KO 11, OM 3, and DE 7 are homologous to 05021-1.1 and 05021-16; KO 24 is homologous to LG 05021-16, and OM 1 is homologous to 05021-19. The presence of segregation distortion in the same region across different mapping populations supports the evidence of genes affecting distortion on those genomic regions. All the distorted markers in population 05021 were skewed toward only one parent. One possible cause could be inadvertent selection due to small population size during the development of the population 05021 (Kianian et al., 2001). Liu et al., 2010 reported that in RIL populations, a high proportion of segregation distortion may be due to artificial sampling and natural selection of many generations. Chromosomal microrearrangements related to introgression of alien segments carrying desirable genes from wild germplasm (Portyanko et al., 2001) and chromosome translocation (Liu et al., 2010) can be also the cause of segregation distortion. A major translocation, between chromosome 7C and 17, in oat affected mapping in other populations (Jellen and Beard, 2000). As spring-type oats (A. sativa), the parents used in this study are expected to have the same allele of this translocation, and consequently no extreme distortion was found on LGs 05021-7.1, 05021-7.2, 05021-7.3, 05021-7.4 homologous to oat chromosome 7C-17A. It is possible then, that other chromosomal rearrangements and minor translocations have influenced segregation distortion in population 05021. The distorted markers, in population 05026, were skewed toward both parents. The apparent preferential transmission of one parental genotype indicated that the regions in question may contain genes that affect gamete and/or hybrid viability (O'Donoughue et al, 1995; Zhu and Kaeppler, 2003).

## **3.5.3.** Map size and cosegregation

The map developed for population 05021 was significantly longer than the one developed for population 05026. The parents used to develop population 05026 are more closely related than those used to develop population 05021. One of the parents of population 05026 'Souris' is a sister line of 'HIFI' and the other parent 'ND030299' has 'HIFI' as a parent (McMullen, communication personal). The shorter map observed in population 05026 may be the result of inadequate polymorphism in some regions with similar ancestry (Hizbai et al., 2012). 487 markers polymorphic in population 05021 were also mapped in population 05026. The 1.5 fold reduction in recombination for regions detected by these markers is quite similar to the two-fold decrease in total length for the two maps. Similar ancestry in some regions, combined with reduced recombination is the best explanation for the differences between the two maps. The map size of the oat consensus map (1838 cM) was in between the two populations (Oliver et al., 2013). Similar map size had been reported for population 05021 (Jin et al., 2000) and also for population 05026 (Zhu and Kaeppler, 2003). Zhu and Kaeppler, 2003 and Kianian et al., 2001 indicated also that map size can be affected by mapping strategy and reduced recombination frequencies.

In general, 45% of the markers in population 05021 and 61% in population 05026 cosegregated with other mapped markers. This amount of cosegregation is much more than those reported in other oat map studies (Zhu and Kaeppler, 2003; Jin et al., 2001; O'Donoughue et al., 1995). The principal reason is that many of the EST markers turned out to be closely linked, because initially those markers were designed for wheat and barley (Howarth et al., 2013).

## **3.6.** Conclusion

In conclusion, DNA markers are useful in the construction of genetic maps of different cereal species (Heun et al. 1991; Messmer et al. 1999; Ramsay et al. 2000). Mapping populations obtained as a result of crossing two different homozygous parents are particularly useful in the process of creating these maps. Molecular breeding in oats has until recently been limited due to lack of available markers. The use of next-generation sequencing (NGS) for genotyping and comparative genomics has revolutionized the field of genetics by the introduction of sequence-based SNP markers. Recently a consensus map for hexaploid oats has been constructed using six mapping populations. Many of the markers placed in the two maps, reported in this study, are not in the consensus map. It is expected that some of the information provided here can be applied to improve the oat consensus map. The constructed maps can also be used in the mapping of quantitative trait loci for important agronomic traits in oats. Future studies should focus in the construction of a physical map for the hexaploid oat, and would constitute the first step in sequencing the oat genome.

#### **3.7. References**

- Dekoeyer, D.L., N.A. Tinker, C.P. Wight, J. Deyl, V.D. Burrows, L.S. O'Donoughue, A. Lybaert, S.J. Molnar, K.C. Armstrong, G. Fedak, D.M. Wesenberg, B.G. Rossnagel, and A.R. McElroy. 2004. A molecular linkage map with associated QTLs from a hulless x covered spring oat population. Theor Appl Genet 108:1285-1298.
- Heun M., A.E. Kennedy, J.A. Anderson, N.L.V. Lapitan, M.E. Sorrells, and S.D. Tanksley. 1991. Construction of a restriction fragment length polymorphism map for barley (*Hordeum vulgare*). Genome, 34: 437-447.
- Hizbai, B.T., K.M. Gardner, C. P. Wight, R.K. Dhanda, S.J. Molnar, D. Johnson, J. Frégeau-Reid, W. Yan, B.G. Rossnagel, J.B. Holland, and N.A. Tinker. 2012. Quantitative trait loci affecting oil content, oil composition, and other agronomically important traits in oat. Plant Genome 5:164-175.
- Jellen, E.N., and J. Beard. 2000. Geographical distribution of a chromosome 7C and 17 intergenomic translocation in cultivated oat. Crop Sci. 400:256-263.

- Jin H., L.L. Domier, X. Shen, and F.L. Kolb. 2000. Combined AFLP and RFLP mapping in two hexaploid oat recombinant inbred populations. Genome 43:94-101.
- Kianian, S.F., S.L. Fox, S. Groh, N. Tinker, L.S. O'Donoughue, P.J. Rayapati, R.P. Wise, M. Lee, M.E. Sorrells, G. Fedak, S.J. Molnar, H.W. Rines, and R.L. Phillips. 2001.
  Molecular marker linkage maps in diploid and hexaploid oat (Avena sp.) p443-462. In R.L. Phillips and I.K. Vasil (eds.) DNA-based markers in plants. Kluwer Academic Publishers. Netherlands.
- Liu, X., L. Guo, J. You, X. Liu, Y. He, J. Yuan, G. Liu, and Z. Feng. 2010. Progress of segregation distortion in genetic mapping of plants. Research Journal of Agronomy 4:78-83.
- Messmer M.M., M. Keller, S. Zanetti, and B. Keller. 1999. Genetic linkage map of a wheat ' spelt cross. Theor. Appl. Genet. 98: 1163-1170.
- Milach, S.C.K., H.W. Rines, and R.L. Phillips. 1997. Molecular genetic mapping of dwarfing genes in oat. Theor. Appl. Genet. 95:783-790.
- O'Donoughue, L.S., Z. Wang, M. Roder, B. Kneen, M. Leggett, M.E. Sorrells, and S.D. Tanksley. 1992. An RFLP-based linkage map of oats on a cross between two diploid taxa (Avena atlantica x A. hirtula). Genome 35:765-771.
- O'Donoughue L.S., M.E. Sorrells, S.D. Tanksley, E. Autrique, A. Van Deynze, S.F. Kianian, R.L. Phillips, B. Wu, H.W. Rines, P.J. Rayapati, M. Lee, G.A. Penner, G. Fedak, S.J. Molnar, D. Hoffman, and C.A. Salas. 1995. A molecular linkage map of cultivated oat. Genome 38:368-380.
- Oliver, R.E., N.A. Tinker, G.R. Lazo, S. Chao, E.N. Jellen, M.L. Carson, H.W. Rines, D.E. Obert, J.D. Lutz, I. Shackelford, A.B. Korol, C.P. Wight, K.M. Gardner, J. Hattori, A.D. Beattie, A. Bjornstad, J.M. Bonman, J.L. Jannink, M.E. Sorrells, G.L. Brown-Guedira, J.W.M. Fetch, S.A. Harrison, C.J. Howarth, A. Ibrahim, F.L. Kolb, M.S. McMullen, J.P. Murphy, H.W. Ohm, B.G. Rossnagel, W. Yan, K.J. Miclaus, J. Hiller, P.J. Maughan, R.R.R. Hulse, J.M. Anderson, E. Islamovic, and E.W. Jackson. 2013. SNP discovery and chromosome anchoring provide the first physically-anchored hexaploid oat map and reveal synteny with model species. PLoS ONE 8:e58068. doi:10.1371/journal.pone.0058068.
- Pal, N., J.S. Sandhu, L.L. Domier, and F.L. Kolb. 2002. Development and characterization of microsatellite and RFLP-derived PCR markers in oat. Crop Sci. 42:912-918.
- Portyanko V.A., D.L. Hoffman, M. Lee, J.B. Holland. 2001. A linkage map of hexaploid oat based on grass anchor DNA clones and its relationship to other oat maps. Genome 44:249-265.
- Portyanko, V.A., G. Chen, H.W. Rines, R.L. Phillips, K.J. Leonard, G.E. Ochocki, and D.D. Stuthman. 2005. Quantitative trait loci for partial resistance to crown rust, Puccinia coronate, in cultivated oat, *Avena sativa L*. Theor Appl Genet 111:313-324.

- Ramsay L., M. Macaulay, S. Degli-Ivanissevich, K. Maclean, L. Cardle, J. Fuller, K.J. Edwards, S. Tuvesson, M. Morgante, and A. Massari. 2000. A simple sequence repeatbased linkage map of barley. Genetics, 156: 1997-2005.
- Singh, R.J., F.L. Kolb. 1991. Chromosomal interchanges in six hexaploid oat genotypes. Crop Sci. 31:726-729.
- Slotta, T.A.B., L. Brady, and S. Chao. 2008. High throughput tissue preparation for largescale genotyping experiments. Molecular Ecology Resources 8:83-87.
- Tanhuanpaa, P., O. Manninen, A. Beattie, P. Eckstein, G. Scoles, B. Rossnagel, and E. Kiviharju. 2012. An updated doubled haploid oat linkage map and QTL mapping of agronomic and grain quality traits from canadian field trials. Genome 55 :289-301.
- Tinker, N.A., A. Kilian, C.P. Wight, K. Heller-Uszynska, P. Wenzl, H.W. Rines, A. Bjornstad, C.J. Howarth, J.L. Jannink, J.M. Anderson, B.G. Rossnagel, D.D. Stuthman, M.E. Sorrells, E.W. Jackson, S. Tuvesson, F.L. Kolb, O. Olsson, L.C. Federizzi, M.L. Carson, H.H. Ohm, S.J. Molnar, G.J. Scoles, P.E. Eckstein, J.M. Bonman, A. Ceplitis, and T. Langdon. 2009. New DArT markers for oat provide enhanced map coverage and global germplasm characterization. BMC Genomics 10:39. doi:10.1186/1147-2164-10-39.
- Voorrips, R.E. 2002. MapChart: Software for the graphical presentation of linkage maps and QTLs. The Journal of Heredity 93:77-78.
- Wight, C.P., N.A. Tinker, S.F. Kianian, M.E. Sorrells, L.S. O'Donoughue, D.L. Hoffman, S. Groh, G.J. Scoles, C.D. Li, F.H. Webster, R.L. Phillips, H.W. Rines, S.M. Livingston, K.C. Armstrong, G. Fedak, and S.J. Molnar. 2003. A molecular map in 'Kanota' x 'Ogle' hexaploid oat (*Avena spp.*) enhanced by additional markers and a robust framework. Genome 46:28-47.
- Zhu, S., and H.F. Kaeppler. 2003. A genetic linkage map for hexaploid, cultivated oat (*Avena sativa L.*) based on intraspecific cross 'Ogle/MAM17-5'. Theor. Appl. Genet. 107:26-35.

# CHAPTER 4. GENETIC ANALYSIS OF QUANTITATIVE TRAIT LOCI AFFECTING AGRONOMIC AND GRAIN QUALITY TRAITS IN TWO SPRING OAT POPULATIONS

## 4.1. Abstract

Grain yield, test weight, 1000 kernel weight, heading date, and plant height are major agronomic traits in cultivated oat (Avena sativa L.). On the other hand,  $\beta$ -glucan content, oil content, groat percentage and dehulling efficiency are major quality and grain physical traits. Information regarding quantitative trait loci affecting those traits would facilitate the development of oat cultivars with desirable quality. QTLs for agronomic and grain physical traits were mapped and characterized in two spring oat populations derived from the crosses 'ND991151/ND030299' (population 05021 – 97 RILs) and 'Souris/ND030299' (population 05026 – 93 RILs). The two populations were evaluated for 4 years at Fargo, ND and 2 years at Casselton, ND. Composite Interval Mapping (CIM) from Windows QTL Cartographer was used for OTL analysis with two framework maps consisting of 640 molecular markers for population 05021, and 398 molecular markers for population 05026, respectively. Nineteen genomic regions on 14 linkage groups were significantly associated with agronomic and grain chemical traits in the population 05021. Fourteen genomic regions on 12 linkage groups were identified for agronomic traits in the population 05026. The same genomic region on LG 05021-16, from 19.4 to 45.8 cM, was associated with thin kernels, test weight, 1000 kernel weight, and oil content. Two QTLs affecting grain yield and test weight had been mapped at the exact same position on LG 05021-34. LG 05026-19 loci, from position 23.7 to 47 cM, and had strong effects on heading date, plant height, and grain yield. These correlated responses could be due to linkage of the underlying QTLs or to pleiotropy. The QTLs consistently detected across environments and between the two populations could serve as starting points for marker-assisted selection.

## 4.2. Introduction

Plant breeding consists essentially of the selection of specific plants with desirable traits with the objective to assemble more desirable combinations of genes in new varieties (Collard and Mackill, 2008). Grain yield, test weight, 1000 kernel weight, heading date, and plant height are major agronomic traits in cultivated oat (Avena sativa L.). On the other hand,  $\beta$ -glucan content, oil content, groat percentage and dehulling efficiency are major quality and grain physical traits. Identification of genes determining important functional agronomic traits had been done in oats over the last 15 years (Okon and Kowalczyk, 2012). One of the most important mapping populations of cultivated oats was generated from the cross Kanota/Ogle (KO) containing 561 genetic markers making up 38 linkage groups (O'Donoughue et al., 1995). Restriction Fragment Length Polymorphism (RFLP) markers from the KO mapping population has been useful in identifying QTLs for grain yield, test weight, groat percentage, days to heading, and plant height (Siripoonwiwat et al., 1996). RFLP markers were also used to look for genomic regions linked to dwarf genes in the OT207/Kanota mapping population (Milach et al., 1997). Major QTLs controlling crown rust resistance and resistance to barley yellow dwarf virus had been reported in the KO population by Bush and Wise, 1996, and Barbosa-Neto et al., 2000, respectively. Vernalization and photoperiod response to heading date and plant height had been detected in the KO population (Holland et al., 1997) and in the Ogle/TAMO-301 (OT) population (Holland et al., 2002). Three Sequence-Characterized Amplified Region (SCAR) markers and one Cleaved Amplified Polymorphic Sequence (CAPS) marker had been identified to be linked with β-glucan content and oil content (Orr and Molnar, 2008). Major QTLs controlling resistance to Puccinia

*coronata* in the field had been detected in the OT mapping population mostly based on Amplified Fragment Length Polymorphism (AFLP) and Simple Sequence Repeat (SSR) markers (Jackson et al., 2010). Diversity Array Technology (DArT) markers had been used to target genomic regions associated with oil content (Hizbai et al., 2012).

The discovery of single nucleotide polymorphism (SNP) markers was an important step in genetic studies. SNP markers are the most common type of DNA-based markers, represent the smallest unit of genetic variation, and can provide a rich source of useful molecular markers (Cho et al., 1999). For example, 90% of the human genome contains SNPs (Kwok and Gu, 1999). Tenaillon et al., 2001 reported 1 SNP every 104 base pair (bp) in the maize genome. Among 15 soybean genotypes, Van et al., 2005 found out that SNPs occurred at a frequency of 1 per 2038 bp in 16302 bp of coding sequence, and 1 per 191 bp in 16960 bp of noncoding regions. As a result, SNPs are useful in the construction of high-density genetic maps and QTL detection because they can be analyzed using high-throughput systems (Van et al., 2005).

The purpose of this chapter was to identify, using SNP and DArT markers, genomic regions associated with genes underlying the expression of several agronomically important traits in two recombinant inbred oat mapping populations.

#### 4.3. Materials and methods

Genotypes from population 05021 and population 05026, described in chapter 2, were grown at two ND locations: Fargo and Casselton. Field experiments were conducted for 4 years at Fargo and during 2 years at Casselton for a total of 6 environments (years and locations combined). A square lattice design was used at Fargo and Casselton during 2008 and 2009 planting seasons as explained in chapter 2, whereas an augmented design was used at Fargo during 2011 and 2012 planting seasons (Table 4.1). The following agronomic traits: grain yield, test weight, heading date, plant height, and thin kernels were evaluated in all the environments. 1000 kernel weight, oil content,  $\beta$ -glucan content, groat percentage, lodging, and dehulling efficiency were assessed only during the 2008 planting season. Genotypic means were obtained using the MIXED procedure from SAS (SAS Institute, Cary, NC) and JMP genomics 6.1 software (SAS Institute, Cary, NC). For the purpose of QTL mapping, and because significant genotype-by-environment interaction were observed for all traits (See chapter 2), combined genotypic means by year and by location were also obtained for a total of 6 combinations (Table

4.2).

Table 4.1. Description of environments used to evaluate two recombinant inbred oat populations for grain quality.

