Evaluation of spring canola (*Brassica napus*) lines derived from Rutabaga (*Brassica napus* var. *napobrassica*) × canola crosses for agronomic and seed quality traits and heterosis

By

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A thesis submitted in partial fulfillment of the requirements for the degree of

Master of Science

in

Plant Science

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Abstract

Brassica napus L. (AACC, 2*n* = 38) canola is the most extensively cultivated *Brassica* oilseed crop in the world; it is one of the most important field crops in Canada. The narrow genetic diversity in this crop is considered one of the major hindrances for continued improvement of this crop for high seed yield and improved agronomic traits. The potential value of the Rutabaga (*B. napus* var. *napobrassica*) gene pool for broadening the genetic base of the Canadian spring *B. napus* canola and for use in the breeding of hybrid cultivars was investigated in this M.Sc. thesis research. For this, the agronomic and seed quality traits including seed yield of a set of F_2- and BC_1-derived inbred lines, developed from two Rutabaga × spring *B. napus* canola crosses, and their test hybrids were evaluated in replicated field trials. The inbred lines were also analyzed by use of SSR markers to estimate the extent of allelic diversity introgressed from Rutabaga into the inbred lines. SSR marker analysis showed that genetically distinct spring *B. napus* canola lines carrying the unique alleles of the A and C genomes of Rutabaga can be obtained from both F_2- and BC_1-derived populations. Some of the inbred lines gave higher seed yield than the spring canola parent and also displayed high heterosis in the test hybrids, which apparently resulted from favorable combinations of Rutabaga and spring *B. napus* canola alleles. Thus, the results from this study demonstrated the potential value of the Rutabaga gene pool for use in the breeding of hybrid spring *B. napus* canola cultivars.

Acknowledgments

It is a great pleasure to acknowledge the people for their support during my M.Sc. program at the University of Alberta, those who made this thesis possible.

I would first like to express my sincere gratitude to my supervisor Dr. Habibur Rahman for giving me the opportunity of being a member of the Canola Breeding Program at the University of Alberta, and for the enthusiasm, encouragement, motivation and advice that he has provided throughout my program. I extremely appreciate his continued support, guidance and contributions during my research and writing of this thesis.

I am also deeply grateful to Dr. Rong-Cai Yang, member of my supervisory committee, for his valuable suggestions and technical supports throughout my research project, and Dr. Barb Thomas, my external examiner, for her insightful comments on the text of my thesis.

I give my sincere thanks to the Canola Breeding Group and the AFNS research support team members: Dr. Berisso Kebede, Jose Salvador Lopez Benites, An Vo, Dr. Neil Hobson, Dr. Rudolph Fredua-Agyeman, Dr. Mehdi Farid, Jory Underwood and Rubeena Shaikh, Dr. Urmila Basu, Nikki-Karyssa Scott, Kelley Dunfield, Victor Manolii, Sarah Jespersen and Robin Miles, as well as the Canola Breeding Group past and present students: Rohit Attri, Rameez Iftikhar, Xin Wang, Bhavik Jani, Azam Nikzad, Gholamreza Habibi, Jakir Hasan and Huiyan Zhao, and all my friends and Staff at the department of AFNS for their support, advice and assistance in the course of my research and writing of my thesis.

For financial support, I thank the Natural Sciences and Engineering Research Council of Canada (NSERC), Crop Production Services (CPS), Alberta Canola Producers Commission,

Alberta Crop Industry Development Fund (ACIDF), and Agriculture and Agri-Food Canada (AAFC).

Last but not least, I want to acknowledge my family. Words cannot express how grateful I am to my wife and beloved daughters for all their support and encouragement throughout this experience.

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List of Symbols and Abbreviations

±	Plus/minus
×	Cross
⊗	Self-pollination
n	Number of observations
ng/µl	Nano gram per micro liter
µl	Microliter
µM	Micromole
µmol/g	Micromoles per gram
°C	Degrees Celsius
BC_1	First backcross generation
BC_1Fn	nth backcross generation
cm	Centimeter
cv.	Cultivar
df	Degree of freedom
dNTP	Deoxynucleotide triphosphate
F_1	First filial generation
F_{st}	F-statistics
g	Gram
ha	Hectare
m	Meter
M	Mole
Mbp	Million base pair

mg	Milligram
min	Minute
ml	Milliliter
mm	Millimeter
mM	Millimole
MMT	Million metric ton
Kg/ha	Kilogram per hectare
p	Probability
r	Coefficient of correlation
R^2	Coefficient of determination
AAFC	Agriculture and Agri-Food Canada
AFLP	Amplified fragment length polymorphism
AMOVA	Analysis of molecular variance
ANOVA	Analysis of variance
CMS	Cytoplasmic male sterility
CPH	Check parent heterosis
CP	Common parent
DH	Doubled haploid
DNA	Deoxyribose nucleic acid
DTF	Days to flowering
DTM	Days to maturity
FAO	Food and Agriculture Organization of the United Nations
GCA	General Combining Ability
HEAR	High erucic acid rapeseed

HOLL	Canola oil containing high oleic acid and low linolenic acid
IN	Inbred line
miRNA	Micro RNA
MPH	Mid-parent heterosis
MS	Mean squares
NIRS	Near infrared spectroscopy
PCR	Polymerase chain reactions
QTL	Quantitative trait loci
RAPD	Random amplified polymorphism DNA
RFLP	Restriction fragment length polymorphism
RNA	Ribonucleic acid
SCA	Specific combining ability
SD	Standard deviation
SE	Standard error
SNP	Single-nucleotide polymorphism
sRNA	Small RNA
SS	Sum of squares
SSR	Simple sequence repeat
Taq DNA polymerase	*Thermus aquaticus* DNA polymerase
TH	Test hybrid
UPGMA	Unweighted pair-group method with arithmetic mean
USDA	United States Department of Agriculture
var.	Variety

Chapter 1

Literature Review

1.1 Introduction

Brassica napus L. belongs to the genus *Brassica* of the mustard family, Brassicaceae. Brassicaceae is one of the most morphologically-diverse plant families comprising more than 300 genera and 3000 species distributed mainly in the temperate areas (Lysak et al., 2009; Al-Shehbaz, 2011; Daun, 2011). The genus *Brassica* includes about 37 species; wide diversity exists within several species of this genus (Dixon, 2007; Cartea et al., 2011). *Brassica* is also featured as one of the most economically-important genus; this includes species producing edible oil, condiment and vegetables for human consumption, fodder and forages for animal feed, and oil for non-food products, such as polymers and surface coatings (for review, see Scarth and Tang, 2006).

Archeological evidence from Xian, China suggests that cultivation of *Brassica* crops dates as far back as 7000 years ago in Neolithic times (for review, see Prakash and Hinata, 1980; Daun, 2011). *Brassica* species were primarily utilized as potherb, vegetable and spice condiment. Utilization of *Brassica* plants as oilseed crop took place far later than their primary culinary usage (for review, see Prakash and Hinata, 1980; Daun, 2011). Earliest evidence of oil extraction from *Brassica* can be traced back to 2000 BC by ancient civilizations in India (for review, see Colton and Potter, 1999; Daun, 2011). Cultivation of *Brassica* crops for extraction of oil in Europe dates back to 13th century, mainly for use of this oil as lamp oil (for review, see Colton and Potter, 1999; for review, see Raymer, 2002). The cultivation of *Brassica* oilseed crop in Canada dates back to the 1930's. *Brassica rapa* L., named as Polish rapeseed as the first seed of this crop was introduced from Poland, was planted in Saskatchewan in the 1930's. With the emergence of the Second World

War, *B. napus*, called as Argentine rapeseed as the seed of this crop was first introduced from Argentina, was also included in the Canadian crop acreage in 1942. Oil of both Polish and Argentine rapeseed was used as lubricant for the steam engines at that time (for review, see Colton and Potter, 1999; Daun, 2011; Eskin, 2013).

Brassica seed oil has been used as edible oil for thousands of years in Asia (Daun, 2011). It was consumed as food oil in Europe in the 20th century during the Second World War (for review, see Colton and Potter, 1999). In Canada, the use of rapeseed oil for edible purposes started in the 1950's (Stefansson and Downey, 1995). However, the presence of a high content of erucic fatty acid in oil and glucosinolate in seed meal imposed limitations for the use of this seed oil and meal for human consumption and as a protein supplement in animal feed, respectively (Daun, 2011; Przybylski and Eskin, 2011). Plants of *B. napus* and *B. rapa*, which seed oil is free from erucic acid were identified in the 1960's (Stefansson, 1960; Downey, 1964, for review, see Stefansson and Downey, 1995) and *B. napus* plants with a low content of glucosinolate in seed meal was identified in 1967 (Kzrymanski, 1967, for review, see Stefansson and Downey, 1995) by Canadian researchers. Use of these genetic variations in breeding resulted the first *B. napus* cultivar Tower, which seed oil contained less than 2% erucic acid and seed meal contained less than 30 μmol glucosinolate per gram, was released in Canada in 1974 (Stefansson and Kondra, 1975; Daun, 2011; Przybylski and Eskin, 2011). This improved *Brassica* oilseed with low erucic acid in oil and low glucosinolate in seed meal was branded as 'canola' (Stefansson and Downey, 1995). With these improvements, the *Brassica* oilseed crops or canola became one of the important oilseed crops in the world. Currently, canola is the second most important oilseed crop in the world after soybean, and is the main oilseed crop in Canada, Western Europe and China, and also a major oilseed crop in India, Australia and Eastern Europe (FAOSTAT, 2016a; USDA, 2016; McVetty et

al., 2016).

1.2 Origin and Evolution of *Brassica* Species

1.2.1 Genome relationship between *Brassica* species

The genus *Brassica* includes six species of great economic importance. Three of these are diploid, namely *Brassica rapa* L. (genome AA, 2*n* = 20), *Brassica nigra* (L.) Koch (black mustard) (genome BB, 2*n* = 16) and *Brassica oleracea* L. (genome CC, 2*n* = 18), and three are amphidiploid, namely *Brassica juncea* (L.) Czern & Coss (Indian or brown mustard) (genome AABB, 2*n* = 36), *Brassica napus* L. (genome AACC, 2*n* = 38) and *Brassica carinata* A. Braun (Abyssinian or Ethiopian mustard) (genome BBCC, 2*n* = 34) (Prakash and Hinata, 1980; Rakow, 2004; Dixon, 2007; Daun, 2011). The genome relationship between these six *Brassica* species was first portrayed by U (1935) in the form of a triangle (U, 1935, for review, see Hayward, 2012) which is commonly known as the "triangle of U" or U-triangle (Figure 1.1). Based on homoeology

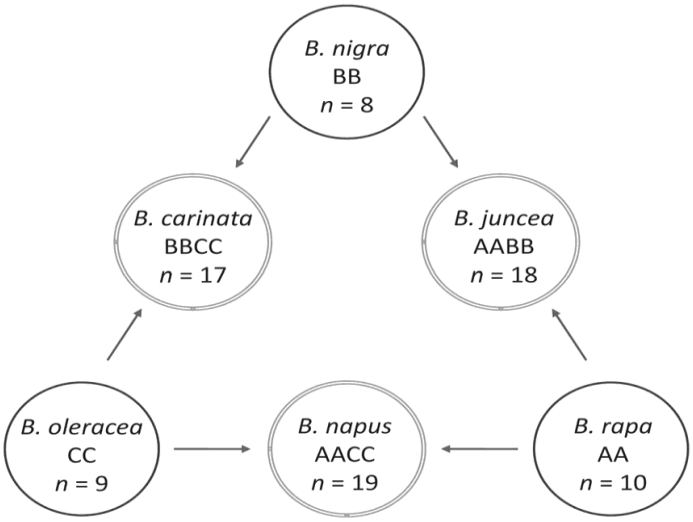

Figure 1.1: The triangle of U. Genome relationships between six species of the genus *Brassica* (U, 1935, cited by Hayward, 2012).

of the chromosome of different species and artificial synthesis of the amphidiploids from the diploids, and analysis of nuclear DNA and genome specific chromosome markers, the cytogenetic

relationships between the *Brassica* species, which initially proposed by U (1935), has been confirmed by different researchers (Attia and Röbbelen, 1986; Attia et al., 1987; Busso et al., 1987; Song et al., 1993; Bohuon et al., 1996; Suwabe et al., 2008).

1.2.2 Evolution of *Brassica napus*

Brassica napus (genome AACC, 2*n* = 38) is the most ancient amphidiploid in the genus *Brassica*. It is the product of interspecific hybridization of the two diploid species *B. rapa* (2*n* = 20) and *B. oleracea* (2*n* = 18) (Prakash and Hinata, 1980; Rakow, 2004). It is believed that *B. napus* evolved in the Mediterranean region where its two parental species occur in nature (Dixon, 2007). Naturalized forms of *B. napus* have also been reported in New Zealand suggesting that *B. napus* may have evolved from various forms of *B. oleracea* and *B. rapa* in more than one geographical region (Rakow, 2004).

1.2.3 Relationship between *Brassica* and *Arabidopsis thaliana*

The relationship between *Arabidopsis thaliana* and the genus *Brassica* has been well documented at the genome level (Cavell et al., 1998; King, 2007). A genome collinearity study by Cavell et al. (1998) demonstrated that the protein coding DNA regions of *Arabidopsis* and *Brassica* show about 87% similarity. Comparative genomics studies have also indicated that the three *Brassica* genomes evolved from a common progenitor which is similar to *Arabidopsis* (King, 2007). It is also hypothesized that the genomes of the diploid *Brassica* species originated from a hexaploid ancestor which has a close relationship to *Arabidopsis* (Parkin et al., 2005). The size of the genomes of the *Brassica* species varies from 470 Mbp in *B. nigra* to 1540 Mbp in *B. carinata*; *B. napus* with 1130–1240 Mbp has the smallest size genome among the *Brassica* amphidiploid species (King, 2007).

1.3 Cultivation and growth habits of *Brassica napus*

Brassica napus is the most extensively cultivated crop species of the genus *Brassica* in the world (for review, see Raymer, 2002; Canola Council of Canada, 2016a). It is also the most productive oilseed crop species of this genus. According to Rakow (2004), the high productivity of *B. napus* is associated with the high number of chloroplast per unit area of leaf, rendering a high photosynthetic rate in this species. Based on vernalisation requirement, *B. napus* can be categorized into three growth habit types: winter, semi-winter and spring (Wang et al., 2011). The winter type requires about 6-8 weeks of vernalisation at rosette leaf stage while the spring type does not require vernalisation to proceed to flowering. The length of the duration of vernalisation to promote flowering in the semi-winter type is less than that of the winter type (Wang et al., 2011). The winter type is mainly grown in Europe, spring type is predominantly grown in Canada and Australia, and the semi-winter type is grown in China (Rakow, 2012). Seed yield of the winter type is almost double when compared to the spring type (Rakow, 2004). These three types are known to be genetically distinct (Diers and Osborn, 1994; Bus et al., 2011); therefore, one of these types can be used in the breeding of the other types (for review, see Rahman, 2013). Another form of *B. napus* is a root-forming type, and is cultivated for vegetable and fodder usages (Rakow, 2004).

1.4 Seed quality of *Brassica napus*

1.4.1 Erucic acid content and its genetic basis

Seeds of *Brassica* oilseed crops contain more than 40% oil (for review, see Raymer, 2002). Several quantitative trait loci (QTL) from the linkage groups of both A and C genome contribute to the total seed oil content in *B. napus* (for review, see Rahman et al., 2013). Unlike many other

vegetable oils, which are composed mostly of C16 and C18 fatty acids, the seed oil of traditional *Brassica* crops contains long chain fatty acids with more than 18 carbons, such as erucic acid (C22:1) (Kondra and Stefansson, 1965; Downey, 1983). In traditional rapeseed, erucic acid content accounted more than 45% of the total fatty acids of the seed oil (Downey, 1983; Ackman, 1990). Erucic acid is undesirable in edible oil; however, rapeseed oil rich in erucic acid is used to produce a diverse range of industrial products such as lubricants, detergents, pharmaceuticals, hydraulic fluids and biodegradable plastics (for review, see Scarth and Tang, 2006). A growing demand for erucic acid-rich oil for industrial applications has triggered the breeding efforts for the improvement of rapeseed cultivars with 50-60% erucic acid in oil content (high erucic acid rapeseed or HEAR, also referred to as industrial rapeseed) (for review, see McVetty and Scarth, 2002). The genetic basis of erucic acid content in seed oil of *B. napus* has been investigated by several researchers. The *Bn.FAE1.1* and *Bn.FAE1.2* genes located on the A and C genome chromosomes A8 and C3, respectively, with additive effect are known to play the major role in the accumulation of erucic fatty acid in *B. napus* rapeseed oil (Harvey and Downey, 1964; Fourmann et al., 1998; Rahman et al., 2008; Cao et al., 2010; Karim et al., 2016).

Due to health concerns arising from erucic acid content in edible oil (Daun, 2011; Przybylski and Eskin, 2011), intensive breeding efforts initiated in Canada to eliminate this fatty acid from *Brassica* seed oil was undertaken. A zero-erucic acid (trace content of erucic acid) line, selected from the German spring rapeseed (*B. napus*) cultivar Liho, was reported in 1961 (Stefansson et al., 1961). By use of this genetic variation, the first low-erucic acid (about 2% erucic acid in oil) *B. napus* and *B. rapa* cultivars, Oro and Span, respectively, were developed. Further breeding almost eliminated this fatty acid (less than 0.5%) from *Brassica* oilseed crops (Stefansson and Downey, 1995; Przybylski and Eskin, 2011). The accumulation of the mutant alleles of the *FAE1*

genes, in fact, is known to be the genetic basis of decreased content of erucic acid in the seed oil of *Brassica* crops (Cao et al., 2010; Karim et al., 2016).

1.4.2 Glucosinolate content in seed meal

Glucosinolates are secondary plant metabolites (phytochemicals), derived from amino acids, and commonly occur in the species of the family Brassicaceae. Based on the precursor amino acid, glucosinolates are classified into three groups: aliphatic (alkyl or straight-chained), aromatic (ring-shaped), and indolyl (indole) glucosinolates. Aliphatic, aromatic and indolyl glucosinolates are biosynthesized from methionine, phenylalanine, and tryptophan, respectively (for review, see Halkier and Du, 1997; Dixon, 2007; Velasco et al., 2008). More than 100 types of glucosinolates are identified in *Brassica* (for review, see Fahey et al., 2001; Mithen and Parker, 2004) of which seven aliphatic, four indole, and one aromatic glucosinolates are detected in *B. napus*. Of the total glucosinolate content of *B. napus* seeds, aliphatic glucosinolates, gluconapin, glucobrassicanapin and progoitrin accounts more than 80% (Kondra and Stefansson, 1970; Velasco et al., 2008), while indolyl glucosinolates accounts about 15% (Newkirk, 2011). Howell et al. (2003) mapped three major and a minor QTL on three C genome linkage groups of C2, C7 and C9, and Rahman et al. (2015) detected similar numbers of QTL on the A genome linkage groups A2, A7 and A9 involved in the control of total glucosinolate content in seed.

Seeds of traditional *Brassica* oilseed cultivars contained a high content (>60 µmol/g seed) of glucosinolates (Newkirk, 2011). Enzymatic hydrolysis of glucosinolates in the presence of the enzyme myrosinase results in several anti-nutritional compounds, such as thiocyanates and isothiocynates. Their presence in seed meal limits utilization of this protein-rich meal in animal feed, especially for monogastric animals (for review, see Halkier and Du, 1997; Daun, 2011;

Przybylski and Eskin, 2011; Rakow, 2012). Canadian researchers found that the Polish *B. napus* forage rape cultivar 'Bronowski' contained a low content (< 30 µmol/g seed meal) of glucosinolates in seeds; this genetic variation has been used exclusively for the development of low glucosinolate *Brassica* oilseed cultivars (Downey, 1983; Stefansson and Downey, 1995). The reduction of glucosinolate content in seed meal has been one of the major achievements in the breeding of *B. napus* for the improvement of its seed meal quality. Today, seed meal of all canola cultivars contain less than 30 µmol glucosinolate per gram of oil free meal.

1.4.3 Canola quality *Brassica* oilseed

The world's first low-erucic, low-glucosinolate, also called 'double low', *B. napus* cultivar Tower and *B. rapa* cultivar Candle were released in Canada in 1974 and 1978, respectively (Stefansson and Downey, 1995; Przybylski and Eskin, 2011). This type of cultivar met the quality standard for oil for human consumption and for seed meal for use in animal feed and was branded by the Canola Council of Canada as 'canola'. The word canola, stands for "Canadian Oil Low Acid"; this is officially defined as the rapeseed with less than 2% erucic acid (22:1) in its oil and less than 30 µmol of any one or any mixture of 3-butenyl glucosinolate (gluconapin), 4-pentenyl glucosinolate (glucobrassicanapin), 2-hydroxy-3 butenyl glucosinolate (progoitrin), and 2-hydroxy-4-pentenyl glucosinolate (napoleiferin) per gram of air-dried, oil-free solid meal (Przybylski and Eskin, 2011; Canola Council of Canada, 2016a; Agnihotri, 2015).

Among the different *Brassica* oilseed crops in the world, *B. napus* canola is the most important one in regards to acreage and production; the other being *B. juncea* and *B. rapa* (Hayward, 2012). Canola seed contains about 44% oil and the seed meal contain about 40% protein (for review, see Raymer, 2002; Canola Council of Canada, 2016a). Currently, canola oil is extensively used for

human consumption and its meal is used as a source of protein in animal feed (Eskin, 2013; Newkirk, 2011).

Canola oil is one of the healthiest oils for human consumption (Canola Council of Canada, 2016c). This oil contains 5-8% saturated, 60-65% monounsaturated and 30-35% polyunsaturated fatty acids (for review, see Raymer, 2002). It has the lowest level of saturated fatty acids among all leading edible vegetable oils (for review, see Mailer, 2009; Canola Council of Canada, 2016c). Saturated fats are believed to increase the risk of coronary heart disease, and therefore, undesirable for human consumption (Hanke et al., 2013; Mathaüs, 2013). The polyunsaturated fat portion of the canola oil is a remarkable source of two essential fatty acids: linoleic acid, an omega-6 fatty acid, and alpha-linolenic acid, an omega-3 fatty acid. These fatty acids are important for body growth and development and are also known to reduce the risk of heart diseases (Canola Council of Canada, 2016b; European Food Information Council, 2016). Linoleic and linolenic acid contents in canola oil are 20% and 10%, respectively (Eskin, 2013; Hanke et al., 2013). Canola oil contains more than 60% oleic acid (for review, see Mailer, 2009; Eskin, 2013). Unlike many other fatty acids, this fatty acid does not increase the level of cholesterol in blood (Canola Council of Canada, 2016c). High oleic low linolenic acid (HOLL) canola is a more recent form of canola quality *B. napus*. This type of oil contains more than 70% oleic acid and about 3% linolenic acid (for review, see Debonte et al., 2012). Compared to conventional canola, HOLL canola contains a decreased content of polyunsaturated fatty acid, mostly alpha-linolenic acid, and an increased content of oleic acid. This makes HOLL canola more resistant to oxidation and is suitable for applications where cooking at high temperatures is needed (for review, see Mailer, 2009; Hanke et al., 2013).

1.5 *Brassica* oilseed production in the world

Based on the harvested area and production of oilseed crops in the world, *Brassica* oilseed crops are ranked second after soybean (FAOSTAT, 2016a; McVetty et al., 2016; USDA, 2016). In 2013-2014 crop year, soybean and *Brassica* oilseed crops were harvested from 117.7 and 35.8 million hectares, respectively. Of the total global production of 540.2 million metric tons (MMT) of oilseeds, soybean and *Brassica* oilseeds had a share of 57.1% and 13.1% (308.4 and 71.0 MMT), respectively; cottonseed, peanut and sunflower had a share of 8.6, 7.7 and 7.6% (46.7, 41.4 and 41.3 MMT), respectively (Table 1.1).

Table 1.1: Global harvested area and production of important oilseeds in 2014.

Oilseed crops	Harvested area (million ha)	Production (million metric tons)
Olives	10.3	15.5
Rapeseed (*Brassica* oilseeds)	35.8	71.0
Soybean	117.7	308.4
Sunflower seed	24.8	41.3
Cotton seed	-	46.7
Peanut	-	41.4
Palm kernel	-	15.9
Total		540.2

(Adapted from FAOSTAT, 2016a and USDA, 2016).

Brassica oilseed crops are mainly grown in Asia, Europe, North America and Australia. In 2014, this crop was harvested from 14.8, 9.1, 8.7 and 2.7 million ha of these regions, respectively. In terms of production, Europe, however, is the leading continent in the world with a production of 28.9 MMT, followed by Asia, North America and Australia with a production of 20.8, 16.7 and 3.8 MMT, respectively (Figure 1.2) (FAOSTAT, 2016a).

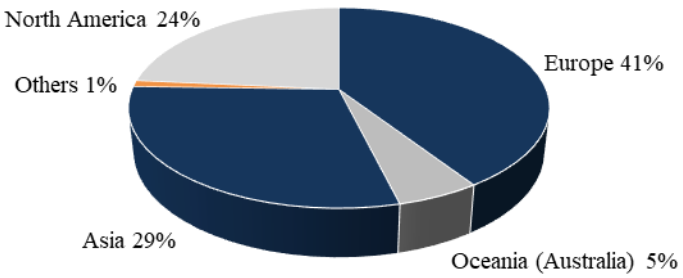

Figure 1.2: Production of *Brassica* oilseed crops on different continents of the world in 2014 (FAOSTAT, 2016a).

Canada is the largest *Brassica* oilseed producing country in the world, followed by China and India (McVetty et al., 2016). In Europe, Germany and France are the major producers; Russia and Poland also grow this oilseed crop at a remarkable scale (Table 1.2).

Table 1.2: The major countries producing *Brassica* oilseed crops in the world in 2014.

Countries	Harvested area (million ha)	Production (million metric tons)
Canada	8.1	16.4
China	6.6	11.6
India	7.2	7.9
France	1.5	5.5
Germany	1.4	6.2
Russian Confederation	1.1	1.5
Poland	1	3.3
Australia	2.7	3.8

(Adapted from FAOSTAT, 2016a and USDA, 2016).

1.6 Production of *Brassica* oilseed crops in Canada

In Canada, canola is the second largest field crop, only behind wheat, with an annual harvested area and production of 9.9 million ha and 25.6 MMT, respectively (FAOSTAT, 2016b; Statistic Canada, 2016d) (Figures 1.3 and 1.4).

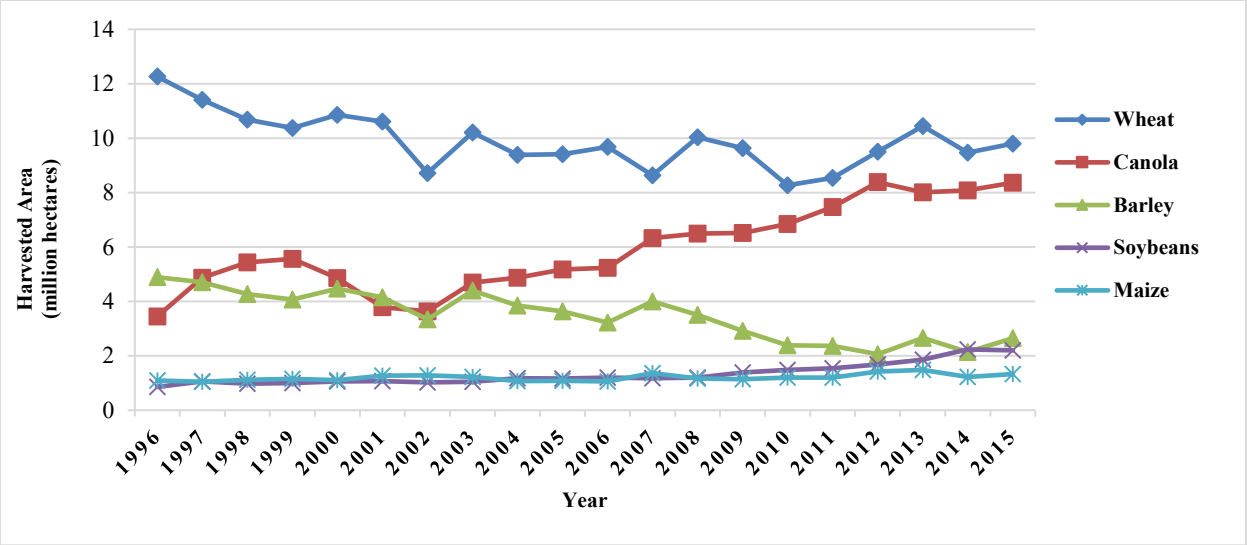

Figure 1.3: Harvested area of the leading crops in Canada in the last two decades (FAOSTAT, 2016b; Statistic Canada, 2016a).

Among the different oilseed crops in Canada, canola is the most important one; the other oilseed crops grown in this country are soybean and flaxseed (Statistic Canada, 2016a). Soybean is grown mainly in Quebec, Ontario and Manitoba, while canola and flaxseed are mostly grown on the Prairie provinces Alberta, Saskatchewan and Manitoba. In 2014, of the total oilseed

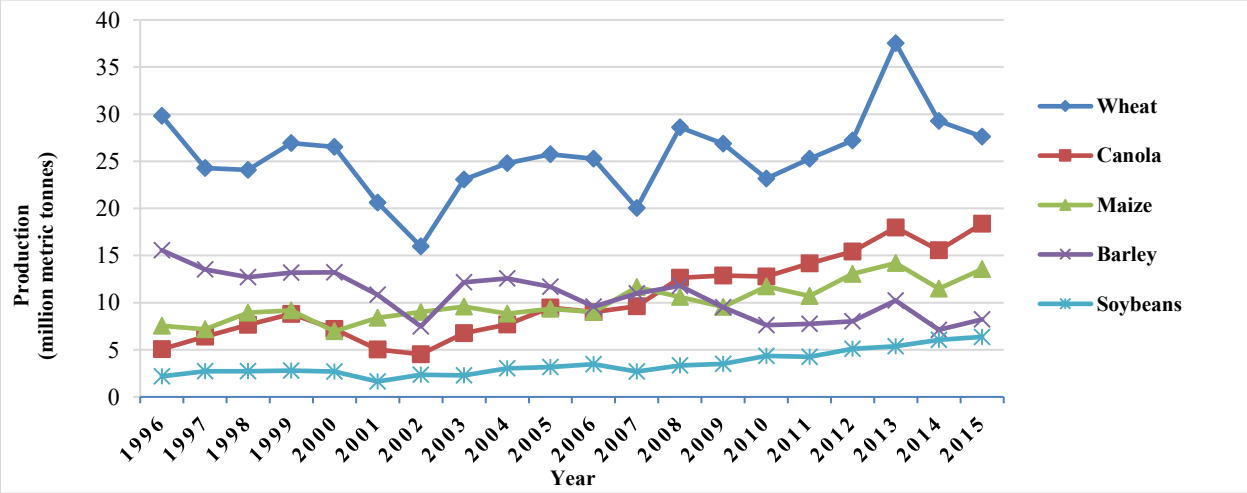

Figure 1.4: Production of the leading crops in Canada in the last two decades (FAOSTAT, 2016b; Statistic Canada, 2016d).

production of 23.6 MMT, the contribution of canola, soybean and flaxseed was 16.4, 6.1 and 0.9

MMT, respectively. Canola alone constitutes about 70% of the total oilseed production in Canada (Statistic Canada, 2016b).

Canadian production of canola has been overwhelmingly escalating over the past four decades, and in 2008, it exceeded 10 MMT (Casséus, 2009; FAOSTAT, 2016a). In 2015, 8.4 million hectares of canola was harvested, producing about 18.4 MMT of seeds. Compared to 2014, an increase of 12% in canola production occurred in 2015 (Statistics Canada, 2016a). The provinces Saskatchewan, Alberta and Manitoba produces more than 95% of the total Canadian canola (Casséus, 2009; Rakow, 2012).

1.7 Importance of canola in the Canadian economy

Canola contributes about $19 billion to the Canada's economy annually and generates about 25% of the total cash receipts for the Canadian farmers. Canola is also considered as a significant value-added industry in Canada, where the processing facilities produce 3 MMT of canola oil for human consumption, and 4 MMT of canola meal for animal feed each year (Canola council of Canada, 2016d).

Canada is one of the major canola exporting countries in the world; about 90% of the total canola produced in this country is being exported as seed or as oil and meal to 55 destinations in the world (Canola council of Canada, 2016d). In 2012, about 60% of the Canadian canola was exported as seed to China, Japan, Mexico and the United States of America (USA) (Canola Council of Canada, 2016d; Statistics Canada, 2016b; Statistics Canada, 2016c). Of the total canola oil extracted in Canada, about 84% is being exported (Casséus, 2009); the USA is the major importer of the Canadian canola oil (Statistics Canada, 2016c). Similarly, the USA is the major user of Canadian canola meal; in 2012, about 85% of the total canola seed meal was exported to the USA

(Statistics Canada, 2016c).

1.8 Genetic diversity in *Brassica napus*

The evolution of *B. napus* from a limited number of genetic variants of its two parental species, and intensive breeding conducted over the last decades within the restricted gene pool has narrowed down the genetic base of this crop (King, 2007; Bonnema, 2012; for review, see Rahman, 2013). Broadening the genetic base of the *B. napus* breeding materials is, therefore, considered a priority for the improvement of this crop (for review, see Rahman, 2013). Currently, most of the Canadian canola cultivars are hybrids; an increase in the level of heterosis in this type of cultivars is needed which can be achieved through broadening the genetic base of this crop (Rahman et al., 2016).

1.8.1 Broadening of genetic diversity in *Brassica napus* using primary gene pool

The genetic base of the breeding materials can be diversified by the use of genetically distinct germplasm of the same species (for review, see Rahman, 2013). For instance, the European winter *B. napus* has been utilized to broaden the genetic base of the spring *B. napus* canola (Kebede et al., 2010) for the development of open-pollinated cultivars (Rahman, 2017). The potential value of this gene pool for use in the breeding of spring *B. napus* hybrid cultivars has also been demonstrated by several researchers. Quijada et al. (2004) developed a genetically diverse spring *B. napus* population through introgression of allelic diversity from French winter type *B. napus* and found that 30% of the test hybrids developed by the use of this population surpassed the seed yield of the commercial canola hybrids. Through a QTL mapping approach, Quijada et al. (2006) identified six genomic regions where the alleles from winter type contributed to seed yield in the hybrids. The usefulness of the European winter *B. napus* canola for the improvement of seed yield

in spring hybrid has also been demonstrated by Rahman et al. (2016). Similarly, the Chinese semi-winter *B. napus* has been used to broaden the genetic base of the German and Canadian spring *B. napus* canola (Qian et al., 2007), and this germplasm was also found promising for the improvement of the performance of the European winter *B. napus* hybrid cultivars (Qian et al., 2009).

1.8.2 Broadening of genetic diversity in *Brassica napus* using secondary gene pool

Introgression of the genome contents of the allied species can broaden the genetic base of the A and C genomes of *B. napus*, and this may increase the level of heterosis in hybrid cultivars (Zou et al., 2010; for review, see Rahman, 2013). The resynthesized *B. napus*, produced from the two progenitor species *B. rapa* and *B. oleracea*, can be used to broaden the genetic base of the *B. napus* gene pool (Lu et al., 2001; Seyis et al., 2003; Rahman, 2005; Girke et al., 2012a). High homoeology between the A and C genome chromosomes of *B. rapa* and *B. oleracea* allows the chromosomes of these two genomes to pair in resynthesized *B. napus*; this can result in homoeologous recombination (Parkin et al., 1995) and can further enhance allelic variation in *B. napus*.

Interspecific crossings of *B. napus* with *B. rapa*, *B. oleracea*, *B. carinata* and *B. juncea*, can also be deployed as a strategy to develop genetically distinct *B. napus* lines through substitution of the genome content of *B. napus* with the A and C subgenomes of its allied species (Bing et al., 1996; Li et al., 2006; Qian et al., 2006; Rahman et al., 2011; Bennett et al., 2012). Qian et al. (2005) reported that hybrids of natural *B. napus* and *B. napus* lines carrying the genome content of *B. rapa* (A_rA_r) exhibit heterosis for seed yield. Likewise, Li et al. (2013) broadened the genetic base of *B. napus* through introgression of the genome contents of *B. rapa* and *B. oleracea*. For this, they crossed *B. rapa* (AA) with a hexaploid (AACCCC), generated from crossing of *B. napus*

(AACC) and *B. oleracea* (CC), and by the use of genome-specific simple sequence repeat (SSR) markers, they found that the interspecific (AA × AACCCC) cross-derived *B. napus* lines were genetically distinct from the cultivated *B. napus*, especially for the A genome. Genetically distinct *B. napus* lines have also been developed through interspecific crossing of spring *B. napus* to *B. rapa* var. yellow sarson and Canadian *B. rapa* canola (Attri, 2015). Bennett et al. (2012) and Rahman et al. (2015) demonstrated that allelic diversity in the C genome of spring *B. napus* canola can be increased through *B. napus* × *B. oleracea* interspecific cross. Indeed, the C genome alleles of *B. oleracea* var. *botrytis*, var. *alboglabra*, var. *italica* and var. *capitata* have been introgressed into spring *B. napus* canola through this approach (Iftikhar, 2015; Wang, 2016). Mid-parent heterosis in test hybrids developed by the use of the lines carrying the genome content of *B. oleracea* found to be two times greater than the level of mid-parent heterosis detected in test hybrids developed by the use of the lines derived from spring × spring or winter × spring *B. napus* crosses (Rahman et al., 2016). The amphidiploid species *B. carinata*, carrying the C genome, can also be utilized to diversify the genetic base of the C genome of *B. napus* (Navabi et al., 2011).

B. napus alleles have also been used for the improvement of other *Brassica* species. For example, the low glucosinolate trait of the *B. napus* cultivar Bronowski was transferred to *B. rapa* for the development of canola quality *B. rapa* cultivars (Scarth et al., 1992). Furthermore, the divergence of the *Brassica* diploid and allotetraploid species has been captured in artificial allohexaploid *Brassica* carrying all three genomes (AABBCC) (Rahman, 2001; Mason et al., 2014). Such allohexaploid can be utilized as a bridge for introgression of desirable alleles from one species to other.