	0 1	J .			
Environ	Location	Year	code	Design	Traits evaluated
ment					
1	Fargo	2008	Far08	Lattice	Yld, kwt, twt, head, height, brk
2	Fargo	2009	Far09	Lattice	Yld, twt, head, height, brk
3	Casselton	2008	Cass08	Lattice	Yld, kwt, twt, head, height, brk,
4	Casselton	2009	Cass09	Lattice	Yld, twt, head, height, brk,
5	Fargo	2011	Far11	Augmented	Yld, kwt, twt, head, height, brk, lodge
6	Fargo	2012	Far12	Augmented	Yld, kwt, twt, head, height, brk,

Table 4.2. Description of combined environments used to evaluate two recombinant inbred oat populations for grain quality.

Enviro	Name	Description	Code	Traits evaluated
nment				
1	Combined 2008	Fargo 2008 + Casselton 2008	Comb08	Yld, kwt, twt, head, height, brk,
2	Combined 2009	Fargo 2009 + Casselton 2009	Comb09	Yld, twt, head, height, brk,
3	Combined Fargo	Fargo 2008 + Fargo 2009	Combfar	Yld, twt, head, height, brk
4	Combined Casselton	Casselton 2008 + Casselton 2009	Combcass	Yld, twt, head, height, brk,
5	Combined 08/09	Fargo 2008 + Casselton 2008 + Fargo 2009 + Casselton 2009	Comb0809	Yld, twt, head, height, brk,
6	Combined 11/12	Fargo 2011 + Fargo 2012	Comb1112	Yld, twt, head, height, brk,

Yld=grain yield, kwt=1000 kernel weight, twt=test weight, head=heading date, height=plant height, brk=percentage of broken kernels

Agronomic and chemical traits were quantitatively mapped by Composite Interval Mapping (CIM) using Windows QTL Cartographer version 2.5 (Wang et al., 2007). CIM method is a combination of interval mapping and multiple regression. Interval mapping fit a linear model at every position in the genome whereas multiple regression fit covariates to control linked and unlinked QTL effects and reduce the model residual (Silva et al., 2012). The general CIM statistical model is as follow:

$$y_i = \mu + Z_i B + \sum_{r=1}^m X_{ir} \beta_r + e_i$$

Where,

 $y_i$  = phenotypic trait value of genotype i

 $\mu$  = overall mean

 $Z_i$  = predictor variables corresponding to the effects of a putative QTL

B = Effects of a putative QTL which depends on the mating design

 $X_{ir}$  = Predictor variables corresponding to the rth cofactor marker

 $\beta_r$  = Coefficient associated with rth cofactor marker

 $e_i = random error.$ 

A forward stepwise regression and backward elimination with the standard CIM model with the following parameters, a probability of 0.1 to enter and leave the model, a window size of 5 cM, and a walk speed of 1 cM, was used to search for the main QTLs. A LOD threshold setting for significant QTLs was determined based on 1000 permutation tests at a significance level of 0.05. QTLs detected using this procedure were considered valid if they were observed at the same position in the genome in at least 50% of the environments and combined environments evaluated, when the signs of the additive effects were consistent across environments (Portyanko et al., 2005). Quantitative trait loci (QTLs) with overlapping support intervals for the same trait

were considered as a single QTL. If the overlapping support intervals represented different traits, the QTLs were assumed to be linked or pleiotropic.

## 4.4. Results

## 4.4.1. Grain yield

A total of 5 QTLs, associated with grain yield, were identified in the population 05021on 3 linkage groups (LG). Linkage group 05021-7.1 corresponding to oat chromosome 7C-17A had 3 QTLs whereas one QTL was observed on each of LG 05021-8 (oat chromosome 8A) and 05021-34. The QTL with the largest effect, which accounts for an average of 21% of the phenotypic variation in grain yield, was located on LG 05021-34 in a region flanked by the DArT markers oPt-15309 and oPt-15736 (Figure 4.1 - 05021-34). This QTL has been consistently observed on 7 of the 12 environments evaluated and the positive allele was contributed by the lower yielding parent 'ND991151' with and additive effect of 247 kg/ha (Table 4.3-I). The second major QTL, accounting for an average of 15% of the phenotypic variation, was mapped in LG 05021-8 in a region containing 3 EST markers (ES15 c11400 361, ES13 c3498 934, and ES03 c695 117), the GBS marker GBS 111982, and the DArT marker oPt-5729 (Figure 4.1 – 05021-8). This QTL was observed in 5 of the 12 environments evaluated and the positive allele was contributed by the higher yielding parent 'ND030299' with an additive effect of 300 kg/ha (Table 4.3-I). Three smaller QTLs, explaining only 11% of the phenotypic variation, were detected in 10 environments at 3 positions along LG 05021-7.1 (Table 4.3-I and Figure 4.1 – 05021-7.1).

On the basis of composite interval mapping (CIM), 3 minor QTL significantly associated with grain yield were identified in the population 05026. All alleles for grain yield were derived from 'Souris', the higher yielding parent. The QTL on LG 05026-5.1, homologous to oat

chromosome 5C, was located in a region containing 9 EST markers and 1 GBS marker tightly linked (Figure 4.2 – 05026-5.1), explained 12% of the total phenotypic variation, and was consistently detected in 8 of 12 environments. The second QTL, observed in 6 environments and accounting for 10% of the phenotypic variation, was mapped on LG 05026-16.1, homologous to oat chromosome 16A, in a region of 10 cM involving 8 EST markers, 2 GBS markers, and 1 DArT marker (Figure 4.2 – 05026-16.1). Located in a region of 10.5 cM, involving 6 EST markers and 2 GBS markers (Figure 4.2 – 05026-19), the third QTL was identified on LG 05026-19 and explained only 11% of the phenotypic variation (Table 4.5-I).

#### 4.4.2. 1000 kernel weight

A total of 3 QTLs, associated with 1000 kernel weight and explaining together 49% of the total phenotypic variation among the progenies, were identified on 3 LGs of population 05021 in all the 3 environments evaluated. All the positive alleles were contributed by 'ND991151', the parent with higher 1000 kernel weight. The QTL with the largest effect, which explains 34% of the phenotypic variation with an additive effect of 2.3 g, was located on LG 05021-16 homologous to oat chromosome 16A at position 62.8 cM and tightly linked to the EST marker ES02_c12776_148 (Figure 4.1 – 05021-16). The two remaining QTLs, with minor effects accounting for 8% and 7% with an additive effect of 1 g, were identified on LG 05021-14.1 (oat chromosome 14D) and LG 05021-18 (oat chromosome 18D), respectively (Table 4.3-II and Figure 4.1 – 05021-14.1 and 05021-18).

A total of 3 minor QTL for 1000 kernel weight, explaining 27% of the total phenotypic variation, were discovered in the population 05026 on linkage groups LG 05026-1.1, LG 05026-5.2, and were observed in all the environments evaluated. Both parents

contributed alleles for higher 1000 kernel weight. 'Souris' provided two alleles and 'ND030299' provided one allele (Table 4.5-II and Figure 4.2 – 05026-1.1, 05026-5.1, 05026-5.2).

## 4.4.3. Test weight

A total of 3 QTLs, associated with test weight, were identified in the population 05021 on LG 05021-14.1 (oat chromosome 14D), LG 05021-16 (oat chromosome 16A), and LG 05021-34. These QTLs, all with minor effects, explained together 34% of the total phenotypic variation among the progenies. Positive alleles on LG 05021-16 and LG 05021-34 were contributed by the lower test weight parent 'ND030299' with an additive effect of 9 kg/m³ and 6 kg/m³, respectively. A positive allele on LG 05021-14.1 was contributed by 'ND991151', the parent with higher test weight with an average additive effect of 12 kg/m³ (Table 4.3-III). The region on LG 05021-14.1 explained 13% of the phenotypic variation and involved 4 EST markers, 1 DArT marker, and 1 GBS marker (Figure 4.1 – 05021-14.1). A region on LG 05021-16, also associated with 1000 kernel weight, explained 9% of the phenotypic test weight variation and comprised a group of 7 EST markers and 2 DArT markers (Figure 4.1 – 05021-34 and explained 12% of the phenotypic variation (Figure 4.1 – 05021-34). This genomic region is also associated with grain yield.

A total of 3 QTLs were found to affect test weight in the population 05026 on linkage groups LG 05026-1.1, LG 05026-4, and LG 05026-5.1. All the positive alleles were contributed by 'Souris' the parent with higher test weight. Together, the 3 QTL explained 39% of the phenotypic variation. The QTL with the largest effect, accounting for 16% of the phenotypic variation and detected in 7 environments, was located on LG 05026-5.1, homologous to oat chromosome 5C, in a region flanked by the EST markers  $ES17_c18602_497$ , and ES05 c8792 292 (Figure 4.2 – 05026-5.1). The QTL on LG 05026-4 had the second highest

effect (explaining alone 12%) and had been identified in a region involving 6 EST markers and 2 DArT markers (Table 4.5-III and Figure 4.2 - 05026-4).

## 4.4.4. Thin kernels

Two QTLs, associated with thin kernels, were identified in the population 05021on genomic regions belonging to LG 05021-16 and LG 05021-31. The two positive alleles were contributed by 'ND030299', the parent with higher percentage of thin kernels. The QTL with the largest effects, which accounted for 14% of the phenotypic variation, was mapped on LG 05021-16 homologous to oat chromosome 16A at position 19.4 to 22.9 cM and was flanked by two EST markers: ES02_c15898_126 and ES02_c12776_148 (Figure 4.1 – 05021-16). This QTL was consistently detected in all the environments evaluated. The other QTL with minor effects explained only 9% of the phenotypic variation and was identified in 5 environments on LG 05021-31 on a region including the DArT marker oPt-8936 (Table 4.3-IV and Figure 4.1 – 05021-31).

A total of 3 QTLs significantly associated with thin kernels were detected in the population 05026 on linkage groups LG 05026-1.1, LG 05026-4, and LG 05026-8.1. Both parents contributed alleles for greater percentage of thin kernels with most contributed by the high thin kernels parent 'ND030299'. Together, the 3 QTL explained 43% of the phenotypic variation associated with thin kernels. The QTL with the largest effect, explaining 23% of the phenotypic variation, was located on LG 05026-8.1, Homologous to oat chromosome 8A, in a genomic region involving 3 EST markers and 3 DArT markers (Figure 4.2 – 05026-8.1). The two other QTL with minor effects were identified on LG 05026-1.1 (oat chromosome 1C), and LG 05026-4 (oat chromosome 4C) (Table 4.5-IV).

#### 4.4.5. Plant height

Two QTLs, found to affect plant height, were identified in the population 05021 on LG 05021-19 and LG 05021-6, and together explained 31% of the total phenotypic variation. The QTL with the largest effect (17% of the phenotypic variation) was located on LG 05021-19 homologous to oat chromosome 19A in a distorted region involving the EST marker ES01_c1793_450 and a group of 4 DArT markers: oPt-15971, oPt-3224, oPt-16979, and oPt-11532 (Figure 4.1 – 05021-19). This QTL was detected on all the environments evaluated and the positive allele was contributed by the taller parent 'ND991151' with an increase on plant height of 3.6 cm. The second QTL which accounted for 14% of the phenotypic variation was identified on LG 05021-6 (oat chromosome 6C) within also a distorted region flanking by the EST marker ES05_c1798_423 and the DArT marker oPt-11599 (Figure 4.1 – 05021-6). The positive allele of this QTL, observed only in 3 environments, was contributed by the shorter parent 'ND030299' with a reduction on plant height of 3.5 cm (Table 4.3-V).

Two QTL, associated with plant height and explaining 32% of the total phenotypic variation, were identified in the population 05026. Souris-derived alleles detected on LG 05026-19, in a region involving 4 EST markers and 2 GBS markers (Figure 4.2 – 05026-19), were associated with shorter plants, reduced plant height by 2.6 cm, and explained 21% of the phenotypic variation. On the other hand, the ND030299 alleles on LG 05026-6 explained 11% of the phenotypic variation and were associated with taller plants (Table 4.5-V).

### 4.4.6. Heading date

Three major QTLs, accounting for 54% of the total phenotypic variation observed for heading date, were discovered in the population 05021 on 3 LGs. The first major QTL and the most important one, detected in 11 of 12 environments, was located on LG 05021-1.2 (oat

chromosome 1C) at a peak position of 31.4 to 46.7 cM involving 3 EST markers, 4 DArT markers, and 1 GBS marker (Figure 4.1 – 05021-1.2). The positive allele for this QTL, explaining 19% of the phenotypic variation, was contributed by the earlier parent 'ND991151' and reduced heading date by 1.1 day. The second major QTL was mapped on LG 05021-14.1 (oat chromosome 14D) in a region flanking by two EST markers: ES15_c12071_400 and ES01_c13432_100 (Figure 4.1 - 05021-14.1) and explained 16% of the phenotypic variation. The positive allele for this QTL, detected in 8 of 12 environments, was contributed by the later parent 'ND030299' and increased heading date by 1 day. The third major QTL, which accounted for 19% of the phenotypic variation, was identified on LG 05021-20 (oat chromosome 20D) in a region flanking by the EST marker ES01_c9095_194 and the DArT marker DS_oPt-2653_332 (Figure 4.1 – 05021-20). The positive allele for this QTL that increased heading date by 1.2 days was contributed by 'ND030299' (Table 4.3-VI).

Five QTL for heading date in five different linkage groups were identified in the population 05026. Together, the main effect of these QTL accounted for 55% of the phenotypic variation observed for heading date. 'ND030299', the later parent, contributed alleles that increased heading date at four QTL on linkage groups LG 05026-20.1, LG 05026-19, LG 05026-6, and LG 05026-7.1 which respectively accounted for 18, 8, 10, and 9% of the phenotypic variation. 'Souris', the earlier parent, contributed alleles for decreased heading date at one QTL on linkage group LG 05026-3 and accounted for 10% of the phenotypic variation. The region on LG 05026-20.1 flanked by the EST markers ES01_c22619_143 and ES01_c9095_194 (Figure 4.2 – 05026-20.1), showed the strongest association with heading date and was consistently detected in all the environments evaluated. The 'ND030299' allele at this locus was associated with an increase in 1.2 days to heading. The second most important locus associated with

heading date involved a group of 6 EST markers, 3 DArT markers, and 1 GBS marker on LG 05026-3 (Figure 5.2 - 05026-3). At this locus, the 'Souris' allele contributed to a decrease in 0.9 day to heading (Table 4.5-VI).

## 4.4.7. Biochemical and other agronomic traits in population 05021

A total of 12 QTLs in 9 LGs were associated with  $\beta$ -glucan content, oil content, groat percentage, dehulling efficiency, and lodging (Table 4.4). At 1 cM from the EST marker ES17_c24_198 on LG 05021-12 (Figure 4.1 – 05021-12), homologous to oat chromosome 12D, only one QTL was detected for  $\beta$ -glucan content. This locus, derived from the parent with higher  $\beta$ -glucan content 'ND030299', accounted for 22% of the total phenotypic variation with an additive effect of 0.4% (Table 4.4-I).

The variation in oil content was associated with 2 QTLs. The two loci explain 43% of the total phenotypic variation among the progenies. One of them with the largest effects was detected on LG 05021-16 (oat chromosome 16A) on a distorted region flanked by two EST markers (Figure 4.1 - 05021-16) and explain 35% of the phenotypic variation. The positive allele was derived from 'ND030299' with an additive effect of 1.4% (Table 4.4-II). Linked to EST marker ES02_c3604_151 (Figure 4.1 – 05021-1.1), the second QTL with minor effects was identified in a distorted region on LG 05021-1.1 homologous to oat chromosome 1C. This locus explained only 8% of the phenotypic variation and was derived from 'ND991151' with an additive effect of 0.6% (Table 4.4-II).

Two QTLs found on LG 05021-13 and 05021-14.1 explained 28% of the phenotypic variation observed in groat percentage. Both alleles that increased groat percentage were contributed by 'ND991151'. The QTL on LG 05021-13 was linked to GBS marker GBS_16970 (Figure 4.1 - 05021-13) at position 95.4 cM and it is also associated with dehulling efficiency

(Table 4.4-IV) whereas the one on LG 05021-14.1 was located at 1 cM from EST marker ES_CC7903_312 (Table 4.4-III). Lodging was associated with two minor QTLs on LG 05021-1.1 and 05021-18, and together explained 23% of the total phenotypic variation. Both loci were derived from 'ND991151' (Table 4.4-V).