1.9 Heterosis

The phenomenon heterosis or hybrid vigor was first proposed by Shull (1914); this is defined as superior performance of the F_1 hybrids over the parents. Exploitation of heterosis in plant breeding started more than 75 years ago (Goldman, 1998; Duvick, 1999); the most obvious example of the exploitation of this phenomenon in crop production is maize, where the hybrid cultivars show significant superiority over the traditional open-pollinated cultivars (Cantrell, 1998; Goldman, 1998). The first maize hybrid cultivars released in the USA in the 1920's surpassed the seed yield of the open-pollinated cultivars by about 15% (Iowa State Dept. of Agric., 1934, reviewed in Duvick, 1999). According to Russel (1991) and Duvick (1992), about 50 to 60% increase in maize grain yield achieved since the 1930's was due to genetic improvement of this crop (Duvick, 1999).

Heterosis can occur for any trait including seed yield (Duvick, 1999; for review, see Schnable and Springer, 2013; Ryder et al., 2014); this can be positive or negative, or in other words, increasing or decreasing (Ryder et al., 2014). The expression of this phenomenon can be changed at different growth stages and can also be affected by the environment (Lefort-Buson and Dattee, 1982; Groszmann et al., 2014).

Although hybrid breeding is well-established in many crops including canola, the genetic basis of heterosis is still not well-understood (Crow, 1999; for review, see Baranwal et al., 2012; Ryder et al., 2014; Li et al., 2015). This phenomenon is considered to be controlled by multiple gene loci where the immediate progeny derived from a cross between two genetically distinct parents exhibits this phenomenon (for review, see Schnable and Springer, 2013). Dominance, overdominance and epistatic action of genes are the most common hypotheses that are widely used to elucidate the genetic basis of heterosis (Crow, 1999; Goodnight, 1999; Ryder et al., 2014). In

B. napus, for instance, QTL exhibiting partial or complete dominance, overdominance and epistatic interaction have been reported to be involved in the genetic control of heterosis for different traits; however, the QTL mostly exhibiting complete dominance and overdominance effects are involved in heterosis for seed yield (Radoev et al., 2008).

Hybrid performance has been recently related to the difference in gene expression between the hybrid and its parental lines. For instance, Chen et al. (2008) found that differential expression of the genes, specifically of the QTL regions involved in seed yield, plays a significant role in seed yield heterosis in *B. napus*. Studies have also shown that the epigenetic regulation of the expression of the genes is associated with heterosis (Hauben et al., 2009; Ryder et al., 2014; Sun et al., 2015). Among the different epigenetic mechanisms, DNA methylation, histone modification and the interference of small RNA (sRNA) molecules were studied to understand the molecular mechanism of heterosis in F_1 hybrids (for review, see Groszmann et al., 2013; Ryder et al., 2014). Study on the patterns of cytosine methylation in maize showed that the level of methylation in hybrids was lower than the average level of methylation in its parental lines. Sun et al. (2015) reported that an increase in the level of DNA demethylation and decrease in methylation can increase the level of expression of the genes in maize hybrids and thus plays a role in heterosis. Study on the mode of action of transcriptomes and epigenomes in hybrids of heterotic parents also showed that gene expression is associated with the modification of histone proteins. He et al. (2010) found that the differential activity of alleles between the hybrids and parental lines of rice (*Oryza sativa*) is correlated with the histone-mediated epigenetic modifications of the transcribed regions of the genes involved in heterosis. Epigenetic adjustment of gene expression mediated by small RNAs can also contribute to heterosis. Study of the role of miRNA-dependent gene regulation in maize suggested that heterosis for embryo germination vigor (Ding et al., 2012) and

elongation of internodes below the ear and ear height (Zhao et al., 2015) can be due to global repression of miRNAs resulting an increase in gene expression.

1.10 Heterosis and genetic diversity in *Brassica napus*

In recent years, exploitation of heterosis has received attention to increase seed yield in *B. napus* canola hybrid cultivars. Heterosis for seed yield has been recorded over 50% in winter and spring canola by Lefort-Buson and Dattee (1982) and Grant and Beversdorf (1985). Several researchers reported that the hybrids of genetically diverse parents show higher heterosis than the hybrids derived from crosses between genetically similar parents (Grant and Beversdorf, 1985; Lefort-Buson et al., 1987a). For example, Lefort-Buston et al. (1987b) reported about 12% high-parent heterosis for seed yield in the hybrids derived from crosses between European and Asian *B. napus*. Ali et al. (1995) also found a positive correlation between genetic divergence of the parents and mid-parent heterosis for seed yield, number of silique per plant and number of seeds per silique in winter *B. napus*. In case of spring *B. napus*, Sernyk and Stefansson (1983) found that hybrids of Marnoo (Australian) × Regent (Canadian) and Karat (European) × Regent (Canadian) out-performed the Canadian cultivar Regent by 38 to 43%. These hybrids had similar agronomic and seed quality traits as the commercial cultivars. Starmer et al. (1998) also reported heterosis for seed yield as well as seed oil content in spring canola hybrids derived from crosses between Canadian and European cultivars. Similarly, Ahmad et al. (2011) found that the hybrids of genetically diverse parents of spring *B. napus* surpasses the seed yield of the hybrids derived from genetically similar parents; however, they did not find any relationship of genetic distance with heterosis for oil content, plant height and maturity. According to Bernardo (1992), hybrid performance can be predicted based on heterozygosity of molecular markers provided that dominance effect of the genes is strong (complete dominance or overdominance), heterotic groups

are complementary, trait heritability is high, and at least 30-50% of the QTL markers are included in the analysis.

In *B. napus*, the genetic divergence between the parents does not always correlate with heterosis, especially in case of extremely diverse parental lines (Diers et al., 1996; for review, see Rahman, 2013). For example, Diers et al. (1996) found that hybrid yield of spring *B. napus* can be predicted more precisely by using genetic distance and general combining ability (GCA) of the parents as independent variables in a multiple linear regression model than by using either genetic distance or general combining ability in a simple linear model. They also reported a significant correlation between genetic distance and specific combining ability (SCA). Thus, selection of genetically diverse superior inbred lines with high general combining ability would be needed to develop high yielding hybrid cultivars. Likewise, Qian et al. (2007) found a significant association between general combining ability of the parents and hybrid performance for seed yield in hybrids derived from crossing of German or Canadian spring canola lines and Chinese semi-winter *B. napus* lines; in this case, the parental genetic divergence was weakly correlated with heterosis. Qian et al. (2009) also found a stronger correlation between GCA of the parents and heterosis than genetic distance of the parents and heterosis in hybrids derived from crossing of Chinese semi-winter to European winter *B. napus*. Furthermore, cytoplasm of the female parent can affect the expression of heterosis; therefore, nucleo-cytoplasmic interaction may also need to be taken into account for full exploitation of this phenomenon (Lefort-Buson and Dattee, 1982).

Of the different primary gene pools of *B. napus,* most of the efforts initiated to date to diversify the genetic base of spring *B. napus* canola is the use of either spring (Rahman et al., 2016) or winter (Quijada et al., 2004; for review, see Rahman, 2013) or semi-winter (Qian et al., 2007; for review, see Rahman, 2013) types of *B. napus*. However, no study has been conducted to evaluate

the potential value of the *B. napus* var. *napobrassica* (Rutabaga) gene pool for broadening the genetic base and increasing the level of heterosis in spring *B. napus* canola for agronomic and quality traits including seed yield. The purpose of this research was to assess the usefulness of this gene pool in the breeding of spring *B. napus* canola hybrid cultivars.

1.11 Research objectives

The long-term objective of this project is to introgress allelic diversity from Rutabaga into spring *B. napus* canola and to develop elite lines for the development of commercial hybrid canola cultivars.

The objectives of this Master's thesis research project were following:

I) Evaluate the spring *B. napus* inbred lines derived from Rutabaga (*B. napus* var. *napobrassica*) × *B. napus* crosses in field trials for agronomic and seed quality traits including seed yield.

II) Evaluate the test hybrids, developed by crossing of the above-mentioned inbred lines to their spring *B. napus* canola parents, for heterosis for different traits including seed yield.

III) Evaluate the above-mentioned inbred lines, derived from Rutabaga × canola crosses, for genetic diversity by use of SSR markers.

Chapter 2

Assessment of spring canola (*Brassica napus*) inbred lines derived from Rutabaga (*Brassica napus* var. *napobrassica*) × canola crosses for agronomic and seed quality traits, and allelic diversity

2.1 Introduction

The changeover from traditional rapeseed to the canola quality crop in the 1970's was a significant breakthrough in the breeding of oilseed *B. napus* (Fu and Gugel, 2010; Przybylski and Eskin, 2011). The removal of the erucic fatty acid from the traditional rapeseed oil has resulted in an excellent fatty acid composition of the seed oil for human consumption and the decrease in the content of glucosinolates of the seed has resulted in an excellent protein-rich meal for use as a protein supplement in animal feed (Canola Council of Canada, 2016c). Improvement of agronomic traits and seed yield in canola have also been achieved through breeding over the past forty years. Prior to the 1990's, canola breeding programs were focused on the development of high yielding open-pollinated cultivars; however, the discovery of cytoplasmic male sterility (CMS) and the occurrence of heterosis in the F_1's of *B. napus* crosses encouraged breeders to develop high yielding hybrid cultivars of this crop (Snowdon et al., 2007). With this improved seed quality and increased productivity, canola has advanced from a marginal crop to the second most important oilseed crop in the world after soybean (Stefansson and Downey, 1995). To meet the global demand of the vegetable oil, increasing the seed or oil yield in this crop is needed.

The seed quality improvement of *B. napus* canola was done using only two genetic variants of *B. napus* – the German cv. Liho (Stefansson et al., 1961) and the Polish cv. Bronowski (Kzrymanski, 1967, reviewed in Stefansson and Downey, 1995). This is one of the causes of the

narrow genetic diversity seen today in *B. napus* canola breeding populations (Fu and Gugel, 2010), and is a hindrance for grouping the breeding materials into distinct heterotic pools, which is needed in a knowledge-based breeding program for development of hybrid cultivars (Girke et al., 2012b). Therefore, broadening of the genetic base of spring *B. napus* canola is needed (for review, see Rahman, 2013).

Several gene pools, genetically distinct from the spring *B. napus* canola, have been identified by different researchers within the genus *Brassica* (Qian et al., 2005; Kebede et al., 2010; Girke et al., 2012a; Bennett et al., 2012; Rahman et al., 2015); these gene pools can be utilized to widen the genetic base of this crop. Introduction of allelic diversity from the secondary gene pool, such as its allied species through interspecific hybridization, is associated with several challenges, such as the difficulty of producing interspecific hybrid plants, sterility in the progeny of the interspecific hybrids, and introduction of undesirable traits from the allied species due to linkage disequilibrium. Therefore, exploitation of the A and C genomes of the allied species for broadening of allelic diversity in spring *B. napus* canola is complicated, and needs intensive breeding efforts (for review, see Rahman, 2013).

Several researchers utilized the primary gene pools of spring *B. napus* canola, such as winter and semi-winter forms of *B. napus*, to increase the diversity of alleles in the breeding materials of spring *B. napus* canola (Quijada et al., 2004; Qian et al., 2007; Kebede et al., 2010; Rahman and Kebede, 2012; Rahman et al., 2016). However, very little research has been conducted to use Rutabaga (*B. napus* var. *napobrassica*), which is known to be genetically distinct from spring *B. napus* canola (Diers and Osborn, 1994, Bus et al., 2011), for broadening the genetic base of spring *B. napus* canola. The variant Rutabaga of *B. napus* has been mainly used as a source of clubroot resistance in the breeding of oilseed *B. napus* (Lüders et al., 2011; Rahman et al., 2014, Hasan and

Rahman, 2016). The potential value of Rutabaga for the improvement of the genetic base of spring *B. napus* canola has been studied by the Canola Program at the University of Alberta. Genetic diversity analysis by use of SSR markers revealed that the canola quality spring growth habit *B. napus* inbred lines derived from Rutabaga × spring *B. napus* canola crosses were genetically distinct from their spring *B. napus* parents (Flad, 2015).

DNA-based molecular markers have been used as a tool to evaluate the extent of genetic diversity exists in breeding populations (for review, see Mondini et al., 2009). The efficiency of a plant breeding program can also be increased by use of molecular markers (for review, see Collard and Mackill, 2008). Restriction fragment length polymorphism (RFLP) markers are the first type of molecular markers that were introduced about thirty years ago for use in breeding. Since then, other types of molecular markers, such as random amplified polymorphism DNA (RAPD), amplified fragment length polymorphism (AFLP) and simple sequence repeats (SSR), have been developed for different applications in a breeding program (for review, see Jonah et al., 2011). Several researchers used SSR markers to examine genetic diversity in *Brassica* gene pools (Hasan et al., 2006; Zhou et al., 2006; Bus et al., 2011). Recently, single nucleotide polymorphism (SNP) markers gained much interest for use in breeding (Ganal et al., 2012; Qian et al., 2014).

The objective of this study was to assess the agronomic and seed quality traits of a set of advanced generation F_2- and BC_1-derived inbred lines developed from two crosses involving a Rutabaga line and two spring *B. napus* cultivars. These populations were also analyzed by use of SSR markers to estimate the extent of allelic diversity introgressed from Rutabaga into the inbred lines.

2.2 Materials and methods

2.2.1 Plant materials

Two spring *B. napus* (AACC, 2*n* = 38) canola cultivars, Hi-Q (a conventional type) and A07-26NR (RoundUp herbicide resistant), both developed by the Canola Program of the University of Alberta, and a Rutabaga (*B. napus* var. *napobrassica*) line, Rutabaga-BF, developed from the Rutabaga cv. Brookfield through single plant selection, were used as parents of the materials used in this study. Rutabaga is also known as swede (for review, see Prakash and Hinata, 1980); this plant forms round or oval shaped-roots, and is commonly grown as a vegetable for human consumption or fodder for animals (Rakow, 2004).

The plant materials used in this research were developed from the following two crosses: Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR. The F_1 plants of the two crosses were self-pollinated to produce F_2 populations. To increase the probability of developing a canola quality line, the F_1 plants were also backcrossed to their respective canola parents, Hi-Q or A07-26NR, to produce BC_1 seeds. The two F_2 and BC_1 populations were subjected to pedigree breeding by the Canola Program for the development of canola quality spring growth habit inbred lines (IN) (Figures 2.1 and 2.2).

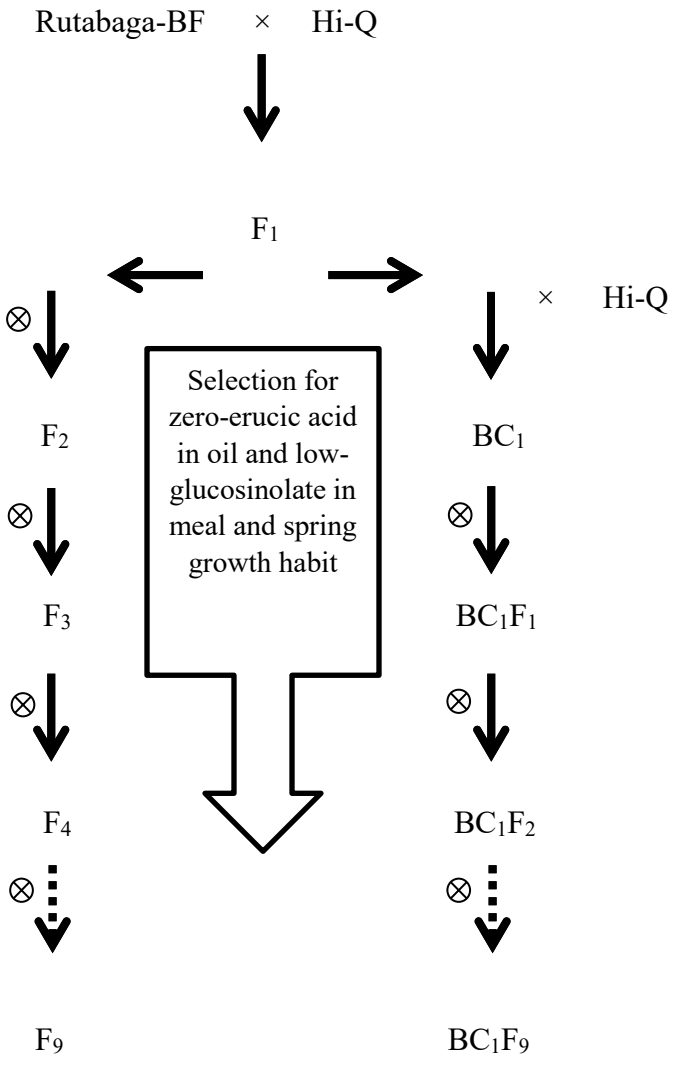

Figure 2.1: A flow diagram showing the development of canola quality spring *B. napus* lines from the Rutabaga-BF × Hi-Q cross. To develop the inbred lines, the F_1 plants were subjected to two breeding pathways: successive self-pollinations (⊗) of the F_2, and backcrossing (BC) of the F_1 to the spring *B. napus* canola parent Hi-Q followed by self-pollination for several generations. Selection for canola quality traits was done in the F_2- and BC_1-derived populations. Twenty five and 26 advanced generation lines derived, respectively, from the BC_1 and F_2 populations were used to evaluate for agronomic and seed quality traits in the field trials.

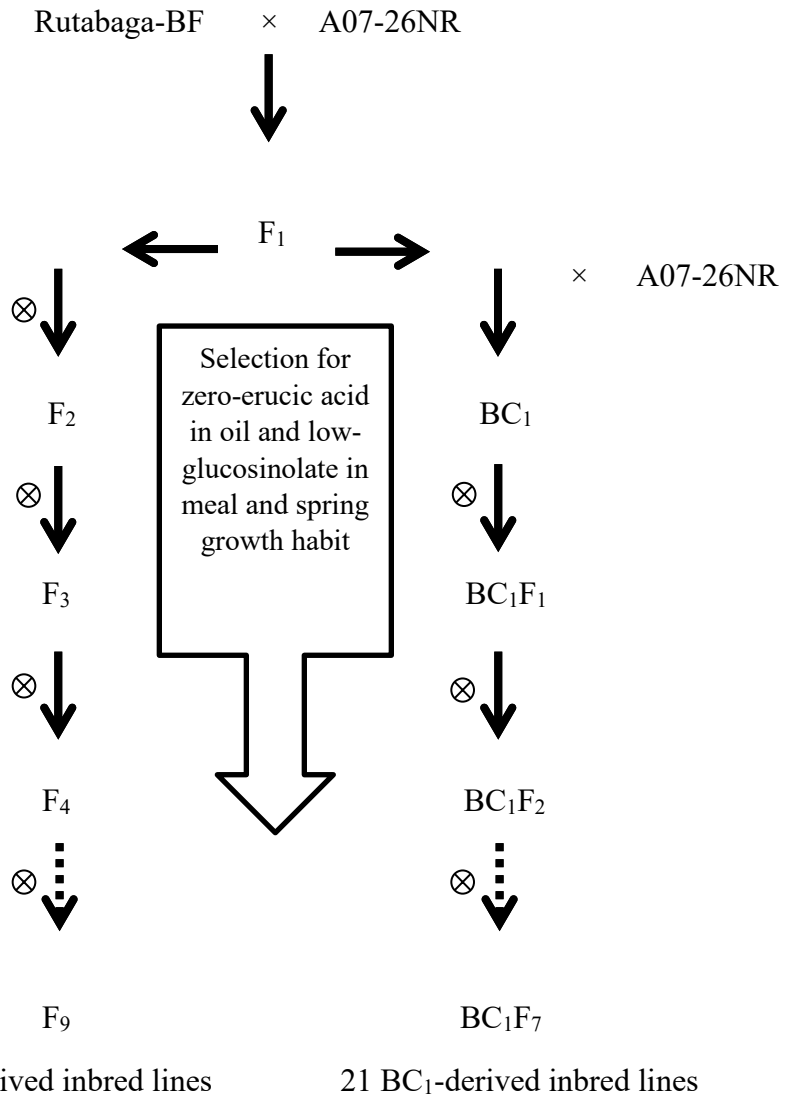

Figure 2.2: A flow diagram showing the development of canola quality spring *B. napus* lines from the Rutabaga-BF × A07-26NR cross. To develop the inbred lines, the F_1 plants were subjected to two breeding pathways: successive self-pollinations (⊗) of the F_2, and backcrossing (BC) of the F_1 to the spring *B. napus* canola parent A07-26-NR followed by self-pollination for several generations. Selection for canola quality traits was done in the F_2- and BC_1-derived populations. Twenty one and 21 advanced generation lines derived, respectively, from the BC_1 and F_2 populations were used to evaluate for agronomic and seed quality traits in the field trials.

For this study, I received seeds of 26 F_9 and 25 BC_1F_9 lines of the Rutabaga-BF × Hi-Q cross, and 21 F_9 and 21 BC_1F_7 lines of the Rutabaga-BF × A07-26NR cross. These populations were grown in the field in summer 2015 and open-pollinated seeds were harvested and used for replicated field trials (Table 2.1).

Table 2.1: The F_2- and BC_1-derived advanced generation inbred lines of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses of *B. napus* used in this study.

Cross	Population	Generation[1]	No. inbred lines
Rutabaga-BF × Hi Q	F_2-derived	IN-F_9	26
	BC_1-derived	IN-BC_1F_9	25
Rutabaga-BF × A07-26NR	F_2-derived	IN-F_9	21
	BC_1-derived	IN-BC_1F_7	21
Total			93

[1] Advanced generation inbred lines (IN) of the F_2- or BC_1-derived population developed from either Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR. For example, IN-F_9 is the ninth generation inbred line derived from the F_2 population of the Rutabaga-BF × Hi-Q cross, and IN-BC_1F_7 is the seventh generation inbred line derived from the BC_1 population of the Rutabaga-BF × A07-26NR cross.

2.2.2 Field trial with the inbred lines

The 93 inbred lines were evaluated at the following sites in Alberta in 2016: Edmonton Research station and St. Albert Research farm of the University of Alberta and a grower's field in Killam (four sites). Field trial in each location was laid out in a randomized incomplete block design with two replications. The *B. napus* canola parents Hi-Q and A07-26NR were also included as checks in the field trials. Due to winter-type growth habit of Rutabaga, this parent of the inbred lines was not included in the field evaluations. By the use of the software program CropStat 7.2 (International Rice Research Institute, Los Baños, Philippines), the inbred lines and the checks were randomly assigned to the experimental plots of four incomplete blocks within each of the

two replications. Plot size was 5 m long and 1.8 m wide (9 m^2) and consisted of six planting rows with 0.25 m space between the rows. Seeding was done with a plot seeder at the rate of 4.5 g seed/plot to achieve plant density of about 75 plants per square meter.

These inbred lines, with the same experimental specifications as in 2016, were also evaluated in field trials at the Edmonton Research station, St. Albert Research farm and Killam (three sites) in 2014 for agronomic and seed quality traits by the Canola Program; I received this data set and included in the statistical analysis of the inbred lines.

2.2.3 Data collection

The following traits were recorded:

1. Plant vigor: Recorded at rosette stage on a 0 - 9 scale, where '9' is very vigorous and '0' is very poor.
2. Days to start of flowering: Recorded when approximately 50% of the plants in the plot had at least one open flower.
3. Plant height: Recorded at the end of flowering in cm.
4. Days to maturity: Recorded when about 50% seeds on the main stem of the plants began to turn to brown or black.
5. Lodging: Recorded before harvest on a 0 - 9 scale, where '0' is very stiff and completely standing, and '9' is totally lodged.
6. Seed yield: Recorded on the whole plot basis and data was converted to kg/ha.

Seed oil (%), protein (%) and glucosinolate (µmol/g seed) contents were estimated by near infrared spectroscopy (NIRS) method (Foss NIR system, model 6500, Eden Prairie, Minnesota, USA) in

the analytical laboratory of the Canola Program of the University of Alberta.

2.2.4 Statistical analysis

The Microsoft Excel software was used to organize the agronomic and seed quality data, and the software program 'R' (R Core Team, 2014), version 3.2.2 was used for statistical analysis of this data. The year/site of the field trials with the inbred lines was considered as an experimental environment. For analysis of variance (ANOVA), environment, replication and block were considered as random-effects, and the cross, the type of population (F_2- or BC_1-derived) and inbred line were considered as fixed-effects. The lmer function of the 'lme4' package (Lenth, 2015) was used to fit a linear mixed-effects model (both random- and fixed-effects factors were incorporated) for each response variable including the agronomic and seed quality traits as follows:

Inbred line.lmer = lmer (Trait ~ Cross + Population + Cross : Population/Inbred line
+ (1|Environment) + (1|Replication) + (1|Block), Inbred line.data)

For each trait, analysis of variance was done to test statistical significance of the fixed-effects (cross, population and inbred line) terms of the model. Lsmeans function of the 'lsmeans' package (Bates et al., 2015) was used to calculate least-squares mean (lsmean) values of the inbred lines, crosses and populations for different traits. The Tukey's test was done to compare the mean values for significant difference ($a \leq 0.05$).

2.2.5 Molecular marker analysis

Simple sequence repeats (SSR) markers were used to evaluate the extent of allelic diversity introgressed from Rutabaga-BF into the inbred lines developed from the Rutabaga-BF × spring canola crosses. The 93 inbred lines, derived from F_2 and BC_1 populations of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses (Table 2.1), were genotyped by polymorphic SSR markers. The majority of the genotyping work was done by the Canola Program. To complete

genotypic data of the 93 inbred lines, I genotyped 45 lines with 54 SSR markers. List of the inbred lines and their DNA codes, and the list of the polymorphic SSR markers used for genotyping are presented in Appendices (Tables A2.1 and A2.2).

2.2.5.1 DNA extraction

About 100 mg young leaf (4-week-old) of the inbred lines and their parents grown in a greenhouse were collected for extraction of DNA. The leaf samples were placed in 1.5 ml Eppendorf tubes and kept at -80 °C until use. For extraction of DNA, about 50 mg of the leaf sample was placed in a 1.5 ml Eppendorf tube, and the tube was immersed in liquid nitrogen for one minute and the frozen leaf sample was immediately grounded using a micropestal. Extraction of DNA was done using a SIGMA DNA extraction kit (Sigma-Aldrich, St. Louis, Missouri, USA) and following the protocol suggested by the manufacturer. The quality and concentration of the DNA was measured using a NanoDrop 2000c spectrophotometer (Thermo Fisher Scientific Inc., Wilmington, Delaware, USA), and the DNA concentration was adjusted to 20 ± 5 ng/µl for use in polymerase chain reactions (PCR).

2.2.5.2 Polymerase chain reactions (PCR)

The inbred lines of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses were genotyped, respectively, by 87 and 105 SSR markers from the 19 linkage groups of *B. napus* (Tables A2.1 and A2.2).

The genomic DNA was amplified using a GeneAmp PCR System 9700 thermal cycler (Applied Biosystems, Foster City, California, USA). For this, a reaction mixture of a total volume of 15.66 µl consisting of 4 µl of the template DNA, 2.5 µl of 10x PCR buffer, 1.0 µl of 25 mM $MgCl_2$, 0.25 µl of each 10 µM forward and reverse primers, 0.25 µl of 2 µM fluorescent dye, 0.25

µl of 10µ mM dNTPs mix (Invitrogen, Life Technologies Inc., Burlington, Ontario, Canada), 0.12 µl of 5.0 µl Taq DNA polymerase (Promega Corporation, Madison, Wisconsin, USA) and 7.04 µl of distilled water was prepared in a microtube of a 96 well PCR plate. PCR programme included initial denaturation for 5 min at 94 °C and 35 cycles of denaturation for 1 min at 94 °C, annealing for 1 min at 60 °C and extension for 1 min at 72°C, followed by final extension at 72 °C for 30 min. The size of the amplicons was analyzed using a capillary ABI sequencer No. 3730 (Applied Biosystems, Foster City, California, USA).

2.2.5.3 Genotyping of the inbred lines and data analysis

The software GeneMarker version 2.4.0 (SoftGenetics, State College, Pennsylvania, USA) was used for binomial classification of the amplicons where the scores 1 and 0 were assigned for the presence and absence of a fragment. The binary score data was transferred to a spreadsheet; data matrix of the F_2- and BC_1-derived inbred lines of the two crosses were created separately. The number, and the percentage (of the total number of detected alleles) of the Rutabaga-BF alleles detected in the F_2- and BC_1-derived populations were calculated for each linkage group.

Dice genetic similarity coefficients (Nei and Li, 1979) between the inbred lines were calculated by using the software program Numerical Taxonomy and Multivariate Analysis System (NTSYSpc 2.2) (Rohlf, 2000). The similarity coefficients were used for cluster analysis following the unweighted pair-group method with arithmetic mean (UPGMA) and using the same software program. By use of the same software program, principal coordinate analysis was also done to estimate the genetic relationship between the F_2- and BC_1-derived populations of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses.

The extent of genetic variation present within and between the populations was evaluated by

analysis of molecular variance (AMOVA) using the software program Arlequin version 3.5 (Excoffier and Lischer, 2010).

2.3 Results

2.3.1 Days to flowering

The results of analysis of variance for days to flowering of the two crosses, two types of population and inbred lines are shown in Table 2.2. Significant variations were found between the two crosses, the BC_1- and F_2-derived populations, as well as among the inbred lines ($p < 0.001$). Interaction between the cross and population type (F_2- or BC_1-derived) was non-significant.

Table 2.2: Analysis of variance for days to flowering (DTF) of the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus*. Two (2014 and 2016) years data was used for analysis of variance.

Source	df	Error df[6]	SS	MS	F value	p value[7]
Environment[1]	6		229.34	38.22		
Replication	1		0.06	0.06		
Block	3		0.09	0.03		
Cross[2] [C]	1	836.39	1089.60	1089.60	563.64	$< 2\times10^{-16}$ ***
Population[3] [P]	1	943.55	177.41	177.41	91.77	$< 2\times10^{-16}$ ***
C × P[4]	1	905.39	3.59	3.59	1.86	0.173
Inbred line (C × P)[5]	89	989.67	1564.64	17.58	9.09	$< 2\times10^{-16}$ ***
Residual	989		1911.90	1.93		
Total	1091		4976.61			

[1] Year/site of the inbred line field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the inbred lines were developed.
[3] Type of population indicates from where, the BC_1 or F_2, the inbred lines were developed.
[4] Combination of the cross and type of population.
[5] Inbred line is nested in C × P.
[6] Due to unbalanced data for days to flowering, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

Of the two populations derived from two crosses, the Conventional (non-RoundUp tolerant) population derived from the Rutabaga-BF × Hi-Q cross flowered later than the RoundUp Ready population derived from the Rutabaga-BF × A07-26NR cross (52.7 ± 2.3 vs. 50.5 ± 2.3 days).

Compared to the F_2-derived population, which on average took 51.3 ± 2.4 days to flower, the BC_1-derived population took about 0.8 days longer (52.1 ± 2.4 days) to flower. Days to flowering of the four populations derived from the two crosses were significantly different ($p < 0.05$). The BC_1-derived population of both crosses took longer time to flower as compared to the F_2-derived population. While comparing with the spring canola parent, the two populations derived from Rutabaga-BF × Hi-Q took significantly longer time to flower than Hi-Q. In case of the population derived from Rutabaga-BF × A07-26NR, the F_2-derived population flowered similar to A07-26NR, while the BC_1-derived population took longer time than A07-26NR (Table 2.3).

Table 2.3: Days to flowering (DTF) of the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus* in 2014 and 2016 field trials.

Cross/*B. napus* parent/ type of population[1]	Inbred line population[7]	2014	2016	Pooled[8]	
		Mean ± SE	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	50.8 ± 1.6 a	54.6 ± 4.0 a	49.8 – 54.6	53.1 ± 2.3 a
	F	50.2 ± 1.6 a	53.9 ± 4.0 b	50.0 – 55.2	52.3 ± 2.4 b
Rut-BF × 26NR[3]	BC	48.9 ± 1.6 b	52.6 ± 4.0 cb	49.3 – 53.5	50.9 ± 2.4 c
	F	46.9 ± 1.6 c	51.8 ± 4.0 e	48.8 – 52.6	49.9 ± 2.4 d
Hi-Q[4]		48.7 ± 1.5 bc	53.1 ± 4.0 b		51.3 ± 2.4 c
A07-26NR[4]		47.1 ± 1.5 bc	50.7 ± 4.0 d		49.1 ± 2.4 d
Rut-BF × Hi-Q		50.5 ± 1.6 a	54.2 ± 4.0 a	49.8 – 55.2	52.7 ± 2.3 a
Rut-BF × 26NR		47.9 ± 1.6 a	52.2 ± 4.0 b	48.8 – 53.5	50.5 ± 2.3 b
BC[5]		49.7 ± 1.7 a	53.7 ± 4.0 a	49.3 – 54.6	52.1 ± 2.4 a
F[6]		48.7 ± 1.7 a	53.0 ± 4.0 b	48.8 – 55.2	51.3 ± 2.4 b

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the inbred lines were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents used as checks in the field trials.
[5] and [6] = BC_1- and F_2-derived inbred lines, respectively.
[7] BC_1- and F_2-derived inbred line population of the two original crosses.
[8] 2014 and 2016 data was used to calculate the pooled lsmeans values of the inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Frequency distribution of the BC_1- and F_2-derived inbred lines of the two crosses is presented in Figure 2.1. Approximately 4% of the BC_1-derived inbred lines of Rutabaga-BF × Hi-Q, and

14.3% and 57.1%, respectively, of the BC_1- and F_2-derived inbred lines of Rutabaga-BF × A07-26NR required fewer days to flower compared to the respective spring canola parent (Figure 2.3).

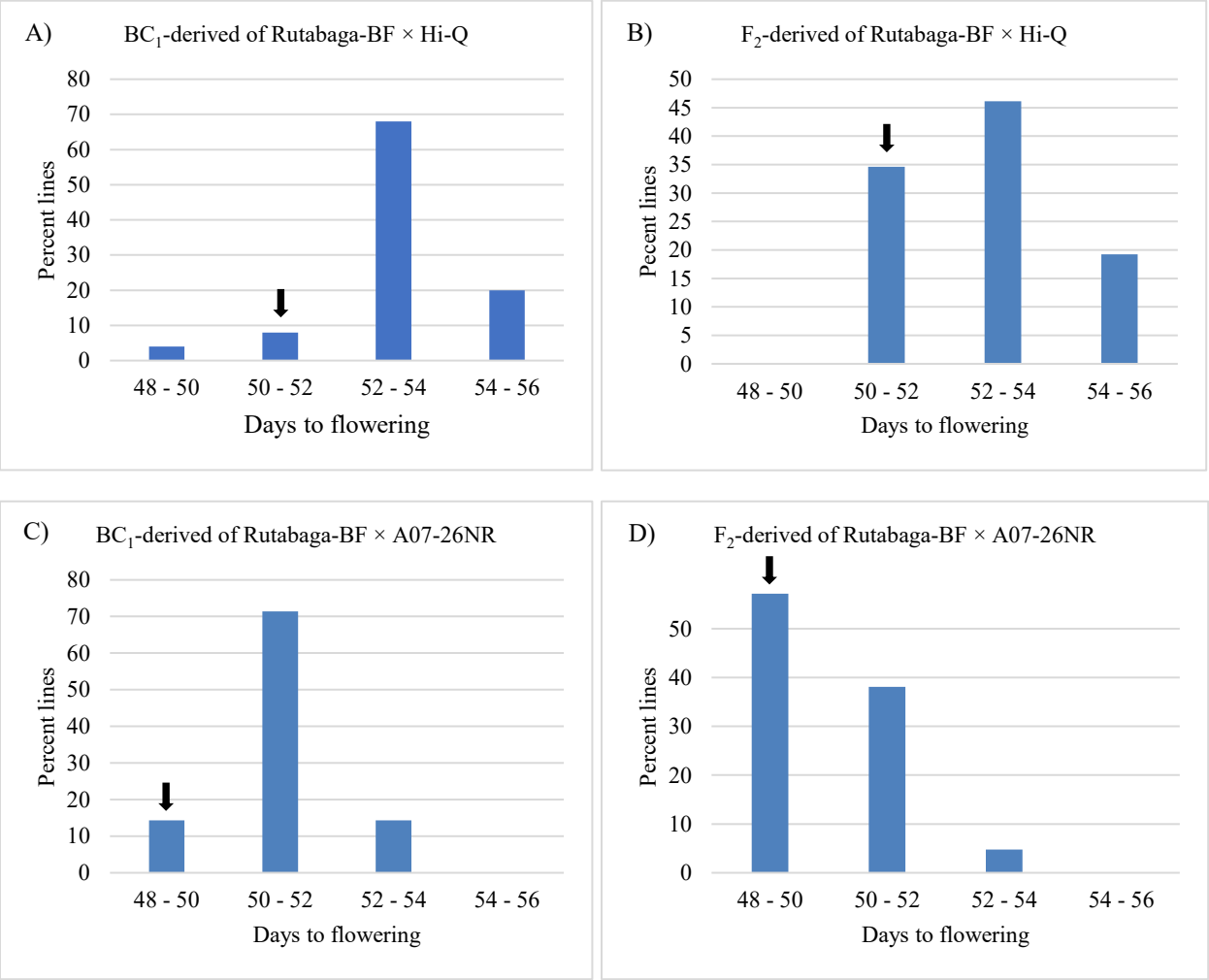

Figure 2.3: Frequency distribution of the BC_1- and F_2-derived inbred lines of Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR of *B. napus* for days to flowering. Pooled data of the 2014 and 2016 field trials for days to flowering was used to categorize the BC_1- and F_2-derived inbred lines of Rutabaga-BF × Hi-Q (graphs A and B) and Rutabaga-BF × A07-26NR (graphs C and D). The arrows show the position of the spring *B. napus* canola parent (Hi-Q or A07-26NR).

The inbred lines CO.BC.IN.18, CO.BC.IN.29, CO.FF.IN.17, CO.FF.IN.18, CO.FF.IN.24, CO.FF.IN.25, CO.FF.IN.26, CO.FF.IN.30, CO.FF.IN.31, CO.FF.IN.39 and CO.FF.IN.40 of Rutabaga-BF × Hi-Q, and RR.FF.IN.27 of Rutabaga-BF × A07-26NR flowered earlier than their spring canola parent (Tables A3.8, A3.9, A3.10 and A3.11).

2.3.2 Days to maturity

Analysis of variance for days to maturity is presented in Table 2.4. Significant difference ($p < 0.001$) among the crosses, populations and inbred lines were found for this trait.

Table 2.4: Analysis of variance for days to maturity (DTM) of the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus*. Two (2014 and 2016) years data was used for analysis of variance.