## 4.5. Discussion

## 4.5.1. Comparison to other QTLs in oat

A good comparison of QTL across populations requires maps with homolog or homeolog linkage groups (Dekoeyer et al., 2004). The Kanota/Ogle mapping population (O'Donoughue et al., 1995) was the most used for the last 20 years in the identification of genomic regions associated with agronomic, disease, and grain quality traits in cultivated oats. The other maps published in the last ten years including the Terra/Marion (TM) population (Dekoeyer et al., 2004), the Kanota/Marion (KM) population (Groh et al., 2001), the Ogle/TAM O-301 (OT) population (Portyanko et al., 2001), the MN841801-1/Noble-2 (MN) population (Portyanko et al., 2005), the Ogle/MAM17-5 (OM) population (Zhu and Kaeppler, 2003), the Aslak/Matilda (AM) population (Tanhuanpaa et al., 2012), the OT207/Kanota (OK) population (Milach et al., 1997), and the Dal/Exeter (DE) population (Hizbai et al., 2012) had been aligned, when possible, with the KO mapping population. The alignment between the recent anchored oat consensus map (Oliver et al., 2013) and the expanded KO map (Tinker et al., 2009) make possible accurate comparison of QTLs between population 05021, 05026, and the populations mentioned before using the oatgenes database (http://avena.agr.gc.ca/oatgenes).
LG	OC	Environments	Peak	Flanking	LOD Score	$R^2$	Additive	Positive
		observed	position	position			effect	allele
			сM	cM	Average	Average		
				I. Grain yield				
05021-7.1	7C-17A	10/13	34.1-34.7	24.1-45.3	3.0	0.10	328	ND030299
			79.8-81.5	75.4-84.0	3.3	0.09	245	ND991151
			93.7-97.7	90.2-100.5	3.6	0.13	306	ND991151
05021-8	8A	5/13	66.2-70.9	65.1-72.8	4.2	0.14	332	ND030299
			76.3-84.3	75.3-87.0	4.1	0.15	268	ND030299
05021-34	-	7/13	5.0-6.7	1.8-8.7	7.5	0.25	324	ND991151
		5/13	12.2-13.7	10.2-15.2	4.9	0.17	170	ND991151
			II.	1000 kernel we	ight			
05021-14.1	14D	3/3	62.8	60.6-65.1	4.3	0.08	1.0	ND991151
05021-16	16A	3/3	19.9	19.4-23.1	14.6	0.34	2.3	ND991151
05021-18	18D	3/3	94.0	88.4-95.4	3.8	0.07	1.0	ND991151
				III. Test weigh	t			
05021-14.1	14D	9/12	41.0-54.0	35.7-59.6	4.8	0.13	12.0	ND991151
05021-16	16A	7/12	19.4-36.8	11.8-40.4	3.1	0.09	8.7	ND030299
05021-34	-	9/12	0.0-12.7	0.0-19.0	3.5	0.12	6.0	ND030299
			]	V. Thins kerne	els			
05021-16	16A	12/12	19.4-22.9	14.0-26.9	6.4	0.14	1.9	ND030299
05021-31	-	5/12	15.2-16.1	13.0-17.1	4.9	0.09	1.9	ND030299
				V. Plant heigh	t			
05021-19	19A	12/12	50.6-58.0	43.4-62.1	5.4	0.17	3.6	ND991151
05021-6	6C	3/12	66.8-71.8	64.8-81.0	4.3	0.14	3.5	ND030299
				VI. Heading da	te			
05021-1.2	1C	11/12	31.4-46.7	27.7-61.6	6.5	0.19	1.1	ND991151
05021-14.1	14D	8/12	63.3-64.3	60.6-66.0	5.5	0.16	1.0	ND030299
05021-20	20D	7/12	47.5-57.5	44.4-58.7	5.8	0.19	1.2	ND030299

Table 4.3. Genomic regions significantly associated with agronomic traits identified by composite interval mapping (CIM) in population 05021.

LG	OC	Peak position	Flanking	LOD Score	$R^2$	Additive	Positive allele
			position			effect	
		cM	cM	Average	Average		
			I. β-glucan con	tent			
05021-12	12D	130.7	125.4-135.3	6.7	0.22	0.38	ND030299
			II. Oil conter	nt			
05021-16	16A	44.3	44.0-45.4	10.9	0.35	1.35	ND030299
05021-1.1	1C	17.5	16.0-19.1	3.4	0.08	0.61	ND991151
		]	II. Groat perce	ntage			
05021-13	13A	95.4	94.3-97.7	3.9	0.13	1.39	ND991151
05021-14.1	14D	77.0	70.0-82.1	5.0	0.15	1.53	ND991151
		Γ	V. Dehulling effi	ciency			
05021-13	13A	95.4	93.9-98.6	3.4	0.12	1.37	ND991151
			V. Lodging				
05021-1.1	1C	0.0	0.0-1.2	3.5	0.13	0.45	ND991151
05021-18	18D	80.8	78.2-86.8	3.8	0.10	0.45	ND991151

Table 4.4. Genomic regions significantly associated with biochemical and other agronomic traits identified by composite interval mapping (CIM) in population 05021.

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LG	OC	Environments	Peak position	Flanking	LOD Score	$\mathbb{R}^2$	Additive	Positive	
		observed		position			effect	allele	
			сM	cM	Average	Average			
				I. Grain yield					
05026-5.1	5C	8/13	67.2-76.8	53.5-78.0	3.4	0.12	156	Souris	
05026-16.1	16A	6/13	41.4-51.4	40.1-55.5	2.7	0.10	140	Souris	
05026-19	19A	9/13	37.7-48.2	23.4-48.2	2.9	0.11	171	Souris	
II. 1000 kernel weight									
05026-1.1	1C	3/3	12.3-15.8	8.5-19.8	2.8	0.10	0.7	Souris	
05026-5.1	5C	3/3	79.2-80.3	76.8-85.5	2.6	0.09	0.7	ND030299	
05026-5.2	5C	3/3	34.6	29.3-42.1	2.2	0.08	0.7	Souris	
				III. Test weight	t				
05026-1.1	1C	9/12	21.6-23.4	15.7-27.4	3.4	0.11	6.0	Souris	
			0.6	0.0-5.3	2.9	0.08	4.1	Souris	
05026-4	4C	10/12	7.2-16.1	4.6-19.9	3.5	0.12	5.7	Souris	
05026-5.1	5C	7/12	13.0-18.5	4.9-25.2	4.1	0.16	7.1	Souris	
05026-18	18D	5/12	3.4-5.1	1.6-7.5	4.0	0.13	8.4	ND030299	
			Ι	V. Thins kerne	ls				
05026-1.1	1C	11/12	15.2-16.8	7.2-21.8	3.9	0.11	0.145	ND030299	
05026-4	4C	8/12	4.6-9.6	0.0-13.3	3.2	0.09	0.130	ND030299	
05026-8.1	8A	12/12	5.1-8.2	0.0-10.4	7.8	0.23	0.196	Souris	
				V. Plant height	ţ				
05026-19	19A	12/12	42.7-47.0	33.9-47.6	6.0	0.21	2.6	Souris	
05026-6	6C	11/12	31.9-40.0	21.1-51.8	3.2	0.11	2.0	ND030299	
			۲	VI. Heading dat	te				
05026-3	3C	12/13	112.2-129.1	102.8-139.1	3.3	0.10	0.9	Souris	
05026-20.1	20D	12/13	6.0-7.0	0.0-7.5	6.1	0.18	1.2	ND030299	
05026-19	19A	13/13	40.7-47.6	24.5-48.2	3.0	0.08	0.3	ND030299	
05026-6	6C	6/13	4.8-7.2	0.9-19.9	3.7	0.10	0.9	ND030299	
05026-7.1	7C-17A	10/13	56.9-64.5	51.9-69.5	2.8	0.09	0.8	ND030299	

Table 4.5. Genomic regions significantly associated with agronomic traits identified by composite interval mapping (CIM) in population 05026.

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Figure 4.1. Linkage groups from the framework linkage map developed for the 'ND991151' x 'ND030299' F₆ recombinant inbred oat population (Population 05021) showing significant QTLs – yld=grain yield, kwt=1000 kernel weight, twt=test weight, brk=thin kernels, head=heading date, height=plant height, bg=β-glucan content, groat=groat percentage, dehull=dehulling efficiency, lodge=lodging.





Figure 4.1. Linkage groups from the framework linkage map developed for the 'ND991151' x 'ND030299' F₆ recombinant inbred oat population (Population 05021) showing significant QTLs - yld=grain yield, kwt=1000 kernel weight, twt=test weight, brk=thin kernels, head=heading date, height=plant height, bg=β-glucan content, groat=groat percentage, dehull=dehulling efficiency, lodge=lodging (Continued).

05021-14.1

#### 05021-18



Figure 4.1. Linkage groups from the framework linkage map developed for the 'ND991151' x 'ND030299' F₆ recombinant inbred oat population (Population 05021) showing significant QTLs – yld=grain yield, kwt=1000 kernel weight, twt=test weight, brk=thin kernels, head=heading date, height=plant height, bg=β-glucan content, groat=groat percentage, dehull=dehulling efficiency, lodge=lodging (Continued).



Figure 4.2. Linkage groups from the framework linkage map developed for the 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) showing significant QTLs – yld=grain yield, kwt=1000 kernel weight, twt=test weight, brk=thin kernels, head=heading date, height=plant height.



Figure 4.2. Linkage groups from the framework linkage map developed for the 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) showing significant QTLs – yld=grain yield, kwt=1000 kernel weight, twt=test weight, brk=thin kernels, head=heading date, height=plant height (Continued).

OTLs for grain yield had been found in this study on LG 05021-7.1, 05021-8, 05021-34 for population 05021, and on LG 05026-5.1, 05026-16.1, 05026-19 for population 05026. Similarly, Siripoonwiwat et al., 1996 reported also major QTL associated with grain yield in the Kanota/Ogle mapping population at two positions along LG KO 16 23, homologous to oat chromosome 8A. The QTL located at position 84 cM is approximately from 15 cM to the one reported in this study on LG 05021-8. A QTL associated with grain yield and linked to RFLP marker bcd1261a had been reported by Siripoonwiwat et al., 1996 on LG KO 1 3 38 homologous to oat chromosome 7C-17A and LG 05021-7.1. The position of the QTL identified on LG 05026-5.1 collocates with a QTL linked to RFLP marker cdo1312b in a homologous region in the Terra/Marion mapping population TM 22 (Dekoeyer et al., 2004). The QTL detected on LG 05026-16.1 is in agreement with the previous report of a QTL for grain yield in the putatively homologous genetic region KO 24 26 34 and KO 11 41 20 45 on the Kanota/Ogle population (Siripoonwiwat et al., 1996; Dekoeyer and Stuthman, 2001), TM 5 on the Terra/Marion population (Dekoeyer et al., 2004), LG 12 in the Aslak/Matilda mapping population (Tanhuanpaa et al., 2012). Siripoonwiwat et al., 1996 found two major QTL affecting grain yield in the Kanota/Ogle population KO 22 at position 112 linked to RFLP marker cdo708b, and at position 93 close to RFLP marker cdo484a. Both of them were identified in the present study on LG 05026-19 homologous to KO 22.

A QTL for 1000 kernel weight had been reported by Tanhuanpaa et al., 2012 in the Aslak/Matilda population LG 7b, and Beer et al., 1997 in the Kanota/Ogle mapping population at two positions on LG KO 16 homologous to oat chromosome 14D and LG 05021-14.1. Similarly, Dekoeyer et al., 2004 reported a QTL associated with 1000 kernel weight linked to phenotypic marker n1(responsible for the hulless character in oat) in the Terra/Marion mapping population

LG TM 5. This region showed homology with oat chromosome 16A, which corresponds to the location identified in this study on LG 05021-16. Tanhuanpaa et al., 2012 reported also this QTL on LG AM17 homologous to LG 05021-16.

Two QTLs associated with test weight had been reported by Siripoonwiwat et al., 1996 in the Kanota/Ogle mapping population LG KO 11 41+20. The first one linked to RFLP marker cdo836arv was mapped at 30 cM, and the second one linked to RFLP marker cdo1090c at 61 cM. This region on KO 11 41+20 showed homology with oat chromosome 16A. Another QTL identified on KO 14 homologous to oat chromosome 14D had been reported by Siripoonwiwat et al., 1996 in a region similar to the QTL found on LG 05021-14.1. Dekoeyer et al., 2004 reported a QTL associated with test weight in the Terra/Marion mapping population TM 5 flanking by the RFLP marker aco118b and the phenotypic marker n1. Linkage group TM 5 is syntenic to oat chromosome 16A and the QTL is located in the same region reported on this study on LG 05021-16. The position of the QTL reported for test weight on LG 05026-1.1 suggests that it had been mapped at approximately 10 cM to a QTL linked to RFLP marker isu2287a reported by Siripoonwiwat et al., 1996 on the Kanota/Ogle population LG KO 11 41+20. The map position of the QTL identified on LG 05026-4 suggests that it corresponds to a QTL reported by Siripoonwiwat et al., 1996 in the Kanota/Ogle population LG KO 32, homologous to oat chromosome 4C.

Dekoeyer et al., 2004 reported a major QTL affecting thin kernels and linked to phenotypic marker n1 in the Terra/Marion mapping population TM 5 homologous to oat chromosome 16A and LG 05021-16.

QTLs have been reported for plant height in the Kanota/Ogle mapping population KO 22, homologous to oat chromosome 19A, LG 05021-19, LG 05026-19, at position 108 cM (Holland

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et al., 1997), 141, 98, and 116 cM (Siripoonwiwat et al., 1996; Milach et al., 1997). A QTL in a similar region has been reported by Tanhuanpaa et al., 2012 on LG 17 of the Aslak/Matilda population. Holland et al., 1997 reported also a QTL on KO 7_10_28 and KO 29_43 homologous to oat chromosome 6C, LG 05021-6, LG 05026-6.

Siripoonwiwat et al., 1996 reported 3 QTLs associated with heading date in the Kanota/Ogle mapping population, two of them on KO 11 41+20 and the last one on KO 37. Holland et al., 2002 reported the same QTL on the Ogle/TAM 0-301 mapping population OT 34 homologous to KO 11 41+20. This region is syntenic to oat chromosome 1C from 49.3 to 73.1 cM which in turn is less than 5 cM from the region identified on LG 05021-1.2. The QTL found in LG 05021-1.2 had been previously reported in a similar region by Tanhuanpaa et al., 2012 on LG AM10 and LG AM12. Another two QTLs associated with heading date had been reported on KO 14, homologous to oat chromosome 14D, at position 14 cM (Holland et al., 1997) and 37 cM (Siripoonwiwat et al., 1996) which correspond to the same region identified on LG 05021-14.1. This QTL had been also discovered by Tanhuanpaa et al., 2012 on LG TM7a. Three QTLs had been reported in two similar genomic regions on LG KO22 and LG OT1 at position 108 cM (Holland et al., 1997), 156 cM (Holland et al., 2002), 171 cM (Siripoonwiwat et al., 1996; Holland et al., 1997). Those two regions in the Kanota/Ogle population and the Ogle/TAM O-301 population are homologous to LG 05026-19. The QTL detected on LG 05026-3 is located in the exact same genomic region of the QTL reported on LG KO42 flanked by RFLP markers isu707a and hkt1c (Siripoonwiwat et al., 1996; Holland et al., 1997).

A QTL located at the same region, to the one found on LG 05021-12 for  $\beta$ -glucan content, had been reported by Dekoeyer et al., 2004 in the Terra/Marion mapping population TM 21 homologous to oat chromosome 12D. The position of the QTL identified on LG 05021-1.1

and affecting oil content collocates with a QTL linked to RFLP marker cdo665 in the Kanota/Ogle mapping population KO 11_41+20 (Kianian et al., 1999), RFLP marker rz69 in the Ogle/Marion mapping population OM 3 (Zhu et al., 2004), and DArT markers oPt-17088_A and oPt-6135 in the Dal/Exeter mapping population DE 13 (Hizbai et al., 2012). The same QTL was also reported by Tanhuanpaa et al., 2012 in the Aslak/Matilda mapping population LG 11 and 15 homologous to LG 05021-1.1. Similarly, a QTL for oil content was identified by Dekoeyer et al., 2004 and linked to phenotypic marker n1 in the Terra/Marion mapping population TM 5 homologous to oat chromosome 16A. This QTL was also found by Tanhuanpaa et al., 2012 on LG 12 and 17 homologous to 05021-16. The position of the QTL on LG 05021-14.1 associated with groat percentage corresponds to that of the QTL identified in the Kanota/Marion mapping population KM 14 and linked to RFLP marker cdo1358f (Groh et al., 2001).

## 4.5.2. Linkage and/or pleiotropic effects

It is interesting that the same distorted genomic region on LG 05021-16, from position 19.4 to 45.8 cM including the common EST markers ES02_c15898_126, ES02_c12776_148, and ES14_c1865_384, affects thin kernels, test weight, 1000 kernel weight, and oil content. Also, two QTLs affecting grain yield and test weight has been mapped at the exact same position on LG 05021-34 from 0 to 15 cM, and overlapped by the DArT markers oPt-14477, oPt-15309, oPt-0350, and oPt-15736. Similarly, on LG 05021-14.1 the region affecting test weight is 9 cM apart from the region associated with 1000 kernel weight, heading date, and groat percentage. These associations were consistent with the phenotypic and genotypic correlations observed between test weight and heading date, test weight and groat percentage, heading date and groat percentage, thin kernels and 1000 kernel weight, thin kernels and oil content, 1000 kernel weight and oil content, grain yield and test weight (Chapter 2-Tables 5, 7, 8). On LG 05021-14.1,

ND99151 alleles increased 1000 kernel weight, test weight, groat percentage, and decreased heading date whereas on LG 05021-16, the ND030299 alleles increased oil content and percentage of thin kernels, but decreased test weight and 1000 kernel weight. Tanhuanpaa et al., 2012 found also that groat percentage and heading date mapped to the same genetic location. These correlated responses could be due to linkage of the underlying QTLs or to pleiotropy. We point out both genetic actions because the resolution of the maps used in this study is not strong enough to discriminate QTLs with pleiotropic effects from tightly linked QTLs affecting the different traits.