Source	df	Error df[6]	SS	MS	F value	p value[7]
Environment[1]	5		522.5	104.5		
Replication	1		0.30	0.30		
Block	3		0.15	0.05		
Cross[2] [C]	1	360.06	985.75	985.75	203.15	$< 2.2\times10^{-16}$ ***
Population[3] [P]	1	521.59	102.00	102.04	21.03	5.7×10^{-6} ***
C × P[4]	1	518.35	0.96	0.96	0.20	0.6561
Inbred line (C × P)[5]	89	805.25	1168.36	13.13	2.71	2.6×10^{-13} ***
Residual	805		104.50	4.85		
Total	906		2884.56			

[1] Year/site of the inbred line field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the inbred lines were developed.
[3] Type of population indicates from where, the BC_1 or F_2, the inbred lines were developed.
[4] Combination of the cross and type of population.
[5] Inbred line is nested in C × P.
[6] Due to unbalanced data for days to maturity, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

The population derived from Rutabaga-BF × Hi-Q required about 103.3 ± 4.2 days to mature; this was 2.5 days later than the population derived from Rutabaga-BF × A07-26NR (100.8 ± 4.2 days). Thus, a greater number of early maturing lines could be found in the population derived from Rutabaga-BF × A07-26NR as compared to the population derived from the Rutabaga-BF × Hi-Q cross. On average, the BC_1-derived population took significantly greater number of days to mature than that of the F_2-derived population (102.5 ± 4.2 vs. 101.8 ± 4.2 days). While comparing with the spring canola parent, the BC_1- or F_2-derived populations were not significantly different from Hi-Q or A07-26NR. Significant variation was found within the populations derived from the

two crosses suggesting the possibility of selection of early maturing lines from these populations (Table 2.5).

Table 2.5: Days to maturity (DTM) of the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus* in 2014 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Inbred line population[7]	2014	2016	Pooled[8]	
		Mean ± SE	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	98.2 ± 2.4 a	109.3 ± 6.9 a	101.4 – 105.5	103.7 ± 4.2 a
	F	97.7 ± 2.4 ab	108.6 ± 6.9 a	101.1 – 105.1	102.9 ± 4.2 b
Rut-BF × 26NR[3]	BC	94.8 ± 2.4 bc	107.2 ± 6.9 bc	99.0 – 104.3	101.1 ± 4.2 c
	F	93.0 ± 2.4 d	106.9 ± 6.9 bc	98.7 - 103.4	100.5 ± 4.2 c
Hi-Q[4]		97.8 ± 2.3 ab	108.1 ± 6.9 ab		102.7 ± 4.2 ab
A07-26NR[4]		93.1 ± 2.3 cd	106.2 ± 6.9 c		99.9 ± 4.2 c
Rut-BF × Hi-Q		98.0 ± 2.3 a	108.9 ± 6.9 a	101.1 – 105.5	103.3 ± 4.2 a
Rut-BF × 26NR		93.9 ± 2.3 a	107.1 ± 6.9 b	98.7 -104.3	100.8 ± 4.2 b
BC[5]		96.4 ± 2.6 a	108.3 ± 6.9 a	99.0 – 105.5	102.5 ± 4.2 a
F[6]		95.5 ± 2.6 a	107.8 ± 6.9 a	98.7 – 105.1	101.8 ± 4.2 b

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the inbred lines were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents used as checks in the field trials.
[5] and [6] = BC_1- and F_2-derived inbred lines, respectively.
[7] BC_1- and F_2-derived inbred line population of the two original crosses.
[8] 2014 and 2016 data was used to calculate the pooled lsmeans values of the inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

The inbred lines CO.BC.IN.18, CO.BC.IN.20, CO.BC.IN.21, CO.BC.IN.27, CO.BC.IN.31, CO.BC.IN.32, CO.FF.IN.13, CO.FF.IN.17, CO.FF.IN.18, CO.FF.IN.22, CO.FF.IN.26, CO.FF.IN.27, CO.FF.IN.30, CO.FF.IN.39, CO.FF.IN.40 of Rutabaga-BF × Hi-Q and the lines RR.BC.IN.18, RR.BC.IN.19, RR.BC.IN.21, RR.BC.IN.31, RR.FF.IN.20, RR.FF.IN.21, RR.FF.IN.23, RR.FF.IN.29 and RR.FF.IN.31 of Rutabaga-BF × A07-26NR matured earlier than their respective spring canola parent (Tables A3.8, A3.9, A3.10 and A3.11).

2.3.3 Plant height

Significant variation between the two crosses and among the inbred lines was found for plant height ($p < 0.001$) (Table 2.6).

Table 2.6: Analysis of variance for plant height of the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus*. Two (2014 and 2016) years data was used for analysis of variance.

Source	df	Error df[6]	SS	MS	F value	p value[7]
Environment[1]	6		1129.86	188.31		
Replication	1		0.05	0.05		
Block	3		0.21	0.07		
Cross[2] [C]	1	249.97	32491.00	32491.00	533.52	$< 2.2\times10^{-16}$ ***
Population[3] [P]	1	514.65	189.00	189.00	3.11	0.0785
C × P[4]	1	357.06	3637.00	3637.00	59.72	1.1×10^{-13} ***
Inbred line (C × P)[5]	89	982.70	24051.00	270.00	4.44	$< 2.2\times10^{-16}$ ***
Residual	989		60230.10	60.90		
Total	1091		121728.22			

[1] Year/site of the inbred line field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the inbred lines were developed.
[3] Type of population indicates from where, the BC_1 or F_2, the inbred lines were developed.
[4] Combination of the cross and type of population.
[5] Inbred line is nested in C × P.
[6] Due to unbalanced data for plant height, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

2016 and pooled (the 2014 and 2016 data combined) data showed that the population derived from the Rutabaga-BF × Hi-Q cross was significantly taller than the population derived from the Rutabaga-BF × A07-26NR cross (127.2 ± 5.3 vs. 115.4 ± 5.3 cm); however, the difference between the populations of these crosses was not significant in 2014. In the case of Rutabaga-BF × Hi-Q, the F_2-derived population was taller than the BC_1-derived population, while the BC_1-derived population was taller than the F_2-derived population in case of Rutabaga-BF × A07-26NR. While combining data of the two crosses, no significant difference for this trait was found between the F_2- and BC_1-derived populations, and these populations were also not significantly different from

their spring *B. napus* parent Hi-Q or A07-26NR. Of the four populations, the shortest population was the F_2-derived populations of Rutabaga-BF × A07-26NR (114.1 ± 5.2 cm) (Table 2.7).

Table 2.7: Plant height (cm) of the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus* in 2014 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Inbred line population[7]	2014	2016	Pooled[8]	
		Mean ± SE	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	118.6 ± 11.4 a	129.3 ± 5.1 b	117.3 – 132.0	124.9 ± 5.2 b
	F	124.3 ± 11.4 a	133.6 ± 5.1 a	120.0 – 139.3	129.7 ± 5.2 a
Rut-BF × 26NR[3]	BC	111.4 ± 11.4 ab	120.9 ± 5.1 c	108.4 – 134.1	116.8 ± 5.2 c
	F	107.9 ± 11.5 b	118.2 ± 5.1 c	108.2 – 124.9	114.1 ± 5.2 d
Hi-Q[4]		127.5 ± 11.1 a	128.4 ± 5.3 b		127.0 ± 5.4 ab
A07-26NR[4]		113.7 ± 11.1 ab	116.5 ± 5.3 c		114.4 ± 5.4 cd
Rut-BF × Hi-Q		121.4 ± 11.0 a	131.5 ± 5 a	117.3 – 139.3	127.2 ± 5.3 a
Rut-BF × 26NR		109.7 ± 11.0 a	119.5 ± 5 b	108.2 – 134.1	115.4 ± 5.3 b
BC[5]		114.7 ± 11.5 a	125.5 ± 5.1 a	108.4 – 134.1	121.2 ± 5.3 a
F[6]		116.4 ± 11.5 a	126.7 ± 5.1 a	108.2 – 139.3	122.9 ± 5.3 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the inbred lines were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents used as checks in the field trials.
[5] and [6] = BC_1- and F_2-derived inbred lines, respectively.
[7] BC_1- and F_2-derived inbred line population of the two original crosses.
[8] 2014 and 2016 data was used to calculate the pooled lsmeans values of the inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Twenty-six (17 BC_1- and 9 F_2-derived) and 18 (4 BC_1- and 14 F_2-derived) inbred lines, respectively, of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses were shorter than their spring canola parent (Tables A3.8, A3.9, A3.10 and A3.11).

2.3.4 Seed yield

Results of the ANOVA for seed yield is presented in Table 2.8. Significant ($p < 0.001$) variation between the two crosses and the two populations (BC_1- and F_1-derived) was found for

seed yield. Variation among the inbred lines was significant ($p < 0.01$). No significant cross × population interaction was found for this trait.

Average seed yield of the inbred lines derived from the two crosses was significantly lower than their spring canola parent; however, the inbred lines derived from Rutabaga-BF × A07-26NR gave higher yield than the inbred lines of the Rutabaga-BF × Hi-Q cross. The F_2-derived population of both crosses gave significantly higher seed yield than the BC_1-derived population. When comparing the four populations, the F_2-derived population of Rutabaga-BF × A07-26NR

Table 2.8: Analysis of variance for seed yield of the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus*. Two (2014 and 2016) years data was used for analysis of variance.

Source	df	Error df[6]	SS	MS	F value	p value[7]
Environment[1]	6		2918718.60	486453.10		
Replication	1		620.70	620.70		
Block	3		0.00	0.00		
Cross[2] [C]	1	976.71	15426998.00	15426998.00	81.38	$< 2.2\times10^{-16}$ ***
Population[3] [P]	1	976.05	12146067.00	12146067.00	64.07	3.3×10^{-15} ***
C × P[4]	1	975.96	65694.00	65694.00	0.35	0.5562
Inbred line (C × P)[5]	89	975.74	33559827.00	377077.00	1.99	5.5×10^{-7} **
Residual	979		185589421.60	189570.40		
Total	1081		249707346.90			

[1] Year/site of the inbred line field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the inbred lines were developed.
[3] Type of population indicates from where, the BC_1 or F_2, the inbred lines were developed.
[4] Combination of the cross and type of population.
[5] Inbred line is nested in C × P.
[6] Due to unbalanced data for seed yield, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

gave the highest seed yield (3442.0 ± 269.3 kg/ha) while the BC_1-derived population of Rutabaga-BF × Hi-Q gave the lowest (2962.4 ± 268.8 kg/ha) seed yield (Table 2.9).

Table 2.9: Seed yield (kg/ha) of the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus* in 2014 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Inbred line population[7]	2014	2016	Pooled[8]	
		Mean ± SE	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	2478.5 ± 204.9 c	3338.6 ± 356.0 d	2766.1 – 3447.8	2962.4 ± 268.8 e
	F	2491.3 ± 206.8 c	3593.7 ± 355.9 c	2813.0 – 3478.3	3154.0 ± 268.9 d
Rut-BF × 26NR[3]	BC	2680.8 ± 206.9 b	3554.2 ± 356.3 c	2872.3 – 3686.6	3176.7 ± 269.1 cd
	F	3028.8 ± 210.1 a	3795.9 ± 356.3 b	2770.0 – 3737.1	3442.0 ± 269.3 b
Hi-Q[4]		2948.7 ± 164.3 ab	3724.2 ± 365.2 c		3376.5 ± 276.8 bc
A07-26NR[4]		3032.9 ± 164.3 ab	4116.7 ± 365.2 a		3672.8 ± 276.8 a
Rut-BF × Hi-Q		2483.7 ± 159.9 a	3468.9 ± 355.2 b	2766.1 – 3478.3	3051.8 ± 270.9 b
Rut-BF × 26NR		2847.7 ± 162.0 a	3675.7 ± 355.3 a	2770.0 – 3737.1	3298.5 ± 271.1 a
BC[5]		2557.2 ± 187.5 a	3436.0 ± 355.1 b	2766.1 – 3686.6	3053.2 ± 270.4 b
F[6]		2778.5 ± 189.1 a	3683.4 ± 355.1 a	2770.0 – 3737.1	3279.8 ± 270.5 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the inbred lines were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents used as checks in the field trials.
[5] and [6] = BC_1- and F_2-derived inbred lines, respectively.
[7] BC_1- and F_2-derived inbred line population of the two original crosses.
[8] 2014 and 2016 data was used to calculate the pooled lsmeans values of the inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Frequency distribution of the BC_1- and F_2-derived inbred lines of the two crosses is presented in Figure 2.4. The proportion of inbred lines yielding greater than 3.4 ton/ha was higher in the F_2-derived lines than the BC_1-derived lines (15.4% vs. 4.0% in Rutabaga-BF × Hi-Q and 66.7% vs. 19.0% in Rutabaga-BF × A07-26NR).

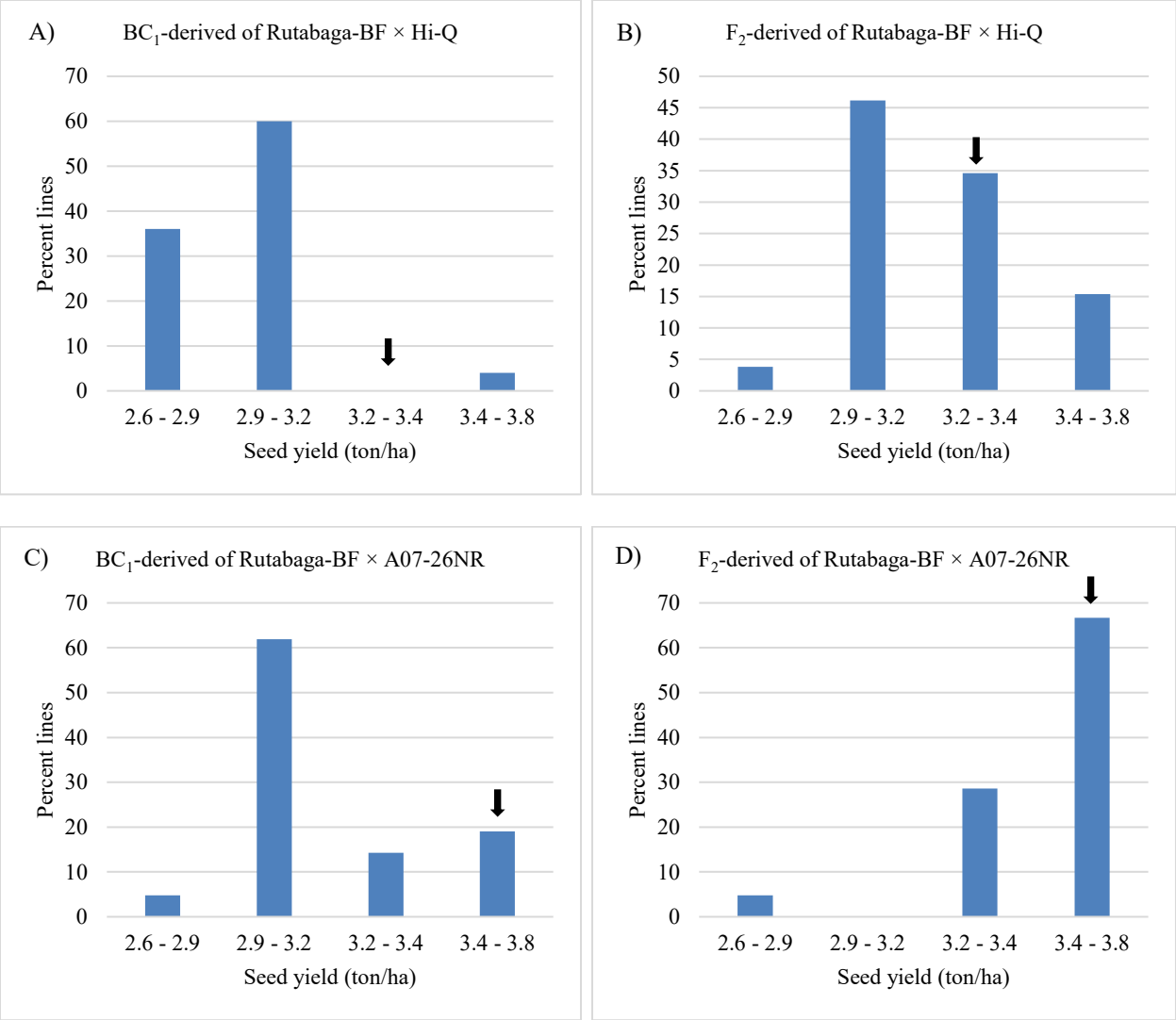

Figure 2.4: Frequency distribution of the BC_1- and F_2-derived inbred lines of Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR of *B. napus* for seed yield. Pooled data of the 2014 and 2016 field trials for seed yield was used to categorize the BC_1- and F_2-derived inbred lines of Rutabaga-BF × Hi-Q (graphs A and B) and Rutabaga-BF × A07-26NR (graphs C and D). The arrows show the position of the spring *B. napus* canola parent (Hi-Q or A07-26NR).

The inbred lines CO.BC.IN.18, CO.FF.IN.18, CO.FF.IN.22, CO.FF.IN.26, CO.FF.IN.30 of Rutabaga-BF × Hi-Q and RR.BC.IN.30 and RR.FF.IN.37 of Rutabaga-BF × A07-26NR gave higher seed yield than their spring canola parent (Tables A3.8, A3.9, A3.10 and A3.11). However, seed yield of the five inbred lines of Rutabaga-BF × Hi-Q was surpassed by the two inbred lines of Rutabaga-BF × A07-26NR.

2.3.5 Seed oil content

The analysis of variance for seed oil content, presented in Table 2.10, revealed that significant variation existed between the two crosses, the two populations as well as among the inbred lines for this seed quality trait ($p < 0.001$).

No significant difference was found between the two types of population of the Rutabaga-BF × Hi-Q cross for seed oil content in both 2014 and 2016; however, the F_2-derived population of Rutabaga-BF × A07-26NR showed higher seed oil content than the BC_1-derived population of this

Table 2.10: Analysis of variance for seed oil content of the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus*. Two (2014 and 2016) years data was used for analysis of variance.

Source	df	Error df[6]	SS	MS	F value	p value[7]
Environment[1]	6		25.35	4.22		
Replication	1		0.01	0.01		
Block	3		0.06	0.02		
Cross[2] [C]	1	820.28	656.02	656.02	517.98	$< 2.2\times10$ ***
Population[3] [P]	1	932.40	72.50	72.50	57.24	9.2×10^{-14} ***
C × P[4]	1	895.01	120.33	120.33	95.01	$< 2.2\times10^{-16}$ ***
Inbred line (C × P)[5]	89	986.52	1494.55	16.79	13.26	$< 2.2\times10^{-16}$ ***
Residual	986		1248.77	1.27		
Total	1088		3617.58			

[1] Year/site of the inbred line field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the inbred lines were developed.
[3] Type of population indicates from where, the BC_1 or F_2, the inbred lines were developed.
[4] Combination of the cross and type of population.
[5] Inbred line is nested in C × P.
[6] Due to unbalanced data for seed oil content, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

cross (Table 2.11). The F_2- and BC_1-derived populations of Rutabaga-BF × Hi-Q were not significantly different from the spring canola parent Hi-Q for this trait. In the case of the Rutabaga-BF × A07-26NR cross, seed oil content of the F_2-derived population was statistically similar to A07-26NR; however, the BC_1-derived population had significantly lower oil content than this

parent. The highest seed oil content was found in the F_2-derived population of Rutabaga-BF × A07-26NR, while the two populations of Rutabaga-BF × Hi-Q had similar and the lowest level of oil content (Table 2.11). On average, the population derived from the Rutabaga-BF × A07-26NR cross had higher oil content than the population derived from the Rutabaga-BF × Hi-Q cross, and the F_2-derived population had higher oil content than the BC_1-derived population.

Table 2.11: Seed oil content (%) of the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi- Q/A07-26NR crosses of *B. napus* in 2014 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Inbred line population[7]	2014	2016	Pooled[8]	
		Mean ± SE	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	46.3 ± 1.9 c	46.7 ± 0.8 c	44.5 – 48.4	46.6 ± 0.8 c
	F	46.1 ± 1.9 c	46.6 ± 0.8 c	44.4 – 48.8	46.5 ± 0.8 c
Rut-BF × 26NR[3]	BC	47.8 ± 1.9 b	47.6 ± 0.8 b	45.8 – 49.3	47.7 ± 0.8 b
	F	50.0 ± 1.9 a	48.8 ± 0.8 a	46.2 – 50.4	49.1 ± 0.8 a
Hi-Q[4]		47.1 ± 1.8 bc	47.0 ± 0.8 bc		47.0 ± 0.8 bc
A07-26NR[4]		49.1 ± 1.8 ab	49.3 ± 0.8 a		49.3 ± 0.8 a
Rut-BF × Hi-Q		46.2 ± 1.9 a	46.7 ± 0.8 b	44.4 – 48.8	46.5 ± 0.8 b
Rut-BF × 26NR		48.9 ± 1.9 a	48.2 ± 0.8 a	45.8 – 50.4	48.3 ± 0.8 a
BC[5]		47.1 ± 2.0 a	47.1 ± 0.8 b	44.5 – 49.3	47.1 ± 0.8 b
F[6]		48.0 ± 2.0 a	47.6 ± 0.8 a	44.4 – 50.4	47.6 ± 0.8 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the inbred lines were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents used as checks in the field trials.
[5] and [6] = BC_1- and F_2-derived inbred lines, respectively.
[7] BC_1- and F_2-derived inbred line population of the two original crosses.
[8] 2014 and 2016 data was used to calculate the pooled lsmeans values of the inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Twenty-two (9 BC_1- and 13 F_2-derived) lines of Rutabaga-BF × Hi-Q and 13 (1 BC_1- and 12 F_2-derived) lines of Rutabaga-BF × A07-26NR had higher seed oil content than their respective spring canola parent (Tables A3.12, A3.13, A3.14 and A3.15).

2.3.6 Seed protein content

The analysis of variance for seed protein content is presented in Table 2.12. Significant variation ($p < 0.001$) was found between the two crosses, populations and among the inbred lines for this trait.

Based on pooled data of 2014 and 2016, mean seed protein content of the population derived from Rutabaga-BF × Hi-Q was significantly higher than the population derived from Rutabaga-BF × A07-26NR, and the F_2-derived population, on average, had higher protein content as compared to the BC_1-derived population (Table 2.13). Among the four populations derived from the two crosses, the highest level of seed protein content was found in the F_2-derived population of Rutabaga-BF × Hi-Q (25.8 ± 1.1%), while the two populations of Rutabaga-BF × A07-26NR

Table 2.12: Analysis of variance for seed protein content of the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus*. Two (2014 and 2016) years data was used for analysis of variance.

Source	df	Error df[6]	SS	MS	F value	p value[7]
Environment[1]	6		51.61	8.6		
Replication	1		0.00	0.00		
Block	3		0.10	0.03		
Cross[2] [C]	1	932.42	266.21	266.21	222.16	$< 2.2\times10^{-16}$ ***
Population[3] [P]	1	974.84	18.36	18.37	15.33	9.7×10^{-5} ***
C × P[4]	1	962.51	55.61	55.61	46.41	1.7×10^{-11} ***
Inbred line (C × P)[5]	89	987.50	1157.73	13.01	10.86	$< 2.2\times10^{-16}$ ***
Residual	986		1079.67	1.20		
Total	1088		2629.29			

[1] Year/site of the inbred line field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the inbred lines were developed.
[3] Type of population indicates from where, the BC_1 or F_2, the inbred lines were developed.
[4] Combination of the cross and type of population.
[5] Inbred line is nested in C × P.
[6] Due to unbalanced data for seed protein content, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

had similar and lowest seed protein content (24.4 ± 1.1 and 24.2 ± 1.1%). Both F_2- and BC_1-derived populations of Rutabaga-BF × Hi-Q were statistically similar to the spring canola parent Hi-Q (2014, 2016 and pooled data); in the case of the Rutabaga-BF × A07-26NR cross, the BC_1-derived population had higher seed protein content than the spring canola parent A07-26NR (Table 2.13).

Table 2.13: Seed protein content (%) of the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus* in 2014 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Inbred line population[7]	2014	2016	Pooled[8]	
		Mean ± SE	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	24.3 ± 2.5 ab	25.9 ± 1.0 bc	23.3 – 27.2	25.1 ± 1.1 b
	F	24.9 ± 2.5 a	26.6 ± 1.0 a	24.8 – 26.7	25.8 ± 1.1 a
Rut-BF × 26NR[3]	BC	23.3 ± 2.5 bc	25.3 ± 1.0 c	22.4 – 27.1	24.4 ± 1.1 c
	F	22.4 ± 2.5 c	25.1 ± 1.0 c	23.4 – 26.1	24.2 ± 1.1 cd
Hi-Q[4]		23.9 ± 2.4 abc	26.0 ± 1.1 abc		25.1 ± 1.1 abc
A07-26NR[4]		22.8 ± 2.4 bc	24.3 ± 1.1 d		23.7 ± 1.1 d
Rut-BF × Hi-Q		24.6 ± 2.4 a	26.2 ± 1.0 a	23.3 – 27.2	25.4 ± 1.1 a
Rut-BF × 26NR		22.9 ± 2.4 a	25.2 ± 1.0 b	22.4 – 27.1	24.3 ± 1.1 b
BC[5]		23.8 ± 2.5 a	25.6 ± 1.0 b	22.4 – 27.2	24.8 ± 1.1 b
F[6]		23.7 ± 2.5 a	25.9 ± 1.0 a	23.4 – 26.7	25.1 ± 1.1 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the inbred lines were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents used as checks in the field trials.
[5] and [6] = BC_1- and F_2-derived inbred lines, respectively.
[7] BC_1- and F_2-derived inbred line population of the two original crosses.
[8] 2014 and 2016 data was used to calculate the pooled lsmeans values of the inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

As compared to the spring canola parent, 29 (9 BC_1- and 20 F_2-derived) and 28 (14 BC_1- and 14 F_2-derived) inbred lines, respectively, of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR cross showed higher seed protein content (Tables A3.12, A3.13, A3.14 and A3.15).

2.3.7 Seed glucosinolate content

The results of ANOVA for seed glucosinolate content is presented in Table 2.14. Significant

variation was found between the two crosses ($p < 0.05$) and significant variation was found among the inbred lines ($p < 0.001$) for this trait.

As compared to Rutabaga-BF × Hi-Q, the population derived from Rutabaga-BF × A07-26NR had about 0.4 µmol/g higher seed glucosinolate content (Table 2.15). The greatest difference among the four populations of the two crosses was only 1.5 µmol/g seed (15.0 ± 0.6 vs. 16.5 ± 0.6 µmol/g seed). The mean glucosinolate content of the F_2- and BC_1-derived populations was very

Table 2.14: Analysis of variance for seed glucosinolate content of the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus*. Two (2014 and 2016) years data was used for analysis of variance.

Source	df	Error df[6]	SS	MS	F value	p value[7]
Environment[1]	6		13.53	2.25		
Replication	1		0.03	0.03		
Block	3		0.14	0.05		
Cross[2] [C]	1	724.42	23.50	23.51	4.81	0.02859 *
Population[3] [P]	1	888.72	0.20	0.24	0.05	0.8241
C × P[4]	1	829.00	293.90	293.94	60.16	2.6×10^{-14} ***
Inbred line (C × P)[5]	89	986.42	5055.90	56.81	11.63	2.2×10^{-16} ***
Residual	986		4821.54	4.89		
Total	1088		10208.74			

[1] Year/site of the inbred line field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the inbred lines were developed.
[3] Type of population indicates from where, the BC_1 or F_2, the inbred lines were developed.
[4] Combination of the cross and type of population.
[5] Inbred line is nested in C × P.
[6] Due to unbalanced data for seed glucosinolate content, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

similar to the spring canola parents (14.3 ± 0.8 and 14.5 ± 0.8µmol/g seed), though in some cases these differences were found to be statistically significant. Thus, the results indicate that majority of the inbred lines derived from the two crosses had low glucosinolate content to meet the canola quality standard.

Table 2.15: Seed glucosinolate content (µmol/g seed) of the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses of *B. napus* in 2014 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Inbred line population[7]	2014	2016	Pooled[8]	
		Mean ± SE	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	15.9 ± 2.1 b	14.2 ± 0.3 d	12.4 – 23.5	15.0 ± 0.6 b
	F	18.8 ± 2.1 a	14.8 ± 0.3 bd	13.7 – 25.5	16.1 ± 0.6 a
Rut-BF × 26NR[3]	BC	18.2 ± 2.1 a	15.4 ± 0.3 a	13.4 – 24.3	16.5 ± 0.6 a
	F	15.1 ± 2.1 b	14.6 ± 0.3 cd	14.4 – 18.0	15.2 ± 0.6 b
Hi-Q[4]		13.8 ± 1.6 b	14.2 ± 0.4 d		14.3 ± 0.8 b
A07-26NR[4]		14.9 ± 1.6 b	13.9 ± 0.4 d		14.5 ± 0.8 b
Rut-BF × Hi-Q		17.3 ± 1.7 a	14.5 ± 0.2 b	12.4 – 25.5	15.5 ± 0.6 b
Rut-BF × 26NR		16.7 ± 1.7 a	15.0 ± 0.2 a	13.4 – 24.3	15.9 ± 0.6 a
BC[5]		17.1 ± 1.5 a	14.8 ± 0.2 a	12.4 – 24.3	15.6 ± 0.6 a
F[6]		16.9 ± 1.5 a	14.7 ± 0.2 a	13.7 – 25.5	15.7 ± 0.6 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the inbred lines were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents used as checks in the field trials.
[5] and [6] = BC_1- and F_2-derived inbred lines, respectively.
[7] BC_1- and F_2-derived inbred line population of the two original crosses.
[8] 2014 and 2016 data was used to calculate the pooled lsmeans values of the inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Twelve (10 BC_1- and 2 F_2-derived) lines of Rutabaga-BF × Hi-Q and 5 (4 BC_1- and 1 F_2-derived) lines of Rutabaga-BF × A07-26NR had lower content of seed glucosinolate as compared to their respective spring canola parent Hi-Q or A07-26NR (Tables A3.12, A3.13, A3.14 and A3.15).

2.3.8 Association between agronomic and seed quality traits

Days to flowering showed a significant positive correlation with days to maturity, and both traits showed a significant positive correlation with plant height in the inbred lines of the two crosses as well as combined data of the two crosses (Table 2.16). Seed yield showed a negative

correlation with days to flowering and days to maturity; however, the correlation was significant in the population derived from the Rutabaga-BF × Hi-Q cross as well as combined data of the two

Table 2.16: Correlation[1] between agronomic and seed quality traits of the inbred lines of the Rutabaga-BF × *B. napus* (Hi-Q/A07-26NR) crosses.

	Cross[2]	Days to maturity	Plant height	Seed yield	Seed oil content	Seed protein content	Seed glucosinolate content
Days to flowering	Cross 1	0.68 ***	0.45 ***	-0.53 ***	-0.64 ***	0.04	-0.07
	Cross 2	0.82 ***	0.83 ***	-0.28	-0.62 ***	0.28	0.43 **[2]
	Combined	0.84 ***	0.77 ***	-0.6 ***	-0.75 ***	0.38 ***	0.04
Days to maturity	Cross 1		0.43 **	-0.41 **	-0.3 *[2]	-0.22	0.07
	Cross 2		0.78 ***	-0.16	-0.62 ***	0.39 *	0.27
	Combined		0.8 ***	-0.52 ***	-0.67 ***	0.37 ***	0.07
Plant height	Cross 1			0.13	-0.32 *	0.05	0.3 *
	Cross 2			-0.13	-0.55 ***	0.22	0.30 *
	Combined			-0.36 ***	-0.65 ***	-0.41 ***	0.15
Seed yield	Cross 1				0.29 *	0.15	0.06
	Cross 2				0.35 *	-0.05	-0.17
	Combined				0.51 ***	-0.20 *	-0.02
Seed oil content	Cross 1					-0.48 ***	0.16
	Cross 2					-0.78 ***	-0.41 **
	Combined					-0.72 ***	-0.04
Seed protein content	Cross 1						0.01
	Cross 2						0.08
	Combined						0.01

[1] The pooled data of the 2014 and 2016 field trials was used to calculate correlation between the traits of the inbred lines of the two crosses.
[2] Cross 1 = Rutabaga-BF × Hi=Q, Cross 2 = Rutabaga-BF × A07-26NR and Combined = combined data of the two crosses.
*, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

crosses. Days to flowering, days to maturity and plant height showed a significant negative association with seed oil content, while seed yield showed a positive correlation with seed oil content (Table 2.16). This indicates that the early flowering lines tended to be shorter in height and produced higher seed yield and had greater seed oil content. Seed oil content showed a negative correlation with seed protein content in both populations (-0.48 *** and -0.78 ***) as well

as in combined data of the two crosses (-0.72 ***) (Table 2.16). No consistent relationship of seed glucosinolate content was found with other traits.

Of the five high-yielding inbred lines of the Rutabaga-BF × Hi-Q cross, CO.FF.IN.30, CO.BC.IN.18, CO.FF.IN.18, CO.FF.IN.22 and CO.FF.IN.26, the lines CO.FF.IN.26 and CO.FF.IN.22 flowered similar to Hi-Q but matured earlier than this parent, and had similar seed oil but higher seed protein content as compared to Hi-Q. In case of the two high-yielding inbred lines of the Rutabaga-BF × A07-26NR cross, RR.FF.IN.37 and RR.BC.IN.30, the line RR.FF.IN.37 took almost the same number days to flower and mature, and had plant height and seed oil content similar to A07-26NR; this line also had slightly higher seed protein content than this spring canola parent (Tables, A3.8, A3.9, A3.10, A3.11, A3.12, A3.13, A3.14 and A3.15).

2.3.9 Introgression of alleles from *Brassica napus* var. *napobrassica* (Rutabaga-BF)

A total of 455 SSR markers were tested for polymorphisms between Rutabaga-BF and Hi-Q, of which 333 (73.2%) were found to be polymorphic between the two parents. Of the total number of polymorphic SSR markers, 87 from the A and C genome were used to genotype the 51 inbred lines derived from the Rutabaga-BF × Hi-Q cross; these markers amplified a total of 217 loci. Thus, the average number of loci per SSR marker was 2.49. Chromosome A2 with 3.17 loci per SSR marker showed the greatest level of polymorphism. Of the 217 loci, 89 (41%) were found to carry Rutabaga alleles, i.e. these alleles were absent in Hi-Q but present in Rutabaga-BF. Thus, the average number of loci carrying Rutabaga-BF alleles could be detected by a SSR maker in this population was 1.02. The chromosomes A2, A4, A5 and A10 carried more than 1.0 loci per SSR markers where Rutabaga-BF-specific alleles could be detected (Table 2.17).

In case of Rutabaga-BF × A07-26NR, 455 SSR markers were tested for polymorphism where 321 (70.6%) were polymorphic between the parents Rutabaga-BF and A07-26NR. Of these polymorphic markers, 105 markers from the A and C genome amplified a total 301 loci in the inbred

Table 2.17: Occurrence of polymorphic loci carrying alleles specific to Rutabaga-BF in advanced generation inbred lines derived from the Rutabaga-BF × Hi-Q cross of *B. napus* detected using 87 SSR markers from A and C genome linkage groups.

Linkage group	No. of SSR markers	Total no. loci[1]	No. loci/SSR marker	No. loci carrying Rutabaga-BF alleles[2]	No. of loci with Rutabaga-BF alleles/SSR markers
A1	6	15	2.5	5	0.83
A2	6	19	3.17	7	1.17
A3	6	14	2.33	6	1.00
A4	7	21	3.00	8	1.14
A5	5	14	2.80	6	1.20
A6	7	17	2.43	7	1.00
A7	7	19	2.71	6	0.86
A8	8	18	2.25	8	1.00
A9	7	16	2.29	7	1.00
A10	6	18	3.00	7	1.17
C1	4	10	2.50	4	1.00
C2	2	4	2.00	2	1.00
C3	3	6	2.00	3	1.00
C5	4	8	2.00	4	1.00
C6	3	6	2.00	3	1.00
C7	1	2	2.00	1	1.00
C8	3	6	2.00	3	1.00
C9	2	4	2.00	2	1.00
Total	87	217	2.49	89	1.02

[1] Total number of loci detected in *B. napus* (Hi-Q) and Rutabaga-BF parents.
[2] Advanced generation F_2- and BC_1-derived lines of the Rutabaga-BF × Hi-Q cross.

line population derived from the Rutabaga-BF × A07-26NR cross; thus, the average number of loci per SSR marker was 2.87. Of the 301 loci, 129 (43%) were found to carry alleles specific to Rutabaga-BF; thus, on average, 1.23 loci carrying Rutabaga-BF alleles per SSR marker were identified in the inbred line population by using the 105 SSR markers. The greatest level of polymorphism was detected for chromosome A1 with 4.40 loci per SSR marker; the chromosomes

A1, A2, C1 and C8 carried more than 1.4 loci per SSR marker where Rutabaga-BF-specific alleles could be detected (Table 2.18).

Table 2.18: Occurrence of polymorphic loci carrying alleles specific to Rutabaga-BF in advanced generation inbred lines derived from the Rutabaga-BF × A07-26NR cross of *B. napus* detected using 105 SSR markers from A and C genome linkage groups.

Linkage group	No. of SSR markers	Total no. loci[1]	No. loci/SSR marker	No. loci carrying Rutabaga-BF alleles[2]	No. loci with Rutabaga-BF alleles/SSR markers
A1	5	22	4.40	7	1.40
A2	6	16	2.67	9	1.50
A3	6	16	2.67	6	1.00
A4	6	17	2.83	8	1.33
A5	6	16	2.67	5	0.83
A6	6	17	2.83	7	1.17
A7	6	19	3.17	7	1.17
A8	6	14	2.33	7	1.17
A9	6	16	2.67	7	1.17
A10	5	10	2.00	5	1.00
C1	6	24	4.00	11	1.83
C2	5	16	3.20	6	1.20
C3	6	15	2.50	8	1.33
C4	5	12	2.40	5	1.00
C5	6	16	2.67	8	1.33
C6	6	19	3.17	7	1.17
C7	3	8	2.67	3	1.00
C8	4	11	2.75	7	1.75
C9	6	17	2.83	6	1.00
	105	301	2.87	129	1.23

[1] Total number of loci detected in *B. napus* (A07-26NR) and Rutabaga-BF parents.
[2] Advanced generation F_2- and BC_1-derived lines of the Rutabaga-BF × A07-26NR cross.