The QTLs observed for oil content on LG 05021-1.1 and LG 05021-16 were identified in distorted regions. The QTLs for oil content reported by Kianian et al., 1999 on KO 11_41+20, Zhu et al., 2004 on OM 3, and Hizbai et al., 2012 on DE 13 are all located in the same homologous distorted region. The identification of QTLs affecting oil content in four mapping populations in the same distorted region furnishes strong evidence that the mechanisms underlying segregation distortion may directly or indirectly affect oil content.

LG 05026-19 loci, from position 23.7 to 47 cM had strong effects on heading date, plant height, and grain yield. Markers ES03_c1075_468, ES_CC11076_204, GBS_25081, ES17_c7110_570, ES01_c27869_512, and GBS_92025 are shared between the three traits. QTLs associated with grain yield, plant height, and heading date had been also reported by Dekoeyer and Stuthman, 2001. Loci in adjacent regions on LG 05026-6 from position 0 to 52.5 cM also had significant effects on heading date and plant height. Correspondingly, the QTL influencing grain yield on LG 05026-5.1 is approximately 3 cM apart from the region correlated with 1000 kernel weight. The same genomic region on LG 05026-4 overlapped by EST markers ES08_c5133_835, ES01_c22916_27, ES03_c5596_272 affect thin kernels and test weight. GBS

marker GBS_55792 and EST marker ES01_c3447_952 exhibited also strong effects on thin kernels, 1000 kernel weight, and test weight. 'Souris' allele increased test weight on LG 05026-1.1, LG 05026-4, LG 05026-5.1, 'ND030299' allele decreased thin kernels on LG 05026-1.1, LG 05026-4, and increased 1000 kernel weight on LG 05026-5.1. These associations are congruent to the phenotypic and genotypic correlations observed between grain yield and 1000 kernel weight, test weight and 1000 kernel weight, thin kernels and 1000 kernel weight, test weight and 1000 kernel weight, thin kernels and 1000 kernel weight, test weight and 1000 kernel weight, thin kernels and 1000 kernel weight, test weight and thin kernels (Chapter 3 – Table 5). On LG 05026-19, 'Souris' allele increased grain yield, and decreased plant height, and 'ND030299' allele increased heading date whereas on LG 05026-6, 'ND030299' allele increased plant height and heading date. These pleiotropic and/or linked genomic regions may have contributed to the significant positive phenotypic and genotypic correlations observed between heading date and plant height, and between grain yield and plant height in the population 05026 (Chapter 3 – Table 5).

# 4.5.3. Comparing the two populations for common QTLs

QTLs are investigated with the objective to use them in molecular breeding. In order to be exploited in a marker-assisted selection program, the QTL of interest identified should be integrated and validated on other population so that a clearer understanding of the QTL can be gained (Rines et al., 2006). This is a reason why comparative QTLs among population is important.

A QTL with major effect on heading date has been identified on LG 05021-20 of population 05021 from position 47.5 to 57.5 cM. This QTL has also been detected on LG 05026-20.1 of population 05026 from position 6 to 7 cM. Two EST markers (ES15_c13627_458, ES01_c9095_194) were shared between the two homologous genomic regions. The positive allele on both QTLs was contributed by 'ND030299', the common parent between the two

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populations. The effects of both QTLs are quite similar, on the population 05021 the QTL explained 19% of the phenotypic variation whereas on the population 05026 it explained 18% of the variation observed.

A QTL identified for plant height on LG 05021-6 of population 05021 was also observed on LG 05026-6 of population 05026. The shared EST marker (ES05_c1798_423) is responsible for the effects observed. The positive allele was also contributed by the common parent 'ND030299'.

An important genomic region on LG 05026-1.1 of population 05026, from position 12.3 to 23.4 cM, associated with 1000 kernel weight, test weight, and thin kernels had been mapped for heading date in the population 05021 on LG 05021-1.2 from position 31.4 to 46.7 cM. The homologous segment between the two populations comprised 6 EST markers (ES01_c30278_396, ES01_c17631_54, ES05_c1532_208, ES01_c3435_183, ES15_c14779_89, and ES01_c3447_952), 1 DArT marker (DS_LB_1139), and 1 GBS marker (GBS_55792). The positive allele, except for thin kernels, was contributed by the non-common parent, 'ND991151' for population 05021, and 'Souris' for population 05026.

# 4.6. Conclusion

Identification of molecular markers linked to genes controlling important functional traits provides a fast and reliable selection of desirable genotypes. Consequently, the identified markers can play an important role in a plant breeding programme and can seed up considerably the selection of genotypes with better agronomic properties (Okon and Kowalczyk, 2012). In this present research, we identified SNP markers that cosegregate or are closely linked to QTL conditioning many important agronomic traits in oat. Future works, to successfully apply those QTLs in a marker-assisted selection (MAS) program, should focus in validating the QTLs reported on another genetic background and more environments, and developing an efficient and economic marker assay for screening large breeding populations. According to Xu and Couch, 2008 there is still a high discrepancy between QTL studies and application of these studies in a MAS program. One of the principal reasons is because the mapping populations, used on those studies, were from parents highly distinctive in their phenotype, and as noted by Bernardo, 2008 it is possible that these QTL will be population specific and therefore less useful in the context of breeding programs. In our case, since we use parents from a breeding program, the QTL reported in this research, have a higher probability of being valid across elite populations. The results, reported in this study, can also serve as an additional resource in the understanding of genetic mechanisms underlying important agronomic traits in cultivated oats.

# 4.7. References

- Barbosa-Neto J.F., W. Siripoonwiwat, L.S. O'Donoughue, S.M. Gray, D.M. Smith, F.L. Kolb, C. Gourmet, C.M. Brown, and M.E. Sorrells . 2000. Chromosomal regions associated with barley yellow dwarf resistance in oat. Euphytica 114:67–76
- Beer, S.C., W. Siripoonwiwat, L.S. O'Donoughue, E. Souza, D. Matthews, and M.E. Sorrells. 1997. Associations between molecular markers and quantitative traits in oat germplasm pool: can we infer linkages? Journal of Agricultural Genomics 3:1. Available at http://wheat.pw.usda.gov/jag/papers97/paper197/indexp197.html.
- Bernardo, R. 2008. Molecular markers and selection for complex traits in plants: Learning from the last 20 years. Crop Sci. 48:1649–1654. doi:10.2135/cropsci2008.03.0131
- Bush A.L., and R.P. Wise. 1996. Crown rust resistance loci on linkage groups 4 and 13 in cultivated oat. J Hered 87:427–432
- Cho, R.J., M. Mindrinos, D.R. Richards, R.J. Sapolsky, M. Anderson, E. Drenkard, J. Dewdney, T.L. Reuber, M. Stammers, N. Federspiel, A. Theologis, W.H. Yang, E. Hubbell, M. Au, E.Y. Chung, D. Lashkari, B. Lemieux, C. Dean, R.J. Lipshutz, F.M. Ausubel, R.W. Davis, and P.J. Oefner. 1999. Genome-wide mapping with biallelic markers in Arabidopsis thaliana. Nat. Genet. 23:203-207.
- Collard, B.C.Y., and D.J. Mackill. 2008. Marker-assisted selection: an approach for precision plant breeding in the twenty-first century. Phil. Trans. R. Soc. 363:557-572.

- DeKoeyer D.L., and D.D. Stuthman. 2001. Allelic shifts and quantitative trait loci in a recurrent selection population of oat. Crop Sci 41:1228–1234
- Dekoeyer, D.L., N.A. Tinker, C.P. Wight, J. Deyl, V.D. Burrows, L.S. O'Donoughue, A. Lybaert, S.J. Molnar, K.C. Armstrong, G. Fedak, D.M. Wesenberg, B.G. Rossnagel, and A.R. McElroy. 2004. A molecular linkage map with associated QTLs from a hulless x covered spring oat population. Theor Appl Genet 108:1285-1298.
- Groh, S., A. Zacharias, S.F. Kianian, G.A. Penner, J. Chong, H.W. Rines, and R.L. Phillips. 2001. Comparative AFLP mapping in two hexaploid oat populations. Theor Appl Genet 102:876-884.
- Hizbai, B.T., K.M. Gardner, C. P. Wight, R.K. Dhanda, S.J. Molnar, D. Johnson, J. Frégeau-Reid, W. Yan, B.G. Rossnagel, J.B. Holland, and N.A. Tinker. 2012. Quantitative trait loci affecting oil content, oil composition, and other agronomically important traits in oat. Plant Genome 5:164-175.
- Holland, J.B., H.S. Moser, L.S. O'Donoughue, and M. Lee. 1997. QTLs and epistasis associated with vernalization responses in oat. Crop Sci 37:1306-1316.
- Holland, J.B., V.A. Portyanko, D.L. Hoffman, and M. Lee. 2002. Genomic regions controlling vernalization and photoperiod responses in oat. Theor Appl Genet 105:113-126.
- Jackson, E.W., D.E. Obert, J.B. Avant, S.A. Harrison, J. Chong, M.L. Carson, and J.M. Bonman. 2010. Quantitative trait loci in the Ogle/TAM O-301 oat mapping population controlling resistance to *Puccinia coronata* in the field. Phytopathology 100:484-492.
- Kianian, S.F., M.A. Egli, R.L. Phillips, H.W. Rines, D.A. Somers, B.G. Gengenbach, F.H. Webster, S.M. Livingston, S. Groh, L.S. O'Donoughue, M.E. Sorrells, D.M. Wesenberg, D.D. Stuthman, and R.G. Fulcher. 1999. Association of a major groat oil content QTL and an acetyl-CoA carboxylase gen in oat. Theor Appl Genet 98:884-894.
- Kwok, P.Y., and Z. Gu. 1999. Single nucleotide polymorphism libraries: why and how are we building them? Mol Med Today 5:538-543.
- Milach, S.C.K., H.W. Rines, and R.L. Phillips. 1997. Molecular genetic mapping of dwarfing genes in oat. Theor. Appl. Genet. 95:783-790.
- Oatgenes: a comprehensive database of oat markers and QTLs. http://avena.agr.gc.ca/oatgenes.
- O'Donoughue L.S., M.E. Sorrells, S.D. Tanksley, E. Autrique, A. Van Deynze, S.F. Kianian, R.L. Phillips, B. Wu, H.W. Rines, P.J. Rayapati, M. Lee, G.A. Penner, G. Fedak, S.J. Molnar, D. Hoffman, and C.A. Salas. 1995. A molecular linkage map of cultivated oat. Genome 38:368-380.

- Okon, S. and K. Kowalczyk. 2012. Description of DNA analysis techniques and their application in oat (Avena sativa L.) genome research. Acta Agrobotanica 65:3-10.
- Oliver, R.E., N.A. Tinker, G.R. Lazo, S. Chao, E.N. Jellen, M.L. Carson, H.W. Rines, D.E. Obert, J.D. Lutz, I. Shackelford, A.B. Korol, C.P. Wight, K.M. Gardner, J. Hattori, A.D. Beattie, A. Bjornstad, J.M. Bonman, J.L. Jannink, M.E. Sorrells, G.L. Brown-Guedira, J.W.M. Fetch, S.A. Harrison, C.J. Howarth, A. Ibrahim, F.L. Kolb, M.S. McMullen, J.P. Murphy, H.W. Ohm, B.G. Rossnagel, W. Yan, K.J. Miclaus, J. Hiller, P.J. Maughan, R.R.R. Hulse, J.M. Anderson, E. Islamovic, and E.W. Jackson. 2013. SNP discovery and chromosome anchoring provide the first physically-anchored hexaploid oat map and reveal synteny with model species. PLoS ONE 8:e58068. doi:10.1371/journal.pone.0058068.
- Orr, W., and S.J. Molnar. 2008. Development of PCR-based SCAR and CAPS markers linked to β-glucan and protein content QTL regions in oat. Genome 51:421-425.
- Portyanko V.A., D.L. Hoffman, M. Lee, J.B. Holland. 2001. A linkage map of hexaploid oat based on grass anchor DNA clones and its relationship to other oat maps. Genome 44:249-265.
- Portyanko, V.A., G. Chen, H.W. Rines, R.L. Phillips, K.J. Leonard, G.E. Ochocki, and D.D. Stuthman. 2005. Quantitative trait loci for partial resistance to crown rust, *Puccinia coronata*, in cultivated oat, *Avena sativa L*. Theor Appl Genet 111:313-324.
- Silva, L.D.C.E., S. Wang, and Z.B. Zeng. 2012. Composite interval mapping and multiple interval mapping: Procedures and guidelines for using windows QTL cartographer. p 75-119. *In* S.A. Rifkin (ed.). Quantitative trait loci (QTL): Methods and protocols. Methods in Molecular Biology 871. Springer Science + Business Media, NY.
- Siripoonwiwat, W., L.S. O'Donoughue, D. Wesenberg, D.L. Hoffman, J.F. Barbosa-Neto, and M.E. Sorrells. 1996. Chromosomal regions associated with quantitative traits in oats. Journal of Agricultural Genomics 2:3. Available at http://wheat.pw.usda.gov/jag/papers96/paper396/oatqtl3g.html.
- Tanhuanpaa, P., O. Manninen, A. Beattie, P. Eckstein, G. Scoles, B. Rossnagel, and E. Kiviharju. 2012. An updated doubled haploid oat linkage map and QTL mapping of agronomic and grain quality traits from canadian field trials. Genome 55 :289-301.
- Tenaillon, M.I., M.C. Sawkins, A.D. Long, R.L. Gaut, J.F. Doebley, and B.S. Gaut. 2001. Patterns of DNA sequence polymorphism along chromosome 1 of maize (*Zea mays ssp. mays L.*). PNAS 98 :9161-9166.
- Tinker, N.A., A. Kilian, C.P. Wight, K. Heller-Uszynska, P. Wenzl, H.W. Rines, A. Bjornstad, C.J. Howarth, J.L. Jannink, J.M. Anderson, B.G. Rossnagel, D.D. Stuthman, M.E. Sorrells, E.W. Jackson, S. Tuvesson, F.L. Kolb, O. Olsson, L.C. Federizzi, M.L. Carson, H.H. Ohm, S.J. Molnar, G.J. Scoles, P.E. Eckstein, J.M. Bonman, A. Ceplitis, and T. Langdon. 2009. New DArT markers for oat provide enhanced map coverage and

global germplasm characterization. BMC Genomics 10:39. doi:10.1186/1147-2164-10-39.

- Van, K., E.Y. Hwang, M.Y. Kim, H.J. Park, S.H. Lee, and P.B. Cregan. 2005. Discovery of SNPs in soybean genotypes frequently used as the parents of mapping populations in the United States and Korea. Journal of Heredity 96:529-535.
- Wang, S., C.J. Basten, and Z.B. Zeng. 2007. Windows QTL cartographer 2.5_011. Department of statistics, North Carolina State University, Raleigh.
- Xu, Y., and J.H. Crouch. 2008. Marker-assisted selection in plant breeding: From publications to practice. Crop Sci. 48:391–407.
- Zhu, S., B.G. Rossnagel, and H.F. Kaeppler. 2004. Genetic analysis of quantitative trait loci for groat protein and oil content in oat. Crop Sci 44:254-260.
- Zhu, S., and H.F. Kaeppler. 2003. A genetic linkage map for hexaploid, cultivated oat (Avena sativa L.) based on intraspecific cross 'Ogle/MAM17-5'. Theor. Appl. Genet. 107:26-35.

# **CHAPTER 5**

## 5.1. General conclusions

Despite more than 20 years of genomic research, cultivated oats has been lagged behind and has not kept pace with other small grains such as wheat and barley in term of genome sequence, QTL analysis and marker-assisted selection. This dissertation provides supplemental steps towards selection of new oat cultivars with desirable combination of genes. Two recombinant inbred oat populations were evaluated in this research for genotypic and phenotypic relationships among important agronomic traits including grain yield, test weight, 1000 kernel weight, thin kernels, plant height, heading date,  $\beta$ -glucan content, and oil content. Two genetic linkage maps were developed from the two populations and QTLs associated with the agronomic traits were assessed and identified. This dissertation comprises 3 main parts beside the general introduction and that conclusion.

The first main part addressed the issues of the efficiency of indirect selection and the magnitude of response to selection by assessing genotypic and phenotypic correlations among traits. Some important conclusions drawn from that part include:

- Genotype main effect was highly significant for all the agronomic traits under study and showed considerable amount of variation. Such wide variation is an indication of the capacity to improve the two populations evaluated with respect to grain quality.
- The absence of significant genotype-by-environment interaction for 1000 kernel weight suggested that it should be practical to select for 1000 kernel weight.
- The absence of strong correlations among traits indicated that improvement of agronomic traits should not be difficult.

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- Grain yield was positively correlated with test weight, plant height, β-glucan content, and dehulling efficiency suggesting that when quality traits are improved higher yields can be achieved.
- The high heritabilities observed suggested that all the traits measured should respond well to selection on a family-mean basis.