On a population basis, no significant difference ($p > 0.05$) was found between the inbred lines derived from BC_1- and F_2- of Rutabaga-BF × Hi-Q for the number of loci carrying alleles introgressed from Rutabaga-BF (27.8 ± 1.8 vs. 29.5 ± 1.7, respectively). On average, these two populations carried 31.2 ± 2.0 and 33.2 ± 1.9% of the total number of loci with Rutabaga-BF alleles amplified by the 87 SSR markers. The extent of variation for number of loci carrying Rutabaga-

BF alleles was also very similar within these two populations - 8 to 38 and 7 to 44 per line in the BC_1- and F_2-derived populations, respectively (Table 2.19).

In case of the Rutabaga-BF × A07-26NR cross, the average number of loci carrying Rutabaga-BF alleles detected in the BC_1-derived population was 31.5 ± 2.9; this is almost 3-times greater than the number of loci carrying Rutabaga-BF alleles detected in the F_2-derived population (12.3 ± 2.9). The difference between the two populations for the occurrence of the number of loci with Rutabaga-BF alleles was statistically significant ($p < 0.05$). On average, 24.4 ± 2.3 and 9.6 ± 2.3% of the total number of loci carrying Rutabaga-BF alleles were detected in the BC_1- and F_2-derived lines. The extent of variation for the number of loci carrying Rutabaga-BF alleles in the BC_1-derived lines was 18 to 42, whereas this varied from 2 to 62 in the F_2-derived lines (Table 2.19).

Table 2.19: Occurrence of the number of loci carrying Rutabaga-BF alleles in the BC_1- and F_2-derived lines of the two Rutabaga-BF × *B. napus* (Hi-Q/A07-26NR) crosses.

Cross	Advanced generation population	Total loci with Rutabaga-BF alleles expected	No. inbred line	No. loci with Rutabaga-BF alleles/ inbred line		% loci with Rutabaga-BF alleles/inbred line	
				Range	Mean ± SE	Range	Mean ± SE
Rutabaga-BF × Hi-Q	BC_1-derived	89	25	8-38	27.8 ± 1.8	9.0-42.7	31.2 ± 2.0
	F_2-derived	89	26	7-44	29.5 ± 1.7	7.9-49.4	33.2 ± 1.9
Rutabaga-BF × A07-26NR	BC_1-derived	129	21	18-42	31.5 ± 2.9	14.0-32.6	24.4 ± 2.3
	F_2-derived	129	21	2-62	12.3 ± 2.9	1.6-48.1	9.6 ± 2.3

2.3.10 Genetic diversity

Analysis of molecular variance (AMOVA) showed that the inbred lines of the two crosses accounted for 80.61% of the total genetic variance, while variation among the four populations of the two crosses accounted for only 19.39% of the total genetic variance (Table 2.20). A similar

extent of genetic variability within the whole inbred line population and among the four populations of the two crosses was also estimated based on Shannon's diversity index (77 and 23%, respectively) (Figure 2.5). Genetic variations among the four populations, however, was significant (Table 2.20).

Table 2.20: Analysis of molecular variance (AMOVA) of the advanced generation inbred lines derived from the Rutabaga-BF × *B. napus* (Hi-Q/A07-26NR) crosses.

Source of variation[1]	df	SS	MS	Estimated variance	Variance (%)	F_{st}	p-value
Among populations	3	179.18	59.73	2.25	19.39	0.194	0.001
Within populations	86	804.26	9.35	9.35	80.61		
Total	89	983.43		11.6	100		

[1] Among populations = between the four inbred line populations derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and within populations = between the inbred lines of the four populations.

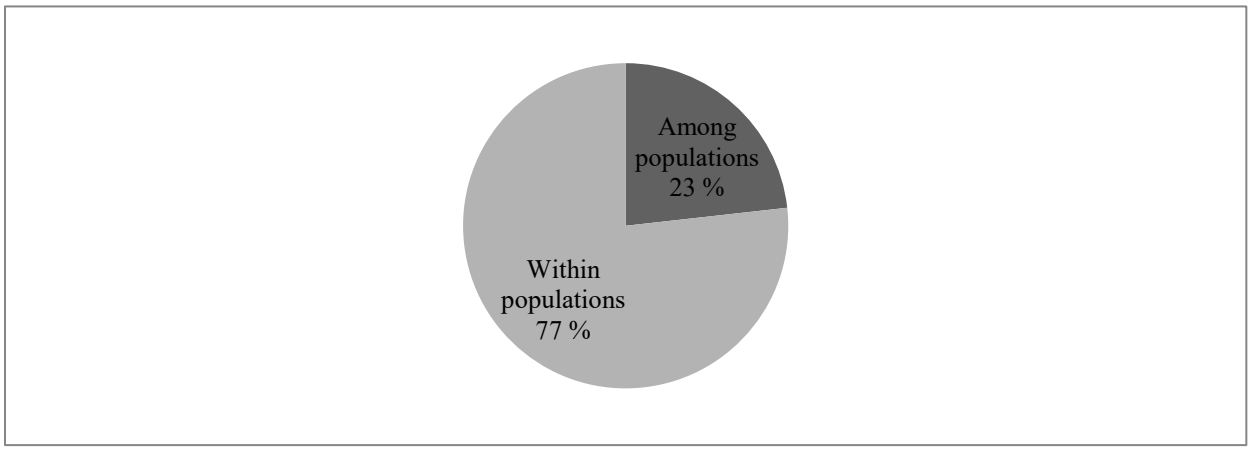

Figure 2.5: Shannon estimates of genetic diversity among and within the populations derived from the Rutabaga-BF × *B. napus* (Hi-Q/A07-26NR) crosses. Among and within populations show the proportion of the whole genetic variation observed between, respectively, the four populations derived from the two crosses, and the inbred lines of these populations.

Genetic variability among the inbred lines derived from the two Rutabaga-BF × *B. napus* crosses was visualized by a dendrogram developed through cluster analysis following an unweighted pair-group method with arithmetic mean (UPGMA). Several inbred lines of the two

crosses exhibited wide genetic dissimilarity to their parents Rutabaga-BF and *B. napus*. Average similarity coefficient of the inbred population of Rutabaga-BF × Hi-Q with the parents Rutabaga-BF and Hi-Q was 0.425 and 0.525, respectively, while it was 0.367 and 0.573, respectively, with Rutabaga-BF and A07-26NR for the inbred lines of the Rutabaga-BF × A07-26NR cross. The average genetic divergence of the inbred population of Rutabaga-BF × A07-26NR (coefficient 0.367) from the common parent Rutabaga-BF was greater than that of the inbred population of Rutabaga-BF × Hi-Q (coefficient 0.425); this indicates that the inbred lines of Rutabaga-BF × A07-26NR showed greater genetic similarity with the *B. napus* canola parent (A07-26NR) as compared to the inbred lines of the Rutabaga-BF × Hi-Q cross (Figures 2.6 and 2.7).

The inbred lines derived from the Rutabaga-BF × Hi-Q cross fell into three genetically distinct groups with similarity coefficient of 0.525 (Figures 2.4). Among these, the Group I was closest to the *B. napus* parent (Hi-Q), and exclusively included the F_2-derived inbred lines. The inbred lines derived from BC_1 were mostly included in Group II. The number of inbred lines in Group III was less than that of Group I and II; only one BC_1-derived line (CO.BC.IN.18) was included in this group (Figure 2.6).

In case of the Rutabaga-BF × A07-26NR cross, the inbred lines formed two distinct groups with a genetic similarity coefficient of 0.573. The Group II included only four lines – all derived from F_2 (RR.FF.IN.17, RR.FF.IN.35, RR.FF.IN.13 and RR.FF.IN.18). In case of Group I, several sub-groups were found where one group was closest to A07-26NR and included the F_2-derived inbred lines. Some of the sub-groups included only the BC_1-derived inbred lines (Figure 2.7).

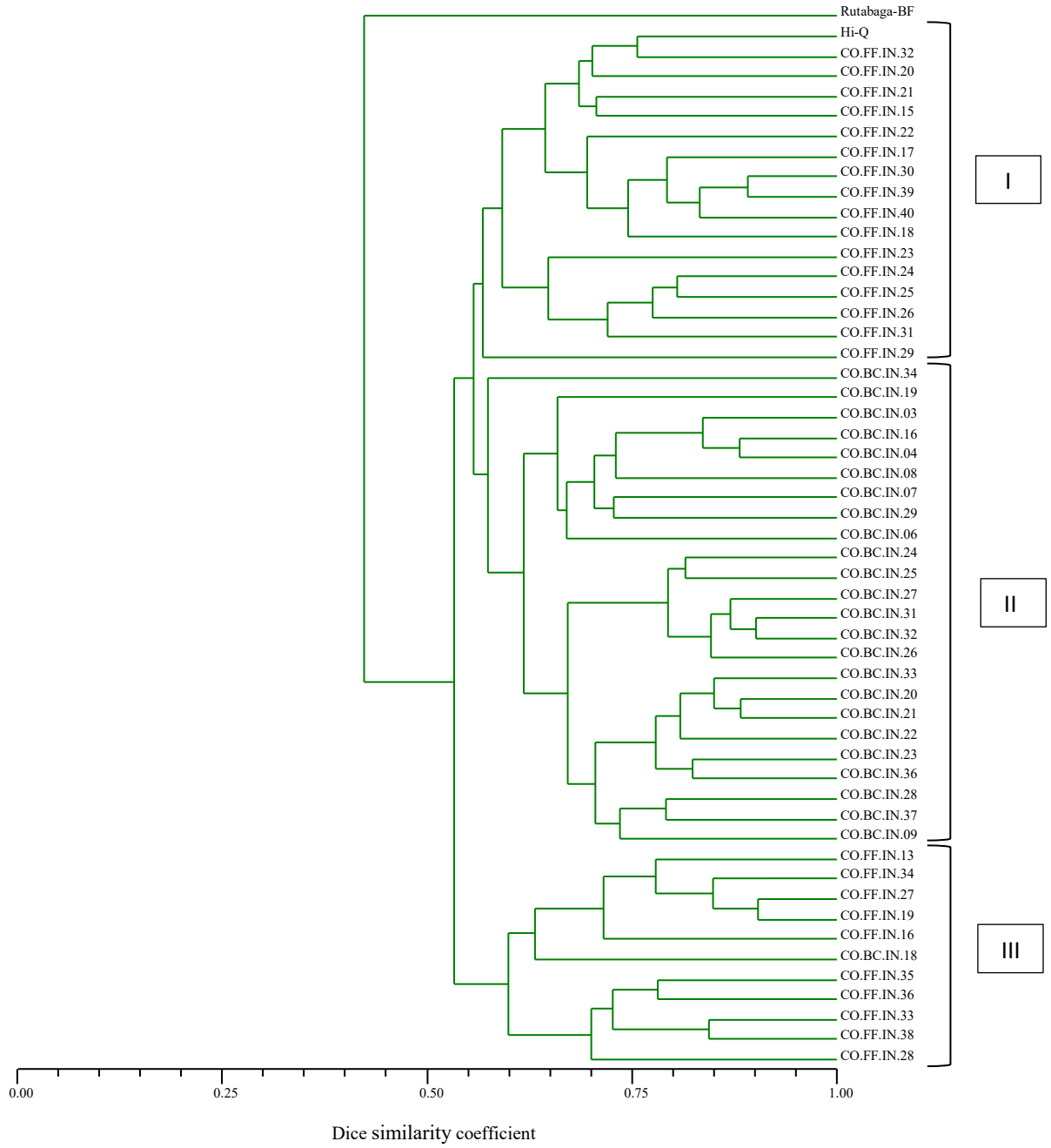

Figure 2.6: Dendrogram showing genetic similarity of the 25 BC_1- and 26 F_2-derived inbred lines of the Rutabaga-BF × Hi-Q cross detected by 87 polymorphic SSR markers through unweighted pair-group method with arithmetic mean (UPGMA).

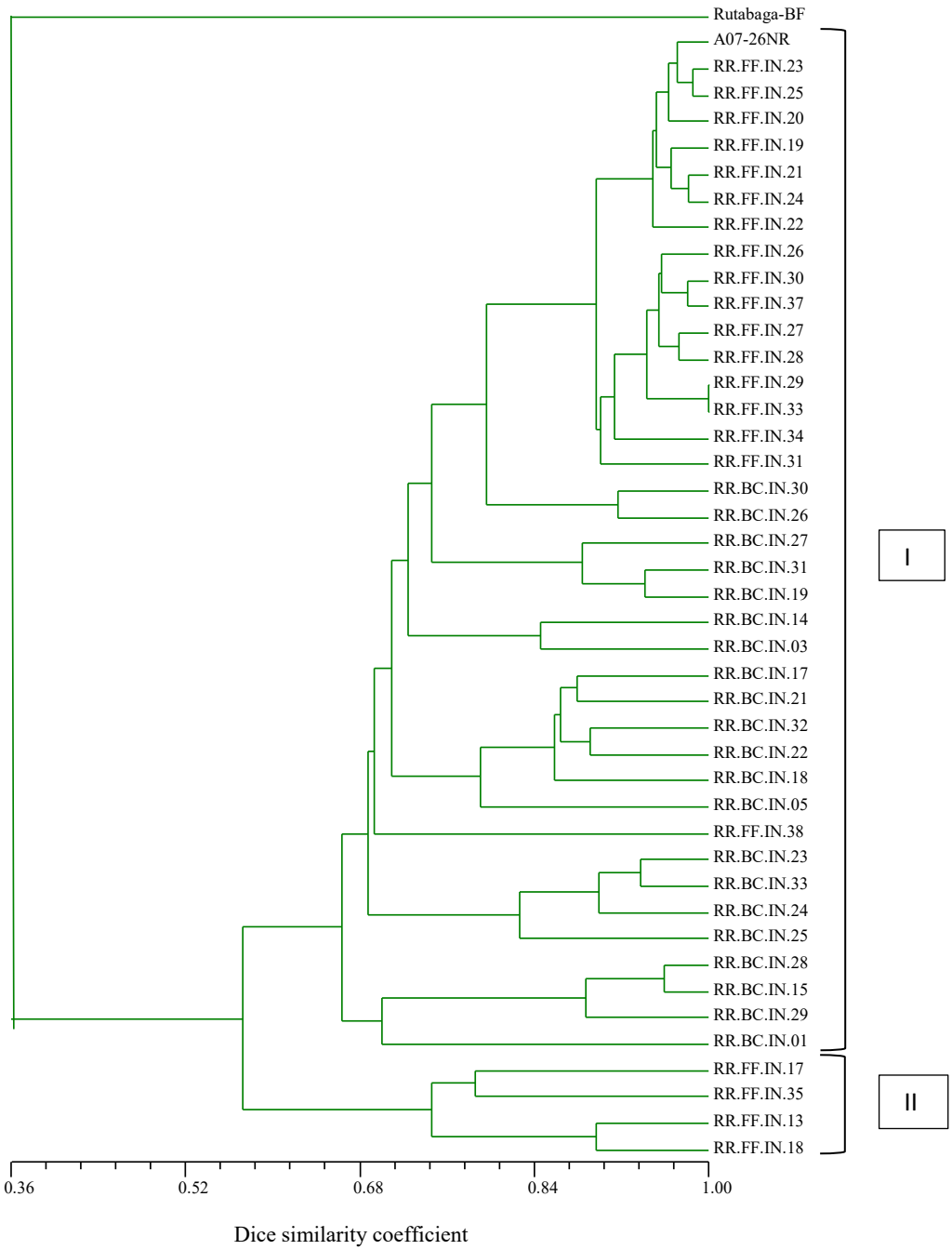

Figure 2.7: Dendrogram showing genetic similarity of the 21 BC_1- and 21 F_2-derived inbred lines of the Rutabaga-BF × A07-26NR cross detected by 105 polymorphic SSR markers through unweighted pair-group method with arithmetic mean (UPGMA).

2.4 Discussion

The transformation of traditional rapeseed to canola type as well as intensive breeding within a restricted gene pool over the past few decades has reduced genetic diversity in *B. napus* canola (Cowling, 2007; Fu and Gugel, 2010; Bus et al., 2011). Success on further improvement of spring *B. napus* canola, therefore, relies on widening the genetic base of the breeding population of this crop (for review, see Rahman, 2013).

In the present study, the use of Rutabaga, which is known to be genetically distinct from other types of *B. napus* (Bus et al., 2011), in the breeding of spring canola revealed that the unique alleles of this variant were introgressed into both A and C genomes of the spring canola inbred lines (Tables 2.17 and 2.18). This resulted in wide genetic dissimilarity among the inbred lines derived from both BC_1 or F_2, suggesting that each inbred line carries unique alleles of the Rutabaga gene pool. The importance of the primary gene pool of *B. napus* (Diers and Osborn, 1994; Bus et al., 2011) for use in broadening the genetic base of spring *B. napus* canola has been well-documented. For example, the European winter and Chinese semi-winter types of *B. napus* have been used by several researchers to broaden the genetic base of spring *B. napus* canola as well as to increase the productivity of this crop (Quijada et al., 2004; Qian et al., 2007; Rahman and Kebede, 2012; Rahman, 2017).

Majority of the inbred lines derived from Rutabaga × spring canola were late flowering; however, some of the inbred lines flowered earlier than the spring canola parent. Flowering time in *B. napus* is a polygenic trait; several regions of the A and C genome chromosomes are found to be involved in the control of this trait (Ferreira et al., 1995; Schiessl et al., 2014; Rahman et al., 2017). Rutabaga is primarily bred for tuberous root where earliness of flowering is not a desired

trait; therefore, introduction late flowering alleles in the progeny derived from the Rutabaga × spring canola crosses was not surprising. In contrast, the occurrence of early flowering inbred lines from these crosses indicates that this variant of *B. napus* also carries alleles which can promote flowering. Similarly, Rahman et al. (2011) reported that the late flowering species *B. oleracea* carries alleles which can improve the earliness of flowering in *B. napus*.

Similar variation for days to maturity was also found in the inbred line population derived from the Rutabaga × spring *B. napus* crosses. While more than 60% of the inbred lines took longer time to mature than the spring canola parents, about 20-30% of the inbred lines of both BC_1- and F_2-derived populations matured similar to or earlier than the spring *B. napus* parent. The significant positive correlation ($r = 0.68$ to 0.82; $R^2 = 0.46$ to 0.67) found between days to flowering and days to maturity in the present study suggests that earliness of flowering can be used as an indicator of earliness of maturity in a breeding program. Positive association of these two traits has also been reported by Miller (2001) in spring *B. napus* canola and by Honsdorf et al. (2010) in winter *B. napus* canola. Plant height also showed a positive correlation with day to flowering ($r = 0.45$ to 0.83; $R^2 = 0.20$ to 0.69) and with days to maturity ($r = 0.43$ to 0.78; $R^2 = 0.19$ to 0.61) in the inbred lines derived from the Rutabaga × spring canola crosses. Significant correlation between days to flowering and plant height has also been reported by Quijada et al. (2006) and Udall et al. (2006) in case of the inbred lines derived from spring × winter *B. napus*, winter × spring *B. napus* and resynthesized × spring *B. napus* crosses. Thus, greater vegetative growth and taller plants seems to be a general feature of the late-flowering inbred lines.