The second main part explored the issues relative to cytogenetic abnormalities in the oat genome and the difficulty of mapping by the construction of two genetic maps and the comparison of those maps with the recently published oat consensus map. The most important conclusions include:

- Thirty linkage groups using 1168 polymorphic markers were formed for population 05021, whereas population 05026 comprised 33 linkage groups using 1024 polymorphic markers.
- The 30 linkage groups of population 05021 varied in size from 15.8 to 225.3 cM, and contained from 3 to 62 markers for a total map size of 2601.7 cM.
- The 33 linkage groups of population 05026 comprised from 2 to 42 markers, varied in size from 2.3 to 143.2 cM for a total map size of 1174.2 cM.
- Twenty six of the 30 linkage groups from population 05021 can be placed on 19 of the 21 oat chromosomes.
- Thirty one of the 33 linkage groups from population 05026 showed homology with 20 of the 21 oat chromosomes.
- The differences noted in the ordering of markers between the two populations and the oat consensus map, are an indication that genomic rearrangements and intervarietal chromosome interchanges exist in the genome of cultivated oat.

• The shorter map observed in population 05026 may be the result of inadequate polymorphism in some regions with similar ancestry.

The purpose of the third main part was to address the QTL mapping in two oat populations, to compare the identified QTLs with those previously published, and to assess linked and/or pleiotropic regions between several agronomically important traits. Key findings discovered in that chapter include:

- Nineteen genomic regions on 14 linkage groups were significantly associated with agronomic and grain chemical traits in population 05021.
- Fourteen genomic regions on 12 linkage groups were identified for agronomic traits in the population 05026.
- The same genomic region on LG 05021-16, from position 19.4 to 45.8 cM, affected thin kernels, test weight, 1000 kernel weight, and oil content.
- Two QTLs affecting grain yield and test weight has been mapped at the exact same position on LG 05021-34.
- The region affecting test weight on LG 05021-14.1 was located 9 cM apart from a region associated with 1000 kernel weight, heading date, and groat percentage.
- Markers on LG 05026-19, from position 23.7 to 47 cM, had strong effects on heading date, plant height, and grain yield.
- The QTL influencing grain yield on LG 05026-5.1 was mapped at approximately
  3 cM from a region correlated with 1000 kernel weight.
- All these correlated responses could be due to linkage of the underlying QTLs or to pleiotropy.

• The identification of QTLs affecting oil content in 4 mapping populations in the same homologous distorted region furnished strong evidence that the mechanisms underlying segregation distortion may directly or indirectly affect oil content.

In conclusion, many linkage maps, over the last 20 years, had been developed for several oat populations. Quantitative trait loci have been identified to be associated with several agronomic traits in cultivated oats. Marker-assisted selection (MAS) has been applied for some economic traits and mostly for disease resistance. Three AFLP markers associated with BYDV had been cloned for MAS purposes (Jin et al., 1998; Jin et al., 1999). Six SCAR and CAPS markers, also linked to BYDV, had been used for MAS (Pal et al., 2002). SNP markers were developed for Pc68 (Chen et al., 2004) and Dw6 dwarfing genes (Kiviharju et al., 2004). Nevertheless, nothing big had been done yet for other agronomic traits. In this point of view, this research will help the oat community with better understanding of the genomic regions underlying some important agronomic traits. In a near future, while waiting for the complete sequencing of the oat genome, the next goal will include the following:

- The utility of the SNP markers, identified and linked to major QTL in oat, needs to be validated in other genetic backgrounds, different locations than the ones used in this study, and years.
- The validated QTLs could be incorporated into breeding cultivars through the use of marker-assisted selection and could also be used in finding candidate genes that explain the genetics of the traits evaluated.

# 5.2. References

- Chen G., J. Chong, M. Gray, S. Prashar, and J.D. Procunier. 2004. Single nucleotide polymorphisms as next generation markers for high throughput screening for crown rust resistance in oat. In: Peltonen-Saino P., Topi-Hulmi M. (eds) Proc 7th Int Oat Conf, Helsinki, Finland, p 86. www.mtt.fi/met/pdf/met51.pdf
- Jin H., L.L. Domier, F.L. Kolb, and C.M. Brown. 1998. Identification of quantitative loci for tolerance to barley yellow dwarf virus in oat. Phytopathology 88:410–415.
- Jin H., L.L. Domier, F.L. Kolb, and C.M. Brown. 1999. Conversion of AFLP markers associated with BYDV tolerance in oats to non-radioactive PCR markers. In: Plant and Animal Genome VII Conf, San Diego, p 396. www.intl-pag.org.
- Kiviharju E., O. Manninen, L. Pietila, and P. Tanhuanpaa. 2004. DNA marker for oat dwarfing gene. In: Peltonen-Saino P., Topi-Hulmi M. (eds) Proc 7th Int Oat Conf, Helsinki, Finland, p 171. www.mtt.fi/met/pdf/met51.pdf
- Pal N., J.S. Sandhu, L.L. Domier, and F.L. Kolb.2002. Development and characterization of microsatellite and RFLP-derived PCR markers in oat. Crop Sci 42:912–918

APPENDIX

## 05021-1.1

0.0 -	∠ GMI_GBS_77337
1.6-\\ <del>`</del>	IGMI DS LB 10716 GMI ES21 c2586 850
3.8 - H	GMI GBS 79453
4.3	GMI_ES02_c6368_605
15.4 1	GMI_DS_CC7541_122
	GMI_ES01_c13467_233 GMI_ES14_c11870_550
17.0	GMI ES02 c28832 308 GMI ES02 lrc17781 234
$\mathcal{N}$	GMI_GBS_5003 GMI_GBS_18040
175 1	GMI_ES02_c3604_154 GMI_GBS_91128
17.5-	GMI ES14 c19327 378 GMI ES22 c928 79
19.1	GMI_ES_CC11177_463
23.6	GMI_DS_CC9481_218 GMI_ES_CC13854_225
and H	GMI_ES01_c22911_435 GMI_ES15_c2934_340
24.1-/	GMI ES05 c14820 212 GMI GBS 6338
as a //	GMI DS LB 72 GMI GBS 97720
29.2-	GMI_GBS_85867 GMI_ES15_c16679_330
27.4	GMI ES22 c6314 447
	GMI_DS_LB_9564 GMI_ES_CC11019_290
29.0	GMI ES02 c8118 320
29.5	GMI_ES05_c22428_236
200	GMI_ES_CC12765_141 GMI_ES15_c1370_537
30.0	GMI_ES15_c5652_491
31.1	GMI_ES05_c14023_258
() ( ( ( ( ( ( ( ( ( ( ( ( ( ( ( ( ( (	GMI_ES03_c16861_126 GMI_GBS_29011
31.6	GMI_ES05_c5968_912 GMI_ES15_c6229_566
<b>∥</b>	GMI_ES15_c911_221
36.5	oPt-15066
43.8	• oPt-11061
Ц	GMI_ES15_c6153_392 GMI_ES14_c5428_351
1	GMI_ES_LB_7961 GMI_ES01_c12570_390
64.1 /	GMI_ES05_c16637_406 GMI_ES05_c20813_204
l (J)	GMI_ES15_c276_702 GMI_ES15_c5908_278
11	IGMI_DS_LB_1713
64.6	<pre>GMI_ES02_c14687_642</pre>
70.0	oPt-17156
71.2	• oPt-8654

### 05021-2

<mark>ר 0.0</mark>		GMI_ES01_c25052_201
15.8 -		oPt-6104
22.2	_	GMI_ES14_c19842_74 GMI_ES17_c3418_95
		IGMI_ES05_c15526_511 GMI_ES14_c7020_89
25.5 -	Π,	GMI_ES05_c8599_963 GMI_DS_LB_4000
20.0	1 18	GMI_ES03_Irc9679_178
27.7	U 🛚	GMI_ES14_c6271_602
10		GMI_ES14_c6885_194 GMI_ES15_c6381_389
N N	J	GMI_ES14_c8931_606 GMI_ES05_c101_122
28.2 ~	$\square$	GMI_DS_CC8468_91 GMI_ES05_c18574_288
	Ħ.	GMI ES05 c5594 313 GMI ES15 c7440 275
Å		GMI ES CC11290 204
29.3	1 🖪	GMI ES15 Irc8841 69 GMI DS CC4033 368
29.9 -	141	GMI_ES05_c16116_187
31.2	1 1	GMI GBS 73687 GMI GBS 64994
31.8 -		GMI_GBS_108223
35.2 -	1_M	GMI ES05 c8417 343 GMI ES15 c13909 303
43.4		oPt-10449
44.9		oPt-11075
53 1		oPt-14513
		GMI ES03 c14095 37 GMI ES03 c22566 720
59 4 J	ורו	GMI_ES03_c3966_68_GMI_ES15_c7005_128
00.4		GMI_ES02_c1433_212
	1 🚺	GMI ES03 c13301 381 GMI ES01 c3302 178
60.0 -	i 🛛	GMI_ES17_c4546_112 GMI_ES01_c24681_389
	I-II	GMI_ES14_c4827_489 GMI_GBS_7561
611		GMI ES CC10136 320
62.2		GMI_ES03 c10621_322
62.7 -		GMI_ES14_c7737_88
	. 0	GMI_ES01_c1635_353 GMI_ES_CC8700_285
63.2	1	GMI ES01 c8470 599 GMI ES CC4978 509
777-	i I <b>V</b>	GMI_ES03_c10410_600
78.8	1 1	GML GBS 70341
		GML ES22 c1298 437 GML ES14 c2320 26
86.0 -	Ы	GMI DS I B 3936 GMI ES17 (899 571
000	1 1	GMI GBS 81139
079-	' U	GML ES05 Irc18863 260
122-	- 1	GML ES15 c7706 583
151	- 1	GML GBS 31405
416		GML DS LB 4073

## 05021-1.2

0.0 -		🖍 oPt-3170
1.1-	-	- oPt-15994
2.3		🛰 oPt-3783
13.7 \		r GML ES14 c3401 306
		IGMI_ES01_09472_428 GMI_GBS_114315
17.0-1		GMLES IB 11704 GMLES22 (10159 379
· · · • 11		GML ES03 (881 241
17.5 -		CML_ES02_C4536_439
17.7		GML DS LB 10008
17.0		CMI_D3_ED_10090
17.97	Η	GWI_ES05_01912_404
18.1-7		GML_E302_IC13446_326 GML_GB3_63637
19.2		• GWI_ES17_09767_102
25.4 ~	_	GMI_ES02_012621_204
26.5-1	Ξ	GMI_ES18_c4764_27 GMI_GBS_85336
/		IGMI_ES15_c16835_340
27.1 /	_	GMI_ES02_Irc13788_346
//		\GMI_GBS_55792 GMI_ES15_c14779_89
27.7	П	GMI_ES05_c1532_208 GMI_ES01_c3435_183
8		GMI_ES01_c3447_952
31.4		GMI_GBS_79511
32.0	=	GMI ES22 c15387 395
33.8-1	_	IGMI DS LB 1139 GMI ES01 c17631 54
34.3	_	GMI ES01 c30278 396
41.0		0Pt-5811
418		0Pt-1032
43.6		0Pt-2998
45.7		oPt-12680
40.1		0.112000
		IGML ES05 c30 044 GML ES05 c16400 86
63.4	θ	CML ES02 c21041 620
		IGINI_E302_021041_039

### 05021-3

00-		GML ES15 c1433 87
40-		GMI ES03 c6194 428
51-	$\square$	GML ES03_03525_308 GML ES03_013250_560
56-1	R	• GML ES05_c1774_174
/	$   \rangle$	GML DS LB 7523 GML DS LB 8152
7.2 -	Ш`	GML ES01 c11827 414
212/		GML DS_CC1149_344
245		GMI GBS 113531
33.8 -	┢╋	GMI_ES02_c19258_432_GMI_ES05_c19273_362
41.4 ~~		oPt-6377
58.7 -	Π.	oPt-2907
60.0		oPt-3838
1 9 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3		GML GBS 13773 GML DS oPt-18257 376
67.1 -	<b>=</b> //	GML ES01 (8255 502
68.2~		GML ES02 c10035 217
68.7 -1	₹.	- GML ES14_08386_549
69.8	Ц)	GMI_ES15_c5060_161
766-1	$\Box V$	GMI_ES01_09955_227
77 1 -//	=(/)	GML ES01 c20367 331
82.5		GMI ES02 Irc27323 95 GMI GBS 6872
		GMI GBS 101811 GMI DS LB 10786
85.2 -	1 18	GMI ES17 c16539 472
85.7 -	1	GMI_ES14_c18975_277
86.8 -11	1 N	GMI DS LB 6241
94.8 -		GMI ES14 c16101 333 GMI GBS 102428
95.9 -1/		GMI DS LB 10489
107.1 🖑	🛚	GMI ES15 c32 761
116.0 -{		GMI_ES15_c14533_341
119.3 -//		GMI_GBS_86839
131.1 ^{_/}	111	BA grs c10318 236
153.2 -		GMI_GBS_67251 GMI_ES22_c7478_431
154.8	₩.	GMI_ES17_c5923_221
155.3 -⁄	~	• GMI_ES14_c19259_657

0.0	∠GMI ES15 c18154 86
2.7 -	GMI ES03 c4446 528
	GMI ES15 c7179 388 GMI ES03 c9610 119
4.3-7	MGMI ES03 c12158 191
5.4	GMI ES22 c182 218
//	GMI ES02 c38195 533 GMI ES05 c24433 354
8.1 - ///-	GMI ES15 c3169 222 GMI DS LB 7769
9.2 -///-	GMI ES02 c4818 463 GMI ES03 c1515 642
	GMI DS CC7482 102 GMI DS LB 10262
11.4 -	GMI DS CC7543 80
17.5 - 17.5	GMI ES01 c22916 27
22.5	0Pt-16952
35.9 -//	GMI_ES08_c5133_835
39.3	YGMI_DS_A3_213_352 GMI_ES_LB_8487
53.4	✓ oPt-1661
60.6 \	/ GMI_ES05_Irc9304_238
624-	GMI_ES05_Irc23969_245 GMI_ES05_c15094_281
03.4	IGMI_GBS_16746
65.1	GMI_DS_LB_7746
66.2 / =	GMI_ES_CC10618_207 GMI_ES05_c5751_396
//	IGMI_DS_CC5554_57
66.7 -//	GMI_ES22_c1834_454 GMI_ES05_c16357_412
71.1 -/	GMI_ES02_c24274_326
71.7	GMI_DS_LB_6584
85.9 -/	NGMI_ES_CC8945_103 GMI_ES17_c9013_268
	GMI_GBS_86251 GMI_ES03_c8203_496
100.1 -	GMI_ES02_c2988_293 GMI_ES03_c12275_310
	IGMI_ES17_c10605_81
101.9	• GMI_GBS_92039
102.5 🌾	GMI_ES03_c18040_202 GMI_ES03_c7869_200
103.1 -//	GMI_ES01_lrc17915_335
104.3 -//	GMI_ES_CC11649_76
104.9 🎢	GMI_ES15_c345_181
116.3 -/	1 oPt-2785
118.1	• oPt-9709

#### 05021-6

0.0 T	GMI_ES01_c12222_428	
6.0 r	GMI_ES22_c13166_513 GMI_ES01_c17867_331	
23.7 🖞	GMI_ES02_Irc12751_147 GMI_DS_CC10035_89	
24.2	GMI_ES02_c4510_183	
24.7	GMI_GBS_95496	
26.4	GMI_ES_LB_9861	
<u></u>	GMI_ES03_Irc23007_427 GMI_DS_LB_4510	
28.6	GMI_ES22_c2572_214 GMI_DS_CC9093_95	
8	IGMI_DS_CC7847_62	
29.1	/ GMI_GBS_18001	
NE VE	GMI_ES02_Irc18844_168 GMI_GBS_12492	
一月	GMI_ES02_c34690_199 GMI_DS_LB_1821	
30.2	GMI_GBS_/20 GMI_ES17_C5666_258	
	GMI_ES02_c1481_139 GMI_GBS_66708	
/H	GMI_GBS_109228 GMI_DS_CC1776_314	
	IGMI_ES15_C10332_306 GMI_ES05_C6476_469	
42.6 - 1	* 0PE-11380	
45.7	GMI_GBS_6566 GMI_ES17_C2308_1026	(
18	IGWI_D5_CC9440_445	
46.3 🗐 🗕	GMI_ES01_014139_496 GMI_GB3_24039	
	ICMI_ES01_0005_677_CMI_DS_CC7705_77	
46.9	GMI_E301_012903_077 GMI_D3_007795_77	
47.5	GMI_ES03_c2934_220_GMI_ES01_c17004_407	
517	GML GBS 3516	1.
53.0	GML ES02 c15952 348 GML ES02 c28827 474	1.
61.0	IGMLES CC4638 119 GML GBS 51953	15
63.7	GMI ES03 c11465 78 GMI ES02 c2118 202	1
64.8	GMI ES15 c10645 354 GMI ES05 c1798 423	10
73.2	oPt-11599	11
90.3	oPt-5847	
1000	GMI_ES15_c14554_458 GMI_ES02_c247_241	
102.0-41-	GMI_ES_LB_4976	2
109.3	GMI_ES14_c453_394 GMI_ES01_c5633_477	2
109.8 -0	GMI_ES03_c11517_459	
114.4	GMI_DS_LB_5045 GMI_ES22_c2410_401	
116.6	GMI_DS_LB_10336	
117.7	GMI_ES14_c5483_345	
119.9	GMI_ES22_c5422_320	
133.9	GMI_ES05_c2253_434	
139.6	GMI_ES01_c14606_61	
140.8	GMI_ES02_C310_458	
141.4	GMI_ES_LB_9771	
144.4	GWI_ESUI_IC914/_285	
151.1 -	"GMI_ES01_01913_998 GMI_ES03_01394_130	
		49

0.0 r		GMI ES15 c6458 250
1.1		GMI_ES02_c17364_288 GMI_ES05 c11331_441
		IGMI ES03 c13946 240 GMI ES05 c26190 676
221		GMI ES02 c2221 329 GMI ES15 c8606 394
		GMI ES17 c11616 253
35-	ù 🚺	GMI_ES22_c8005_394
42-	d 📕	GMI_ES03_lrc10769_351
4.9	W 🚺	GMI GBS 9614 GMI ES02 c13236 178
14.9	W 🚺	GMI ES05 c17311 95
15.4 -		GMI ES05 c553 469
17.0	st.	GMI ES05 c20981 389
18.6 -	N 🖊	GMI ES05 c2760 657
19.1 -	17	- GMI ES22 c11452 253
34.2 -	.     .	GMI_ES02_c15552_416
35.3 -		GMI ES05 c26873 318
05.0	×	GMI DS LB 5972 GMI ES02 c11794 636
35.8 -	71 N	GMI DS LB 1766
36.3	í <b>A`</b>	GMI ES CC14804 235
40.4		GMI ES14 c769 500 GMI DS LB 10794
43.4 -	//	GMI_ES01_Irc8208_413 GMI_ES03_c21288_339
100		GMI_ES02_c12844_488 GMI_ES17_c653_217
43.9 -		GMI ES15 c3717 245
46.1		GMI GBS 21414
51.2 ^{-J}	M 🖪	GMI DS CC6822 86 GMI ES17 c3006 879
	N 🛛	GMI ES22 c1052 894 GMI ES15 c10501 398
50.4		GMI ES15 Irc9414 222 GMI ES CC12161 200
53.4 -	nel.	GMI ES05 c9138 267 GMI GBS 90539
1	() <b>- 1</b>	GMI_ES22_c18205_366
53.9		GMI_ES22_c8057_338
59.0	ii 👖	GMI_ES17_c3625_404
60.1	0-0	GMI_ES01_c16767_69 GMI_ES02_c3374_73
72.5		GMI_DS_oPt-1466_323
73.0	11	GMI_ES01_c3480_411
90.8		GMI_ES14_c10034_107
072	- <b>1</b>	GMI_ES17_c4047_360 GMI_ES17_c4047_539
91.5	<u>"-1</u>	IGMI_GBS_19697 GMI_ES03_c6656_436
09.5 4	N <b>- </b>	GMI_ES22_c3573_197 GMI_ES17_c4716_700
90.0	ή 📘	IGMI_ES15_c12291_689
100.1	A	GMI_ES17_09953_261
103.4 -		GMI_GBS_58632
106.8 -		GMI_ES17_c1186_142
109.6		GMI_DS_LB_7757
120.5		oPt-7110
136.1		oPt-6125
144.7 -		IGMI_ES15_c18028_289 GMI_ES17_c5197_503
150.0 -		GMI_ES05_c26263_333
150.5		GMI_ES05_04270_561
151.0 -		GMI_ES15_06191_3/0
153.2		GNI_ESU2_015089_196
154.9		GMI_ESU2_02/14_3/3
171.2		GNI_ESU5_IC12954_281
171.7		GMI_ESZZ_013490_299 GMI_ES01_03990_524
		TGMI_GBS_57836

#### 05021-7.2

05021-5



### 05021-7.1

0.0	0Pt-14651	050
2.6	0PF-12279 GML ES05 Irc10874 605	0.0-
1.1	GMI GBS 38366 GMI DS LB 10188	0.0
8.3 -	GMI_ES02_c16231_321	
10.0 -	GMI_ES05_c9987_517	
19.9	GMI_ES01_01513_651	
20.4 -	GMI_ES_LD_11072	
24.2 -	IGMI ES LB 4317 GMI ES LB 4318	
30.1 -	oPt-5521	
34.7 -	• oPt-8261	
48.2	GMI_ES15_C6451_437	
48.7 -	GML_ESUT_C10257_104	
69.0 -	GMI GBS 115834 GMI DS LB 2910	
	GMI_ES02_c4957_300	
	GMI_ES01_c1275_493 GMI_ES13_c18923_600	
74 5	GMI_ES_CC/307_489 GMI_ES22_09256_285	
/1.5 m	GMI_GB3_102741 GMI_E302_012013_349	
	IGMI DS CC10034 60	
	GMI_ES15_c14857_328 GMI_ES_LB_11223	
	GMI_ES01_c13746_583 GMI_ES01_c8527_320	
72.7	GMI_ES15_C4721_315 GMI_ES17_C573_426	
W-	GML DS LB 7816 GML DS LB 3957	13 1 -
	GMI GBS 17655 GMI GBS 34487	
91	GMI_ES05_c3839_412 GMI_ES05_c16268_365	
79.8 -	GMI_ES17_c6857_627 GMI_GBS_116433	14.8-
	GMI_GBS_5298 GMI_ES05_c11265_115	
l l	IGMI_GD5_47090 GMI_GD5_23743	164-
80.3 J	AGMI DS LB 812 GMI GBS 5207	
V	GMI_GBS_41996 GMI_ES22_c14022_362	
815	GMI_GBS_19541 GMI_ES01_c2511_375	
1.0	GMI_ES02_c677_709 GMI_GBS_13559	
	IGMI_ES05_C2696_443 GMI_ES05_C27097_64	050
83.4 -   =	IGMI ES21 c1671 340	
84.0 -/// =	GMI_ES22_c884_375	0.0
93.7	0 oPt-5552	
101.0	0PE-1652 0PE-17793	
102.2	oPt 4615	
108.9		
100.0	oPt-12985	
111.7	oPt-12985 oPt-15250	
111.7 120.6	oPt-12985 oPt-15250 GML_ES17_c15366_116	
111.7 - 1 120.6 - 1 121.1 - 1 121.6 - 1	OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2775_246	
111.7 - 120.6 - 121.1 - 121.6 -	OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 IGMI_ES17_c1242_703 GMI_ES_LB_7322	10.9-
111.7 120.6 121.1 121.6 122.7	OPt-12985 OPt-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c172448_349 GMI_ES02_c28150_198	10.9 -
111.7 120.6 121.1 121.6 122.7	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c17448_349 GMI_ES02_c28150_198 GMI_ES15_c5837_115	10.9 —
111.7 120.6 121.1 121.6 122.7	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 - GMI_ES15_c7755_266 - GMI_ES15_c7755_266 - GMI_ES17_c2276_224 - GMI_ES17_c1242_703 GMI_ES_LB_7322 - GMI_ES15_c17448_349 GMI_ES02_c28150_198 - GMI_ES15_c5837_115 - GMI_ES15_c5837_157 - GMI_ES15_c5837_157 - GMI_ES15_c5837_157	10.9 —
111.7 J 120.6 J 121.1 J 121.6 J 122.7 J	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 - GMI_ES15_c7755_266 - GMI_ES15_c7755_266 - GMI_ES17_c1242_703 GMI_ES_LB_7322 - GMI_ES15_c17448_349 GMI_ES02_c28150_198 - GMI_ES15_c5837_115 - GMI_ES01_c6200_493 - GMI_ES01_c6200_493 - GMI_ES15_c6808_218_GMI_ES17_c19933_225 - GMI_ES01_c12756_163 - GMI_ES01_c12756_163 - GMI_ES01_c12756_163 - GMI_ES15_c6805_218_GMI_ES17_c19933_225 - GMI_ES01_c12756_163 - GMI_ES15_c6805_218_GMI_ES17_c19933_225 - GMI_ES15_c7805_163 - GMI_ES15_c7805_163 - GMI_ES15_c7805_163 - GMI_ES15_c785_163 - GMI_ES15_c785_173 - GMI_ES15_073 - GMI_ES15_07	10.9-
111.7 J 120.6 121.1 J 121.6 J 122.7 J	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 [GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c717448_349 GMI_ES02_c28150_198 IGMI_ES15_c5837_115 GMI_ES01_c6200_493 [GMI_ES15_c6808_218 GMI_ES17_c19933_225 IGMI_ES17_c8008_218 GMI_ES17_c19933_225 IGMI_ES17_c8008_49 GMI_ES_LB_11611	10.9
111.7 J 120.6 J 121.1 J 121.6 J 122.7 J	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES15_c7748_349 GMI_ES02_c28150_198 GMI_ES15_c5837_115 GMI_ES15_c58008_218 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES15_c806_849 GMI_ES17_c19933_225 GMI_ES17_c806_849 GMI_ES2_LB_11611 GMI_ES8_24408 GMI_ES02_c17762_447	10.9 16.7 17.2
111.7 J 120.6 J 121.1 J 121.6 J 122.7 J	OPt-12985 OPt-12985 OPt-15250 GMI_ES17_c15366_116 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c17448_349 GMI_ES02_c28150_198 GMI_ES15_c5837_115 GMI_ES15_c5837_15 GMI_ES15_c8008_218 GMI_ES17_c19933_225 IGMI_ES01_c12759_163 IGMI_ES15_c806_849 GMI_ES_LB_11611 GMI_ES_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_Irc14087_138	10.9 16.7 17.2
111.7 120.6 121.1 121.6 122.7 123.8 124.3	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c7148_349 GMI_ES02_c28150_198 GMI_ES15_c75448_349 GMI_ES02_c28150_198 GMI_ES15_c5837_115 GMI_ES01_c6200_493 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES17_c806_849 GMI_ES_LB_11611 GMI_ES17_c806_849 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_IC14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_GBS_77_c604_41_OMI_ES01_IC14087_138 GMI_GBS_80720 GMI_ES02_c3392_447	10.9 − 16.7 〜 17.2 〜
111.7 1 120.6 1 121.1 1 121.6 1 122.7 1 123.8 1 124.3 1	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c17448_349 GMI_ES02_c28150_198 GMI_ES15_c5837_115 GMI_ES01_c6200_493 GMI_ES15_c6808_218 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES17_c806_849 GMI_ES12_LB_11611 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_Ic14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES17_c5090_114 GMI_ES01_Ic14087_138 GMI_ES17_c609_114 GMI_ES01_Ic10107_481 GMI_GBS_825_C6193_U_ES12_c445_511	10.9
111.7 . 120.6 . 121.1 . 121.6 . 122.7 . 123.8 . 124.3 . 129.4 .	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES15_c7755_266 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c17448_349 GMI_ES02_c28150_198 GMI_ES15_c6803_115 GMI_ES01_c6200_493 GMI_ES15_c6008_218 GMI_ES17_c19933_225 GMI_ES01_c12759_163 GMI_ES17_c806_849 GMI_ES1_E_11611 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_IRC14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES17_c5090_114_GMI_ES01_c10107_481 GMI_GBS_35_GMI_ES17_c1465_511 GMI_GBS_363_GMI_ES17_c1465_511 GMI_GBS_363_GMI_ES17_c1465_511 GMI_GBS_363_GMI_ES17_c1465_511 GMI_GBS_363_GMI_ES02_c3206_288	10.9
111.7 4 120.6 4 121.1 1 121.6 4 122.7 1 123.8 4 124.3 1 129.4 1	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES15_c7755_266 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c7847_349 GMI_ES02_c28150_198 GMI_ES15_c6803_115 GMI_ES01_c6200_493 GMI_ES15_c6803_218 GMI_ES17_c19933_225 GMI_ES01_c12759_163 GMI_ES17_c806_849 GMI_ES_LB_11611 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_ICT14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES17_c5090_114 GMI_ES01_c10107_481 GMI_GBS_85 GMI_ES17_c1465_511 GMI_ES03_c13481_505 GMI_ES05_c2906_288 GMI_ES05_c11155_383 GMI_DS_LB_4840	10.9- 16.7~ 17.2~ 24.3-
111.7 J 120.6 1 121.1 1 121.6 J 122.7 J 123.8 1 124.3 J 129.4 J	OPt-12985 OPt-12985 OPt-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c17448_349 GMI_ES02_c28150_198 GMI_ES15_c17448_349 GMI_ES17_c19933_225 GMI_ES01_c6200_493 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES01_c12759_163 GMI_ES15_c8008_218 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_c10107_481 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES15_c6193_114_GMI_ES01_c10107_481 GMI_GBS_835 GMI_ES17_c1465_511 GMI_ES05_c11348_505 GMI_ES05_c2906_288 GMI_ES05_c1115_383 GMI_DS_LB_4840 GMI_ES03_c2839_38 GMI_GBS_103591	10.9 16.7 17.2 24.3
111.7 J 120.6 1 121.1 1 121.6 1 122.7 J 122.7 J	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c7148_349 GMI_ES02_c28150_198 GMI_ES15_c7036_218 GMI_ES17_c19933_225 IGMI_ES01_c12759_163 IGMI_ES15_c8008_218 GMI_ES17_c19933_225 IGMI_ES17_c806_849 GMI_ES_LB_11611 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_IIC14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 IGMI_ES15_c501_4GMI_ES01_c10107_481 GMI_ES3_c5GMI_ES17_c1465_511 GMI_ES3_c13481_505 GMI_ES05_c2906_288 GMI_ES03_c13481_505 GMI_ES05_c2906_288 GMI_ES03_c2839_38 GMI_GBS_103591 GMI_GBS_00720 GMI_ES02_c07845_455	10.9 16.7 17.2 24.3
111.7 4 120.6 4 121.1 1 121.6 4 122.7 4 122.7 4 123.8 4 124.3 4 129.4 4	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c77448_349 GMI_ES02_c28150_198 GMI_ES15_c7808_218 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES01_IC14087_138 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_IC14087_138 GMI_GBS_855 GMI_ES17_c1465_511 GMI_ES17_c5090_114 GMI_ES01_c10107_481 GMI_ES03_c13481_505 GMI_ES05_c2906_288 GMI_ES03_c2839_38 GMI_OS_LB_4840 GMI_ES03_c2839_38 GMI_GS_103591 GMI_GBS_108124 GMI_GBS_0076_GMI_GBS_21446_ GMI_GBS_0076_GMI_GBS_21446_ GMI_GBS_0076_GMI_GBS_21446_ GMI_GBS_0076_GMI_GBS_21446_ GMI_GBS_0076_GMI_GBS_21446_ GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53 GMI_GBS_0076_GMI_GBS_21446_53	10.9 16.7 17.2 24.3 30.7
111.7 4 120.6 4 121.1 1 121.6 4 122.7 4 122.7 4 122.8 4 124.3 4 129.4 4 129.4 4	OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c71448_349 GMI_ES02_c28150_198 GMI_ES15_c5837_115 GMI_ES01_c6200_493 IGMI_ES15_c692.18 GMI_ES17_c19933_225 IGMI_ES17_c806_849 GMI_ES12_LB_11611 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_Ic14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES15_c6193_143 GMI_ES01_Ic14087_138 GMI_GBS_835 GMI_ES12_c1465_511 GMI_ES8_35 GMI_ES15_c1465_511 GMI_ES03_c13481_505 GMI_ES05_c2906_288 GMI_ES05_c11155_383 GMI_DS_LB_4840 GMI_ES03_c13481_505 GMI_ES05_c2906_288 GMI_ES03_c2839_38 GMI_DS_LB_4840 GMI_ES03_c13481_24 IGMI_GBS_0976 GMI_GBS_24442 IGMI_GBS_90976 GMI_GBS_90976 GMI_GBS_909	10.9
111.7 4 120.6 4 121.1 4 122.7 4 122.7 4 122.8 4 124.3 4 129.4 4 129.9 4 129.9 4	OPE-12985 OPE-12985 OPE-12985 OPE-15250 GMI_ES17_c1256266 GMI_ES15_c7755_266 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES17_c1242_703 GMI_ES02_c28150_198 GMI_ES15_c5837_115 GMI_ES01_c6200_493 GMI_ES15_c6808_218 GMI_ES17_c19933_225 GMI_ES15_c6908_218 GMI_ES17_c19933_225 GMI_ES15_c693_143 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_IC14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES15_c6193_143 GMI_ES01_IC14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES17_c5090_114 GMI_ES01_IC14087_138 GMI_GBS_835 GMI_ES17_c1465_511 GMI_ES03_c13481_505 GMI_ES05_c2906_288 GMI_ES05_c11155_383 GMI_DS_LB_4840 GMI_ES03_c2839_38 GMI_GBS_103591 GMI_GBS_09076 GMI_GBS_24442 GMI_ES02_c14270_228 GMI_ES03_c1840 GMI_ES03_c11836_276	10.9
111.7 4 120.6 4 121.1 4 121.6 4 122.7 4 122.7 4 122.8 4 124.3 4 129.4 4 129.9 4 129.9 4 130.4 4 139.7 1	OPt-12985 OPt-12985 OPt-15250 GMI_ES17_c15366_116 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c17448_349 GMI_ES02_c28150_198 GMI_ES15_c17448_349 GMI_ES02_c28150_198 GMI_ES15_c17448_349 GMI_ES17_c19933_225 GMI_ES01_c12759_163 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES01_c12759_163 GMI_ES15_c6193_143 GMI_ES01_c1017_481 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_c1017_481 GMI_GBS_835 GMI_ES17_c1465_511 GMI_ES17_c509_114 GMI_ES01_c1017_481 GMI_ES03_c13481_5503 GMI_ES15_c2906_288 GMI_ES03_c13481_55383 GMI_DS_LB_4840 GMI_ES03_c1839_38 GMI_GBS_103591 GMI_GBS_90976 GMI_GBS_24442 GMI_ES02_c14270_228 GMI_ES02_c185_0441 GMI_ES03_c185_0441 GMI_ES03_c185_046 GMI_ES03_c11836_276 GMI_ES03_c12850_294	10.9 16.7 ~ 17.2 24.3 30.7 34.6 ~-
111.7 4 120.6 1 121.1 1 121.6 1 122.7 5 123.8 1 124.3 5 129.4 1 129.4 1 129.9 1 130.4 1 139.7 1 141.3 1	OPt-12985 OPt-12985 OPt-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c71748_349 GMI_ES02_c28150_198 GMI_ES15_c5837_115 GMI_ES15_c708_218 GMI_ES17_c19933_225 IGMI_ES01_c12759_163 IGMI_ES15_c8008_218 GMI_ES17_c19933_225 IGMI_ES17_c806_849 GMI_ES_LB_11611 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_IIC14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 IGMI_ES15_c509_114 GMI_ES01_c10107_481 GMI_GBS_85_GMI_ES17_c1465_511 GMI_ES03_c13481_505 GMI_ES05_c2906_288 GMI_ES05_c11155_383 GMI_GBS_103591 GMI_GBS_07281_186 GMI_DS_CC7846_153 GMI_GBS_90976 GMI_GBS_24442 IGMI_GBS_60480 GMI_ES03_c11836_276 GMI_GBS_09046 GMI_ES03_c11836_276 IGMI_GBS_09041_405 IGMI_GBS_09041_405 IGMI_GBS_09041_405 IGMI_GBS_09041_405 IGMI_GBS_09041_405 IGMI_GBS_09041_405 IGMI_GBS_09041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGMI_GBS_04041_405 IGM	10.9 16.7 ~ 17.2 24.3 30.7 34.6 ~-
111.7 4 120.6 4 121.1 121.6 4 121.6 4 121.6 4 121.6 4 121.6 4 122.7 4 123.8 4 124.3 4 129.4 4 139.7 - 141.3 - 165.4 -	OPt-12985 OPt-12985 OPt-15250 GMI_ES17_c15366_116 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c1242_703 GMI_ES_LB_7322 GMI_ES15_c7148_349 GMI_ES02_c28150_198 GMI_ES15_c7048_349 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES15_c8008_218 GMI_ES17_c19933_225 GMI_ES17_c806_849 GMI_ES_LB_11611 GMI_GBS_24408 GMI_ES02_c17762_447 GMI_ES15_c6193_143 GMI_ES01_Ic14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES15_c6193_143 GMI_ES01_Ic14087_138 GMI_GBS_80720 GMI_ES02_c3392_447 GMI_ES15_c6193_143 GMI_ES01_c10107_481 GMI_GBS_805_C011ES17_c1465_511 GMI_ES03_c13481_505_GMI_ES05_c206_288 GMI_ES03_c13481_505_GMI_GBS_103591 GMI_GBS_108124 GMI_GBS_90976_GMI_GBS_24442 GMI_GBS_90976_GMI_GBS_24442 GMI_ES02_c14270_228 GMI_GBS_80480 GMI_ES03_c11836_276 GMI_ES05_c13201_125 GMI_GBS_80600 GMI_ES05_c13201_125 GMI_GBS_505_c13201_125 GMI_GBS_505_c13201_125 GMI_ES05_c13201_125 GMI_ES05_c13201_125 GMI_ES05_c13201_125 GMI_ES05_c13201_125	10.9 16.7 17.2 24.3 30.7 34.6
111.7 4 120.6 4 121.1 1 121.6 4 122.7 4 122.7 4 122.8 4 124.3 4 129.4 4 129.4 4 129.9 - 130.4 - 139.7 4 139.7 - 141.3 - 165.4 - 165.4 - 165.9 -	OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985 OPE-12985	10.9 16.7 17.2 24.3 30.7 34.6

### 05021-7.3











0.0 J	r oPt-7556	6
5.2	oPt-17014	6
21.4	GMI_DS_LB_5997	30
32.6 -	GMI_ES02_c25243_77	
35.9 -	GMI_GBS_99439 GMI_ES02_c38462_575	41
37.0 -	IGMI_DS_0Pt-17670_78 GMI_ES05_c14477_258	41
38.6 -	GMI_ES05_IIC16479_180 GMI_ES05_C23907_190	
20.4	IGMI_ES17_012906_275	43
39.11	GMI_ES05_C3230_665	45
30.6 -	GML_503_32290 GML_1314_014307_103	49
00.0	GML_GBS_99288_GML_ES17_c3477_85	50
45.4	GML ES17 c12536 430	50
46.5	GMI DS LB 1859 GMI ES02 c8737 267	01
53.6	GMI ES03 c5662 209	51
54.7	GMI_ES02_c8277_506	
58.0	GMI_GBS_100951	52
58.5	GMI_ES15_c5831_418	
59.6	GMI_ES03_c2612_410	54
1	GMI_ES05_c7724_591 GMI_DS_LB_1354	56
61.2 ₁	GMI_ES17_c3134_489 GMI_ES02_c22830_111	60
	/IGMI_ES15_05554_377	62
62.3 -	GMI_ES02_C2959_310 GMI_ES02_C4007_487	02
	//GWI_GBS_66666	63
65.5	/ GMI_ESUS_IIC21145_126	
66.6	GMI_C3_CD_12079 GMI_D3_CD_10039	65
00.0	IGMI_ES05_Irc12441_760_GMI_GBS_72768	66
67.1 -//	IGMI_ES17_c18901_119	76
68.2	M GMI ES02 c15462 413	96
85.3	GMI_ES17_c12269_176	00
104.5	- GMI_GBS_78545	87
105.0 -//	GMI_DS_LB_2922	90
107 7 -//=	GML ES22 c10434 196	
···· //	0111_2022_010101_100	
110.4	GMI_ES05_c1719_239	0
110.4 115.3	GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c4125_413	0
110.4 115.3 119.5	GMI_ES05_c1719_239 GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400	0
110.4 115.3 119.5	GMI_ES05_c1719_239 GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 IGMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6420_CMI_ES14_c1701_574	<b>0</b> (
110.4 115.3 119.5 121.7	GMI_ES05_c1719_239 GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 IGMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_580	<b>0</b> (
110.4 115.3 119.5 121.7	GMI_ES05_c1719_239 GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 IGMI_ES14_c1133_505_GMI_DS_LB_10723 GMI_DS_LB_6430_GMI_ES14_c1791_574 IGMI_ES15_c2518_589 GMI_ES05_c4008_426_GMI_ES02_c3556_502	<b>0</b> (
110.4 115.3 119.5 121.7 122.4 123.8	GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES05_c4098_426 GMI_ES02_c3556_502 GMI_ES05_c4098_426 GMI_ES02_c3556_502 GMI_ES05_c4098_9GMI_ES05_c8628_73	0 (
110.4 115.3 119.5 121.7 122.4 123.8 126.0	GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES05_c4098_426 GMI_ES02_c3556_502 IGMI_GBS_94839 GMI_ES05_c8628_73 GMI_ES14_c5917_794	<b>0</b> (
110.4 115.3 119.5 121.7 122.4 123.8 126.0 126.5	GMI_ES05_c1719_239 GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES05_c4098_426 GMI_ES02_c3556_502 GMI_ES05_c4098_426 GMI_ES05_c8628_73 GMI_ES14_c5917_794 GMI_ES_C27433_103	<b>0</b> 0
110.4 115.3 119.5 121.7 122.4 123.8 126.0 126.5 126.5 128.7	GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES05_c4098_426 GMI_ES02_c3556_502 GMI_GBS_94839 GMI_ES05_c8628_73 GMI_ES14_c5917_794 GMI_ES12_C7433_103 GMI_GBS_107339 GMI_ES05_c10102_262	<b>0</b> (
110.4 115.3 119.5 121.7 122.4 123.8 126.5 126.5 126.5 128.7 128.7 129.2	GMI_ES05_c1719_239 GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES05_c4098_426 GMI_ES02_c3556_502 GMI_GBS_94839 GMI_ES05_c8628_73 GMI_ES14_c5917_794 GMI_ES14_c5917_794 GMI_ES_CC7433_103 GMI_GBS_107339 GMI_ES05_c10102_262 GMI_ES05_c10573_58	0 0 0
110.4 115.3 119.5 121.7 122.4 123.8 126.5 126.5 128.7 128.7 129.2 129.7	GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES15_c2518_589 GMI_ES5_94899 GMI_ES05_c8628_73 GMI_ES54_c5917_794 GMI_ES14_c5917_794 GMI_ES_C7433_103 GMI_ES5_C7433_103 GMI_ES5_05_c10573_58 GMI_ES17_c24_198	0 0 0
110.4 115.3 119.5 121.7 122.4 123.8 126.0 126.5 128.7 129.7 129.7 129.7 129.7 138.1	GMI_ES05_c1719_239 GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES5_c4098_426 GMI_ES02_c3556_502 HGMI_GBS_94839 GMI_ES05_c8628_73 GMI_ES14_c5917_794 GMI_ES_CC7433_103 GMI_ES_CC7433_103 GMI_ES505_c107339 GMI_ES05_c10102_262 GMI_ES05_c10573_58 GMI_ES17_c24_198 - OPt-0553	0 ( ( (
110.4 115.3 119.5 121.7 122.4 123.8 126.5 126.5 126.5 128.7 129.7 129.7 129.7 138.1 138.1 138.1 138.1	GMI_ES05_c1719_239 GMI_ES03_c4125_413 GMI_ES03_c12156_400 GMI_ES14_c1133_505 GMI_DS_LB_10723 GMI_DS_LB_6430 GMI_ES14_c1791_574 GMI_ES15_c2518_589 GMI_ES05_c4098_426 GMI_ES02_c3556_502 IGMI_GBS_94839 GMI_ES05_c8628_73 GMI_ES14_c5917_794 GMI_ES_CC7433_103 GMI_GBS_107339 GMI_ES05_c10102_262 GMI_ES05_c10573_58 GMI_ES17_c24_198 OPL0553 OPL13898	0 ( (
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0.0 \ oPt-5020
6.1 \ / 0Pt-10221 6.8 \ / 0Pt-11595
30.2 1 GMI_ES17_c8314_651 GMI_ES01_c24259_455
41.0-1 GMI_ES05_c14960_147 GMI_GBS_78960
GMI_ES15_c2202_214 GMI_ES15_c17272_315
43.2 - GMI_ES01_c12816_41
45.8 - 10 GMI_ES22_C4889_60
49.8 1 GMI_DS_LB_4706 GMI_ES15_c4157_324
51.4 JUIGMI_ES15_c16874_366 GMI_DS_LB_9347
51.9 J GMI_ES01_c27997_135 GMI_ES15_c5396_587
52 4 GMI_ES02_c16824_373 GMI_ES03_c3636_522
GMI_ES15_66921_451
54.0 GMI_GBS_66062
60.1 GMI_ES02_Irc14207_485
62.8 – GMI_ES15_c2305_730 GMI_ES_CC3511_395
63 3 GMI_GBS_24858 GMI_GBS_19438
65.5 GMI_ES05_c2185_605_GMI_ES15_c120/1_400
66.0 ^J - VIGMI_ES01_c1307_465 GMI_GBS_80580
76.0 GMI_ES17_c1537_1127 GMI_ES17_c10505_001
86.1 GMI_ES05_c6752_720
87.2 GMI_GBS_61527 GMI_GBS_90753
90.0 GMI ES17 08241 378
05021-14.2
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05021-14.2 0.0 - 0Pt-10501 6.9 GMI_GBS_112167 9.1 GMI_DS_A3_340_378 GMI_DS_LB_4867 22.6 GMI_GBS_83715 GMI_ES_LB_11741 GMI_GBS_46202 23.1 GMI_DS_LB_1995 23.6 GMI_ES02_c11450_462
05021-14.2 0.0 - oPt-10501 6.9 GMI_GBS_112167 9.1 GMI_DS_A3_340_378 GMI_DS_LB_4867 22.6 GMI_GBS_83715 GMI_ES_LB_11741 GMI_GBS_46202 23.1 GMI_DS_LB_1995 23.6 GMI_ES02_c11450_462 27.1 GMI_ES17_c3665_257
05021-14.2 0.0
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05021-14.2 0.0 - 0Pt-10501 6.9 GMI_GBS_112167 9.1 GMI_DS_A3_340_378 GMI_DS_LB_4867 22.6 GMI_GBS_83715 GMI_ES_LB_11741 GMI_GBS_46202 23.1 GMI_DS_LB_1995 23.6 GMI_ES02_c11450_462 27.1 GMI_ES17_c3665_257 30.0 IGMI_DS_LB_8147 GMI_ES02_Irc21522_135
05021-14.2 0.0 - 0Pt-10501 6.9 GMI_GBS_112167 9.1 GMI_DS_A3_340_378 GMI_DS_LB_4867 22.6 GMI_GBS_83715 GMI_ES_LB_11741 GMI_GBS_46202 23.1 GMI_ES_LB_1995 23.6 GMI_ES02_c11450_462 27.1 GMI_ES17_c3665_257 30.0 GMI_DS_LB_8147 GMI_ES02_lrc21522_135 36.3 - 0Pt-11716





#### 05026-1.1







### 05026-1.2

GMI_ES05_c16637_406 GMI_ES15_c5908_278 GMI_ES05_c20813_204 GMI_DS_LB_1713 GMI_ES15_c276_702 14.5 1

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- 17.4 18.5 11
- GM_DS_LB_1713 GMI_ES15_c276_702

   GMI_ES15_c6153_392 GMI_ES14_c5428_351

   GMI_ES15_c6153_392 GMI_ES14_c5428_351

   GMI_ES16_12570_390

   IGML_DS_LB_9301 GMI_ES01_c22545_242

   IGML_SE_LB_11702 GMI_ES05_c2407_680

   IGML_ES_c751_580 GMI_ES05_c12229_212

   IGML_ES1_c751_580 GMI_ES05_c12229_212

   IGML_ES1_c2254_56 GMI_ES02_c4801_78
  19.7
- IGMI_ES17_220294_211 GMI_ES11_22261_56 GMI_ES22_c4891_78 GMI_ES02_c13712_506 GMI_ES02_c13712_383 IGMI_ES22_c667_711 GMI_ES02_c19578_292 20.3
- 22.6 GMI_DS_LB_10291
- 22.6 GMI_CS_LB_10291 23.2 GMI_ES22_c3138_583 1GMI_ES02_c14384_591 GMI_ES22_c2788_756 24.3 GMI_ES15_c2773_491

#### 05026-3

	ONIL OB0. 00005
0.0 1	GMI_GBS_20325 GMI_ES05_c12055_245
16.0	GMI_L303_C12033_243
10.9	CMI E815 c1422 97
20.7	CMI E902 c6104 429
23.91	GWI_E505_06194_420
20.1	CMI_003_09003
20.3 1	CMI DS LB 7522 CMI DS LB 9152
26.9	GMI_DS_LD_7525 GMI_DS_LD_0152
200	GMLES05_015200_560 GMLES01_011627_414
20.0 1	GMI_ES IS_0005_000
45.4 1	GMI_ESUS_C3356_266 GMI_EST7_C3764_732
	GWI_DS_CC10995_70 GWI_GBS_64241
	GMI_ESU1_0461_1288 GMI_ES_CC10160_63
46.5	GML_E5_CC1007_01GML_D5_L6_2902
U 🚺	GWI_DS_CC0134_229 BA_gIS_C0269_136
1 - <b>0</b>	GMI_ES14_C14040_375 GMI_ES05_C20438_78
	GMI_ES05_011113_348 GMI_GBS_7855
49.4	GMI_GBS_113531
58.7	GMI_ES05_019273_362 GMI_ES02_019258_432
67.3 1	GMI_GBS_13773 GMI_DS_0Pt-18257_376
67.9	GMI_ES01_08255_502
69.6 1	GMI_ES02_010035_217
70.2	GMI_ES02_C24184_389
72.5	GMI_ES14_08386_549
73.1	GMI_ES22_0526_367
73.7	GMI_ES02_01819_259
76.2	GMI_GBS_23770
80.8	GMI_ES05_c10478_455
83.1	AGMI_GBS_102428 GMI_ES14_c16101_333
84.2	GMI_DS_LB_6241
81.1	GMI_ES14_018975_277
883-A	GMI_ES17_016539_472 GMI_GBS_101811
	IGMI_DS_LB_10786
92.5 2	NGMI_ES02_IC27323_95 GMI_GBS_6872
111.2	GMI_ES17_C4051_315 GMI_GBS_79375
H	IGMI_ES03_05837_61
A	GMI_ES12_08736_490 GMI_ES22_09270_194
114.2	GMI_ESU1_0377_521 GMI_GBS_39204
114.9 1	GMI_DS_LB_10721
116.3	GWI_GBS_93276 GWI_GBS_76760
// 🛝	GWI_ESUS_024965_506 GWI_ES22_02904_506
116.9	GWI_ES17_013962_600 GWI_ES05_019505_225
	GWI_ESU2_C21260_142 GWI_ES_CC7714_103
0.10	CML ESOE (1993 386 CML ESOE (31230 343
117.5	GMI_ES05_01525_206 GMI_ES05_021529_245
1	IGMI_ES02_023100_443
120.2	GWI_ES01_010727_290 GWI_DS_LB_0009
- 11	CML EQ05 (0704 100 CML EQ02 (0252 65
101.0	GWI_L303_L9704_109 GWI_E322_L2332_b3
121.3	GWI_E302_0041_720 GWI_E314_0982_713
122.0	GML ES05_020576_210
123.0 1	GMI_L303_C20370_219
124.1 1	GWI_DS_LD_0303
143.2 *	• BA_gis_010318_236

Figure A2. Molecular marker linkage map from 'Souris' x 'ND030299' F6 recombinant inbred oat population (Population 05026) (red=EST markers, blue=GBS markers, green=DArT markers).

0.0-1	
0.6	GMI_GBS_73993 GMI_ES05_c18925_99 GMI_ES03_c2590_477 GMI_ES14_c19125_558 GMI_ES05_c959_1393 GMI_GBS_102362
1.2	VGMI_ES02_c12727_423 GMI_GBS_17395
4.6	GMI_ES_LB_8487
5.8 —	GMI_DS_A3_213_352
7.2-	GMI_ES08_c5133_835
8.6	GMI_ES01_c22916_27
10.0-	GMI_GBS_57848 GMI_ES14_c18722_288
10.0	/IGMI_GBS_108402 GMI_ES03_c5596_272
13.1 \	GMI_DS_CC7482_102 GMI_DS_LB_10262
13.7 ₁	GMI_DS_CC7543_80
14.3 -	GMI_DS_LB_7769 GMI_ES03_c1515_642
1.0	GMI_ES02_c4818_463 GMI_ES_LB_9026
1	_/ GMI_ES05_c24433_354 GMI_ES17_c10605_81
15.4	GMI_ES15_c3169_222 GMI_ES02_c2988_293
	IGMI_ES02_038195_533 GMI_GBS_86251
16.1	GMI_ES03_c12275_310 GMI_ES03_c8203_496
18.3	GMI_GBS_92039
19.4	GMI_ES03_09610_119 GMI_ES22_0182_218
N	- OMI E004 10045 005
20.0	GMI_ES01_IC17915_335
20.6	GMI_ES03_012158_191
21.2~	
21.8-	
22.4	- GMI_ES_CCT1649_76
23.5-4	GMI ES14 C9791 550



## 05026-5.1

		GML GBS 2049 GML ES14 c11245 392
0.0		GML ES17 c18602 497
18.5		GML ES05 (8702 202
21.5 -		GML ES02 c16427 394
23.2		GML ES14_015814_503
20.2		CML DS 1 B 6800
24.0		CML CPS 12607
25.2 -		GWI_GB5_13007
27.9	1	GWI_ES14_01961_351 GWI_ES02_I023466_262
		IGMI_ES03_09025_529
45.3		GMI_ES02_016894_445
66.5 -		GMI_ES02_c13236_178
67.2		GMI_ES03_Irc10769_351 GMI_GBS_9614
01.2		IGMI_ES22_c8005_394
68.7		GMI_ES02_c2221_329 GMI_ES17_c11616_253
69.3 -		GMI_ES15_c8606_394
60.0		GMI_ES02_c17364_288 GMI_ES05_c11331_441
09.9	4 J	GMI_ES05_c26190_676
71.0		GMI_ES15_c6458_250
	li 📕	GMI_ES15_c19863_323 GMI_GBS_72260
73.9		GMI ES05 c18726 496 GMI ES02 c2752 372
		GMI_ES01_c25986_126 GMI_ES05_c12542_273
74.5 -	H.	GML GBS 113240
		GML GBS 103122 GML GBS 23519
75.6		GMI ES05 c789 549 GMI ES15 c1937 496
		IGML ES22 (9147 376 GML ES01 (17476 92
76.2 -		GML ES01_03480_411 GML ES02_0514_867
10.2		CML DS LB 5451 CML ES01 (rc12306 230
76.0		CML ES15 c1176 234
10.0		GMI_ES15_CT176_554
		GWI_ES05_IC27474_450 GWI_ES05_C15259_451
	QI 🚺	GWI_ES02_037327_088 GWI_DS_LB_10066
	A 🚺	GMI_ES01_C2025_808 GMI_ES02_C4756_515
		GWI_ESU1_C11753_732 GWI_ES15_C5451_344
77.4	L 🛛	GMI_ES05_013239_749 GMI_ES02_026443_296
		GMI_ES01_c8245_248 GMI_ES01_c22998_155
		GMI_ES01_c1947_88 GMI_ES02_c2693_341
	1	GMI_GBS_5940 GMI_ES17_c12009_600
	H	IGMI_ES02_c885_586 GMI_ES01_lrc8457_64
78.0 -	JEL	GMI_DS_LB_10436
	Y	GMI_ES15_c12600_230 GMI_ES15_c8706_404
70 6	Æ	_GMI_ES02_c16428_446 GMI_DS_LB_8184
10.0 -		GMI_ES01_c9205_212 GMI_ES03_c17348_240
	И 🚺	GMI_GBS_44600
70.0	(I) <b>= </b> \	GMI_ES_CC11658_395 GMI_ES02_c495_1649
19.2 -		GMI ES21 c9389 9 GMI ES15 c2810 658
80.3 -		GMI_ES15_c1393_161
	J 🛾	IGMI ES15 c16762 310 GMI ES15 c6652 253
	<b>/ – I</b> )	GMI GBS 35979 GMI ES15 c6914 663
		GMI ES03 c447 586 GMI ES17 c8372 460
		GMI GBS 19697 GMI ES17 c4047 539
80.9 -		GML ES03 c6656 436 GML GBS 9676
		GMI_ES17_c4047_360 GMI_DS_LB_6884
		GMI_ES22_c1930_948_GMI_ES15_c12818_361
		GML ES01_c3247_533 GML ES01_c9687_75
	1	GML ES15 (8101-231
815	6 🛛	GML_ES07_c4037_548
85.0		GML ES CC13304 332
01.0	6 <b> </b>	GML ES02 (15080 106
02.5		CML ES14_c12590_171
92.0	í	CMI ES02 c21425 103 CMI CBS 73404
94.0		CML ES15 c6101 370
94.1		CML ES02 c0749 570
96.0		GWI_E305_19740_379
90.0		GWI_GDS_/210 CMI_CRS_114111
90.0		
99.2		GWI_ESU2_020223_200
01.0		GMI_ES17_01186_142
08.2		GWI_GBS_15811
19.5		GMI_ES02_C5703_238
20.1		GMI_ES01_c3990_524 GMI_GBS_57836
20.1		IGMI_ES03_Irc10199_274
20.7 -		GMI_ES22_c13490_299
22.4 -		GMI_ES05_Irc12954_281
287		GML ES02 c23703 243

0.0 0.6 4.8 5.4 6.0 6.6 7.2 8.3 8.9	GMI_ES05_c497_505 GMI_ES01_c513_769 GMI_ES05_c2253_434 GMI_ES05_c2253_434 GMI_ES22_c546_592 GMI_DS_LB_5802 GMI_ES02_c5812_271 GMI_ES_LB_5045 IGMI_ES_LB_9771 GMI_ES02_c310_458 GMI_ES_CC7312_286 GMI_ES01_Irc29198_650 IGMI_ES05_c9758_147 GMI_ES01_Irc9147_285 IGMI_ES01_Irc9147_285 IGMI_ES_LB_9918 GMI_ES_LB_7662	
34.3 36.0	GMI_ES15_c10645_354 GMI_ES05_c1798_423 GMI_ES02_c2118_202 GMI_ES03_c11465_78	
52.5 53.1 54.2	IGMI_GBS_95496_GMI_ES02_c4510_183      IGMI_ES_IB_9861      GMI_GBS_109117      IGMI_ES22_c2572_214_GMI_DS_CC9093_95      IGMI_DS_CC7847_62_GMI_GBS_18001      IGMI_GBS_12492_GMI_ES02_c34690_199	
56.5 /	GMI_ES02_Irc18844_168 GMI_GBS_720 GMI_ES05_c6476_469 GMI_ES03_Irc23007_427 GMI_DS_LB_1821 GMI_ES17_c5666_258 GMI_DS_CC1776_314 GMI_GBS_109228 GMI_GBS_66708 GMI_ES02_c1481_139 GMI_DS_LB_4510_GMI_ES15_c10932_305	
73.2 ~ 75.0 ~	GMI_ES01_c17867_331 GMI_ES22_c13166_513	

### 05026-7.1

0.0	- GMI_ES17_c6425_188
00.0	- CML CBS 62001
20.0	GMI_GB5_65991
36.2 1	IGMI ES02 Irc12740 554 GMI GBS 19256
38.0	GMI DS LB 4204 GMI DS LB 10835
40.4	GMI_ES03_c1581_689
41.0	GMI_DS_LB_795
42.7	GMI_ES03_c5009_566
44.6	[GMI_ES15_01400_411_GMI_ES02_06639_318]
45.3-	GML_ES22_C2100_244
40.0 V	IGMI DS CC2571 369 GMI DS LB 9572
47.6-	- GMI ES LB 9068 GMI ES01 c4259 207
Н	GMI_DS_LB_370
//-	( GMI_ES02_c35886_610 GMI_GBS_29787
49.5	[']GMI_GBS_107491 GMI_ES17_c14163_394
510/1	IGMI_ES_LB_/159
51.9*	- GMI_D3_LB_6061
64.5 —	— GMI_ES01_c20980_333
71.5	- GMI ES17 c3776 638




#### 05026-8.1



#### 05026-8.2





Figure A2. Molecular marker linkage map from 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) (red=EST markers, blue=GBS markers, green=DArT markers) (Continued).

# 05026-11



Figure A2. Molecular marker linkage map from 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) (red=EST markers, blue=GBS markers, green=DArT markers) (Continued).

37.0-

38.1 ~

GMI_ES14_C19908_282 GMI_ES14_Inc6920_447 GMI_ES05_C8958_463 GMI_ES05_C261_587

JGMI_ES_CC11933_314 GMI_ES02_c7694_423

GMI_ES17 c7973 279

2.3



Figure A2. Molecular marker linkage map from 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) (red=EST markers, blue=GBS markers, green=DArT markers) (Continued).

4.3- GMI ES01 c13068 54

GMI_GBS_112713 GMI_ES15_c5315_156 GMI_GBS_95417 GMI_GBS_73388

5.2~

## 05026-15

05026-16.2 0.0--

14.7-15.8

16.4 -

17.0-





GMI_ES_CC6557_563 GMI_GBS_66183 GMI_GBS_75418	0.01 GMI_ES15_67233_246 8.7 GMI_GBS_3090 15.91 GMI_ES15_610847_268 28.01 GMI_ES14_6429_194 36.4 GMI_ES_CC2716_392 37.01 GMI_ES_CC2716_392 GMI_ES2_C38960_34E_GMI_ES15_67879_555 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES15_67879_555 55.5 GMI_ES02_C38960_34E_GMI_ES02_6786_55 55.5 GMI_ES02_C38960_34E_GMI_ES02_6786_55 55.5 GMI_ES02_C38960_34E_GMI_ES02_6786_55 55.5 GMI_ES02_6786_55 55.5 GMI_ES02_6786_55
GMI_ES22_c2039_647 GMI_DS_LB_1346 GMI_ES05_c10407_153 GMI_ES02_c21558_350 GMI_ES11_c9638_786 GMI_ES01_c8445_691 GMI_GBS_101905 GMI_ES03_c12754_237 GMI_ES03_c12754_237 GMI_ES03_c11491_497 GMI_ES_LB_6699 GMI_ES03_irc22616_280 GMI_ES_LB_7601 GMI_ES01_c9384_567 GMI_ES_L7527	38.11 GM_ES03_C12159_493 38.71 GM_ES01_C21179_347 GMI_ES01_C3388_616 GMI_ES05_C14456 70 GMI_ES05_C422_582 GMI_ES03_C9203_225 GMI_ES15_C2738_176 GMI_DS_LB_7758 IGMI_ES17_C9934_427 GMI_ES01_C19797_80 GMI_ES15_C19218_469 GMI_ES02_C27935_359 IGMI_ES17_C780_489 GMI_ES02_C27935_359 IGMI_ES17_C780_489 GMI_ES22_Irc17882_62 IGMI_ES17_C780_489 GMI_ES02_C17882_62 IGMI_ES17_C780_2001_ES03_C9714_720 GMI_ES17_C650_702 GMI_ES03_C9714_720 GMI_ES03_C2344_498 GMI_ES15_C7254_710 IGMI_ES01_C6455_522 GMI_DS_LB_3341
GMI ES17 c3309 96 GMI ES02 lrc13826 351	46.7 IGM_ES02_c20364 439 GMT_ES_LB_11858 IGM_ES05_c12170_315 47.8 48.4 48.4 49.0 GM_GBS_9398 GMI_ES05_c3954_482 48.4 49.0 GMT_ES05_c23665_336 GMT_ES02_c8953_242 50.8 GMT_ES02_c1493_323 GMT_ES01_c18017_440 51.4 GMT_ES02_c1493_323 GMT_ES01_c18017_440 51.4 GMT_ES02_c1493_323 GMT_ES01_c18017_440 51.4 GMT_ES01_c13907_104 GMT_ES03_c1096_257 GMT_ES15_c7106_329 GMT_ES17_c15455_93 GMT_ES05_c8742_452 GMT_ES05_c2351_213 GMT_ES05_c8742_452 GMT_ES05_c2351_213 GMT_ES05_c8742_452 GMT_ES05_c2351_213 GMT_ES05_c9397_421 GMT_ES03_c16835_129 GMT_ES05_c9397_421 GMT_ES03_c16835_129 GMT_ES05_c9397_421 GMT_ES05_c4394_195 GMT_ES05_c13000_165 GMT_ES05_c15948_428
- <b>16.2</b> → GMI_ES05_c6521_452	GMI_ES15_c5368_259 GMI_ES05_c20848_84 GMI_ES02_c6920_272 GMI_ES02_c6920_143 GMI_ES17_c20215_324 05026-17 0.0 GMI_ES15_c8998_976
	1.7 ——— GMI_ES15_c8164_563
- GMI ES14 c14823 110	2.8 GMI_ES15_c7755_266 GMI_ES17_c2276_224 GMI_ES17_c15366_116 3.4 GMI_ES15_c5837_115 GMI_ES15_c17448_349
GMI_ES17_c19025_110 GMI_ES17_c2787_171 GMI_ES03_c1068_262 GMI_GBS_98449 GMI_ES02_c17401_928 GMI_ES17_c1612_641 GMI_ES01_c1881_299 GMI_ES05_c1299_884 GMI_ES22_c2548_559 GMI_ES14_c2532_571 - GMI_ES03_c10711_406	4.5 GMI_ES02_c28150_198 GMI_ES17_c19933_225 GMI_ES01_c12759_163 GMI_ES_LB_7322 GMI_ES15_c8008_218 GMI_ES17_c1242_703 GMI_ES01_c6200_493

Figure A2. Molecular marker linkage map from 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) (red=EST markers, blue=GBS markers, green=DArT markers) (Continued).

## 05026-18



## 05026-19

0.0	A	- GMI_ES02_c14478_154
1.1	П	MGMI_DS_LB_10834 GMI_ES14_c2285_377
	1 [	GMI ES02 c27120 208 GMI ES03 c9677 427
2.2		GMI_ES02_c14927_478 GMI_DS_0Pt-13151_665
	n	GMI ES02 c25424 168
5.9		YGMI ES15 Irc9062 227 GMI ES01 c19245 178
16.6 -		GMI_ES14_c118_502 GMI_ES05_c2066_503
10.0		/IGMI_ES22_c4463_275
20.8 -	4	GMI_ES22_c11596_73 GMI_ES15_c12694_287
20.0		/IGMI_GBS_3525
22 7-		GMI_ES17_c2454_883 GMI_ES01_c16071_418
23.7	Н	/IGMI_ES03_c10715_468
43.4 ₁ \	11	GMI_ES_CC11076_204 GMI_ES03_c1616_774
45.2	Г	GMI_GBS_25081
45.8-		GMI_ES17_c7110_570
46.4		GMI_GBS_80511 GMI_ES01_c27869_512
47.0		GMI_ES05_c4438_302 GMI_GBS_92025
47.6		GMI_GBS_10457 GMI_ES15_c8569_380
47.01		GMI_ES13_c14470_87 GMI_ES21_c6485_431
48.21		GMI_ES01_c2436_405
		GMI_GBS_100319 GMI_ES_LB_11958
- 0		GMI_ES05_c14901_270 GMI_ES16_c20195_287
1		GMI_ES02_c1833_408 GMI_DS_LB_9310
i i		GMI_DS_LB_6019 GMI_ES_LB_11974
48.8 1		GMI ES22 c18838 446 GMI ES02 c16381 584
- N	Н	/ GMI_ES03_c9122_309 GMI_DS_LB_950
l (		GMI_ES_LB_11633 GMI_GBS_96826
S S		GMI_ES_CC15240_111 GMI_ES17_c7039_546
	U	GMI ES15 c17597 267



Figure A2. Molecular marker linkage map from 'Souris' x 'ND030299' F₆ recombinant inbred oat population (Population 05026) (red=EST markers, blue=GBS markers, green=DArT markers) (Continued).