More than 90% of the inbred lines derived from the Rutabaga × spring canola crosses gave lower seed yield than their spring canola parent; however, few (5+2 = 7 lines of the two crosses)

gave higher seed yield as compared to the spring canola parent. Quijada et al. (2006) also found low seed yield in a majority of the spring *B. napus* lines derived from European winter × spring and Chinese semi-winter × spring *B. napus* crosses. Seed yield in *B. napus* is controlled by several QTL from both the A and C genomes (Quijada et al., 2006; Udall et al., 2006; Radoev et al., 2008; Rahman et al., 2017); therefore, variation for seed yield observed in the inbred lines derived from the Rutabaga × spring canola crosses apparently resulted from variable combination of the Rutabaga and spring canola alleles generated through segregation of these two types of alleles. The higher seed yield in the few inbred lines, apparently, resulted from favorable combination of Rutabaga and spring canola alleles. This agrees with Rahman (2017) that exotic alleles of the primary gene pool can be used to increase seed yield in spring canola; however, undesired alleles often get introduced into the breeding population (Quijada et al., 2006; Udall et al., 2006; Kebede et al., 2010) which would need repeated cycle of breeding for removal.

In the present study, a strong negative correlation was found between seed yield and flowering time. Similar results have also been reported by Udall et al. (2006) and Raman et al. (2016). Such correlation can result, at least partly, from pleiotropic effects of the same or closely linked QTL governing flowering time and seed yield as suggested by Udall et al. (2006) and Raman et al. (2016) while working with doubled haploid populations derived from winter × spring *B. napus*, resynthesized × spring *B. napus*, and spring × spring *B. napus* crosses. However, correlation between theses two traits was not statistically significant in case of the inbred lines derived from the Rutabaga-BF × A07-26NR cross; this is likely due to the lower number of Rutabaga alleles introgressed into this population as compared to the population derived from the Rutabaga-BF × Hi-Q cross (Table 2.19).

Analysis of seed quality traits revealed that about 30 - 40% of the inbred lines surpassed the spring canola parent for seed oil content. Oil content is a quantitative traits controlled by a large number of loci (for review, see Rahman et al., 2013). Zhao et al. (2005) and Chen et al. (2010) reported that favorable QTL alleles for this trait can be found in both European and Chinese *B. napus* and accumulation of these alleles can increase oil content in *B. napus*. Thus, the results from this study suggests that Rutabaga also carries alleles which can increase oil content in spring canola. Seed protein content in *B. napus* is also a polygenic trait (Zhao et al., 2006); this trait generally shows strong negative correlation with seed oil content (Grami et al., 1977). As expected, a negative correlation ($r = -0.48$ to -0.78; $R^2 = 0.23$ to 0.61) between these two traits was also found in this study. The association between oil and protein content can be due to pleiotropic effects of the majority of the QTL governing these two traits or location of the QTL for these traits in the same genomic region (Zhao et al., 2006). Some of the high-yielding inbred lines, such as CO.FF.IN.30, CO.FF.IN.18 and CO.FF.IN.26, however, showed early-flowering and maturity properties along with 0.2-1.2% increase in oil content and 0.4-1.3% increase in protein content when compared with the spring canola parent. Increase in both oil and protein contents in these lines might have resulted from accumulation of protein QTL alleles, which are independent of oil QTL alleles. This type of protein QTL has also been reported by Zhao et al. (2006) in a doubled haploid population derived from a German × Chines *B. napus* cross.

Theoretically, it was expected that the BC_1-derived population would carry fewer number of Rutabaga alleles; however, in practice, a greater number of Rutabaga alleles was found in the BC_1-derived population as compared to the F_2-derived population. This might have resulted from the greater selection pressure applied to the F_2 and subsequent generation populations of the two crosses to develop canola quality inbred lines. Theoretically, relatively lower selection pressure

would be needed to develop a canola quality line from the BC_1-derived population where backcrossing to the *B. napus* canola expected to increase the probability of the occurrence of a canola quality line. When comparing these two populations, the F_2-derived lines performed better than the BC_1-derived lines for days to flowering (51.3 ± 2.4 vs. 52.1± 2.4 days), days to maturity (101.8 ± 4.2 vs. 102.5 ± 4.2 days), seed yield (3279.8 ± 270.5 vs. 3053.2 ± 270.4 kg/ha), seed oil content (47.6 ± 0.8 vs. 47.1 ± 0.8 %) and seed protein content (25.1 ± 1.1 vs. 24.8 ± 1.1 %). This apparently resulted from the introduction of greater number of undesirable Rutabaga alleles in the BC_1-derived population due to low selection pressure. This is also evident from the fact that the 47 F_2-derived lines descended from 10 F_3 plants, while the 46 BC_1-derived inbred lines descended from 18 BC_1 plants.

In conclusion, this study showed that the alleles of Rutabaga can be used for broadening the genetic base of spring *B. napus* canola, and favorable alleles for different agronomic and seed quality traits including seed yield and oil content can be found in this variant for increasing the productivity of spring canola. Indeed, a few inbred lines showed superiority for the major agronomic and seed quality traits, such as days to flowering and maturity, seed yield, and seed oil and protein content, over the *B. napus* parent. These inbred lines, therefore, can be used in the breeding programs for the improvement of the spring *B. napus* canola.

Chapter 3

Evaluation of the inbred lines derived from Rutabaga (*Brassica napus* var. *napobrassica*) × spring *Brassica napus* canola for heterosis

3.1 Introduction

Increasing the level of heterosis and productivity of the hybrid cultivars plays an important role in crop production in the world (for review, see Schnable and Springer, 2013). Using hybrid cultivars, the productivity and profitability of major field crops, such as maize, rice and sunflower has been increased substantially at the farm level (for review, see Groszmann et al., 2013). The development of male sterility, for use as a pollination control mechanism in *B. napus* in the 1970's (Stefansson and Downey, 1995), and the findings of the high level of heterosis in F_1 progeny derived from cross between *B. napus* parents (Lefort-Buson and Dattee, 1982; Grant and Beversdorf, 1985; Lefort-Buson et al., 1987a) stimulated canola breeders to exploit the phenomenon known as heterosis and the development of hybrid cultivars in this crop. The first commercial hybrid *B. napus* cultivar, Hyola 40, was released in Canada in 1989 (Canola Council of Canada, 2016e). The greater yields in hybrid compared to open-pollinated cultivars is the primary reason for popularity of this type of cultivar. Currently, hybrid cultivars are grown on more than 95% of the canola acreage in Canada; about 25% of the increase in production of this crop in the recent years in this country is attributed to the use of hybrid cultivars (McVetty et al., 2016).

Many hypotheses have been proposed to explain the genetic basis of heterosis; however, very little is known about the molecular mechanisms that result in the superiority of the hybrid over its parents (Crow, 1999; Goodnight, 1999; for review, see Baranwal et al., 2012; Ryder et al., 2014).

The importance of genetic divergence between the parents for heterosis has been well-documented in maize (Melchinger and Gumber, 1998; for review, see Baranwal et al., 2012 and Schnable and Springer, 2013), and the same has also been found in *B. napus* canola (Lefort-Buston et al., 1987b; Ali et al., 1995; Ahmad et al., 2011; for review, see Rahman, 2013). However, the narrow genetic diversity reported in *B. napus* canola (for review, see Rahman, 2013) is one of the major constrains for continued improvement of the hybrid cultivars; broadening of allelic diversity in the breeding population of this crop is, therefore, important (Gehringer et al., 2007).

Spring *B. napus* carrying diverse alleles, introgressed from genetically distinct germplasms of its primary gene pool, has demonstrated higher levels of heterosis in hybrids (for review, see Rahman, 2013). For example, Quijada et al. (2004) reported that spring *B. napus* hybrids developed from crossings of spring *B. napus* lines and inbred lines carrying genome content of French winter *B. napus* surpassed seed yields of commercial hybrid cultivars. Likewise, Rahman et al. (2016) found that the test hybrids of inbred lines derived from spring × winter crosses exhibited almost 3.5 times greater heterosis as compared to the level of heterosis found in test hybrids developed by use of lines derived from spring × spring *B. napus* cross (12.2% vs. 3.5%). *B. napus* lines carrying genome contents introgressed from Chinese semi-winter type *B. napus* also formed a strong heterotic group; the test hybrids, produced by crossing of these lines to a Canadian spring *B. napus* canola, exceeded the seed yield of the commercial hybrids (Udall et al., 2004). However, no research has been conducted so far to assess the potential of heterosis of the spring *B. napus* canola inbred lines carrying genome contents of *B. napus* var. *napobrassica* (Rutabaga) – despite this gene pool being genetically distinct from spring and winter types (Bus et al., 2011).

The purpose of this research was to evaluate the performance of the spring *B. napus*

canola inbred lines carrying genome contents of Rutabaga for heterosis for different agronomic and seed quality traits including seed yield.

3.2 Materials and methods

3.2.1 Production of test hybrids based on the F_2- and BC_1-derived inbred lines

For this study, test hybrid (TH) seed was produced by crossing the parental spring *B. napus* canola cultivar/line to the advanced generation inbred lines derived from Rutabaga-BF × spring *B. napus*. I received seeds of 26 F_9 and 25 BC_1F_9 lines of Rutabaga-BF × Hi-Q and 21 F_9 and 21 BC_1F_7 lines of Rutabaga-BF × A07-26NR, and their 93 test hybrids from the Canola Program of the University of Alberta for field trial in 2015 (Table 3.1). I produced the same set of the test hybrid seeds in 2015-16 winter for field trial in the summer of 2016. To produce test hybrid seeds, the 93 inbred lines and the two maternal parents of the test hybrids, Hi-Q and A07-26NR, were grown in a greenhouse (21°/18° ± 2°C day/night) of the University of Alberta. The inbred lines of Rutabaga-BF × Hi-Q were crossed as male to Hi-Q while the inbred lines of Rutabaga-BF × A07-26NR were crossed as male to A07-26NR. For this, the female plants were manually emasculated and cross-pollinated with fresh pollen from the male plants. Paper envelope was used to cover the pollinated buds to avoid any other cross-pollination. The plants of the inbred lines were also self-pollinated by bag isolation to produce seeds of the paternal parents of the test hybrids for the trials.

3.2.2 Field trails

I evaluated the 93 test hybrids and their parents (the inbred lines and spring *B. napus* canola, Hi-Q or A07-26NR) in field trials at the Edmonton Research Station (ERS) (seeds produced by the Canola Program) in summer 2015 and at St. Albert Research Farm (seeds produced in 2015-16 winter) of the University of Alberta in summer 2016. The design of the field trial was an

Table 3.1: List of the test hybrids produced by use of the advanced generation lines derived from F_2 and BC_1 of the Rutabaga-BF × spring *B. napus* canola crosses.

Cross	Inbred line[1]		No. inbred lines	Test hybrid[2]			
	Population	Generation		Maternal parent	Paternal parent (inbred line)	Population	No. test hybrid
Rutabaga-BF × Hi-Q	F_2-derived	IN-F_9	26	Hi-Q	F_9	TH-F_9	26
	BC_1-derived	IN-BC_1F_9	25		BC_1F_9	TH-BC_1F_9	25
Rutabaga-BF × A07-26NR	F_2-derived	IN-F_9	21	A07-26NR	F_9	TH-F_9	21
	BC_1-derived	IN-BC_1F_7	21		BC_1F_7	TH-BC_1F_7	21
Total			93				93

[1] Advanced generation inbred lines (IN) of the F_2- or BC_1-derived population developed from either Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR. For example, IN-F_9 shows the ninth generation of the inbred line derived from the F_2 population of the Rutabaga-BF × Hi-Q cross, and IN-BC_1F_7 shows the seventh generation of the inbred line derived from the BC_1 population of the Rutabaga-BF × A07-26NR cross.

[2] Test hybrid (TH) seeds were produced by crossing the parental spring *B. napus* canola cultivar/line (Hi-Q or A07-26NR) as female to the inbred lines as male. For example, TH-F_9 is the test hybrid produced by crossing either Hi-Q or A07-26NR to the ninth generation inbreed line derived from the F_2 of Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR.

incomplete block with two replications. To achieve a high level of accuracy in the measurement of heterosis, the test hybrids and their respective parent lines (Table 3.1) were grown side-by-side where the test hybrid plot was always being placed in between the two parents (Figure 3.1). Thus, the test hybrid and its two parents constituted an experimental unit (triplet). This arrangement also facilitated visual comparison and scoring of different traits of the test hybrids and their respective parents. The triplets of Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR were randomized and assigned to the experimental plots in the two separate sets of incomplete blocks within each replication using the software program CropStat 7.2 (International Rice Research Institute, Los Baños, Philippines).

In 2015, the field trial was seeded by hand. Each plot was 1.2 m long and 1 m wide (1.2 m^2) and consisted of three planting rows with 25 cm space between the rows. Of the three rows, the middle row was seeded with 132 seeds, while the other two rows were seeded with 60 seed per row.

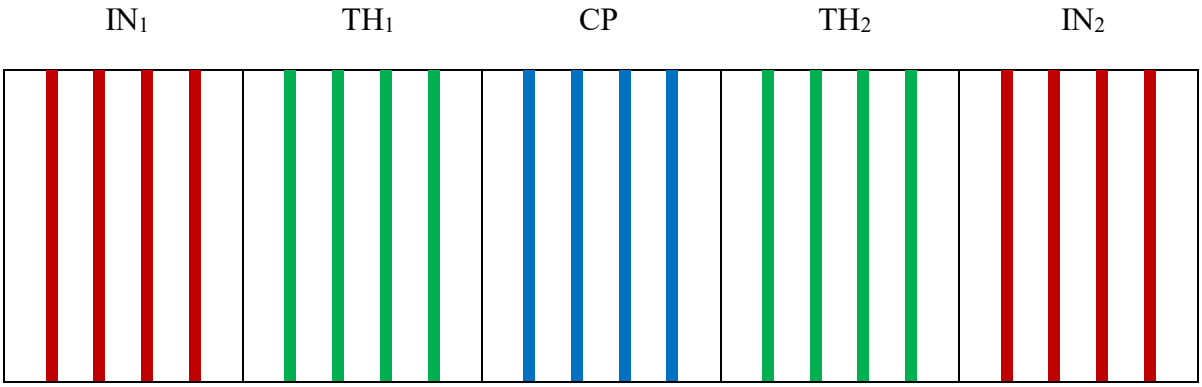

Figure 3.1: Field layout of test hybrids and their respective parents. IN = inbred line, CP = common spring canola parent (Hi-Q or A07-26NR), and TH = test hybrid of Hi-Q × IN or A07-26NR × IN.

The seeds were seeded in 22 spots, maintaining equal space between the spots. To maintain plant density of 75 plants per square meter, thinning was done at 4-leaf stage and 20-22 plants (one in each spot) were retained in the middle row. In 2016, seeding was done with a plot seeder. Each plot was 2 m long and 1.3 m wide (2.6 m^2) and consisted of four rows with 25 cm space between the rows. To achieve the same plant density as in 2015 trials, 1.3 g seed per plot was used.

3.2.3 Data collection

The following agronomic traits were recorded:

1. Plant vigor, was recorded on a 0 - 9 scale, where '9' is very vigorous and '0' is very poor.

2. Days to flowering, was recorded when approximately 50% plants in the plot had at least one open flower.

3. Plant height (cm), was measured at the end of flowering.

4. Days to maturity, was recorded when almost 50% of seeds on the main stem of the plants began to turn to brown or black.

5. Lodging, was recorded on a 0 - 9 scale, where '0' is very stiff and completely standing, and '9' is totally lodged.

6. Yield, was measured on the whole plot basis and data was converted to kg/ha.

Seed oil (%), protein (%) and glucosinolate (µmol/g seed) contents were measured by near infrared spectroscopy (NIRS) (Foss NIR system, model 6500, Eden Prairie, Minnesota).

The 93 test hybrids and their parents, with the same experimental specifications as in 2015 and 2016, were also evaluated in a field trial in 2014 at the Edmonton research station by the Canola Program of the University of Alberta. In this case, plot size was 2 m long 1-row. I received agronomic and seed quality data from this trial and incorporated in statistical analysis.

3.2.4 Data analysis

Data from 2014, 2015 and 2016 field trials were organized using Microsoft Excel. A mixed-effects model was used for statistical analysis of data using the software program 'R' (R Core Team, 2014), version 3.2.2, where environment (year/site), replication and block were considered as random-effects, and the cross, population, and test hybrid were considered as fixed-effects factors. Using lmer function from the lme4 package (Lenth, 2015), a linear mixed-effects model (both random- and fixed-effects factors were incorporated) was fitted for each agronomic and seed quality trait of the test hybrids as follows:

Test hybrid.lmer = lmer (Trait ~ Cross + Population + Cross : Population/Test hybrid
+ (1|Environment) + (1|Replication) + (1|Block), Test hybrid.data)

Analysis of variance was done for each trait to test for statistical significance of the fixed-effects variables. Mean values of the agronomic and seed quality traits of the fixed-effects variables were compared using the Tukey's honest significant test ($a \leq 0.05$). Least-squares mean values of the fixed-effects terms (cross, population, and test hybrid) for all traits were calculated using lsmeans function in the lsmeans package (Bates et al., 2015).

Mid-parent heterosis (MPH) was calculated using the formula ((test hybrid – ((inbred line + *B. napus* parent)/2))/ ((inbred line + *B. napus* parent)/2)) × 100 for all agronomic and seed quality traits. Heterosis over the check *B. napus* parent (CPH) was estimated by use of the formula ((test hybrid – *B. napus* parent)/ *B. napus* parent) × 100. As in the statistical procedure followed for evaluation of the test hybrids, two identical linear mixed-effects models were fitted for MPH and CPH values of the test hybrids for the agronomic and seed quality traits as follows:

MPH.lmer = lmer (Trait.MPH ~ Cross + Population + Cross : Population/Test hybrid
+ (1|Environment) + (1|Replication) + (1|Block), MPH.data)

CPH.lmer = lmer (Trait.MPH ~ Cross + Population + Cross : Population/Test hybrid
+ (1|Environment) + (1|Replication) + (1|Block), CPH.data)

These models were then used for analysis of variance (ANOVA) of the MPH and CPH values of all traits. Least-squares means for the fixed-effects factors such as cross, population and test hybrid was calculated for MPH and CPH and comparison of the MPH and CPH means for these fixed-effects terms was done by Tukey test ($a \leq 0.05$).

3.3 Results

3.3.1 Days to flowering

Analysis of variance for days to flowering of test hybrids of the inbred lines of the two crosses and the two types of population (F_2- and BC_1-derived), and mid-parent heterosis (MPH) and heterosis over the *B. napus* parent (Hi-Q or A07-26NR) (CPH) for this trait is presented in Table 3.2. Significant variation between test hybrids of the two crosses as well as within the whole test hybrid population was found for this trait; however, variation between the test hybrids of the two types of population was not significant. In case of MPH and CPH, variation between the two crosses was not significant; however, significant variation between the two types of population

was found for MPH only. Variation within the whole population was also significant for MPH as well as CPH for days to flowering.

Comparison of mean data of the test hybrids, and MPH and CPH of the test hybrids for days to flowering showed that the four test hybrid populations derived from the two Rutabaga-BF × spring canola crosses are statistically similar (Table 3.3). The test hybrid population of the Rutabaga-BF × Hi-Q cross flowered significantly later than the test hybrid population of the Rutabaga-BF × A07-26NR cross (49.5 ± 3.7 vs. 48.7 ± 3.7 days); however, no significant difference between the two crosses was found for MPH and CPH. All four test hybrid populations showed negative MPH and CPH for this trait. When comparing the two types of population, greater level of negative MPH observed in the test hybrids of the BC_1-derived lines than test hybrids of the F_2-derived lines (-2.7 ± 1.1 vs. -1.8 ± 1.1%) (Table 3.3).

Twenty test hybrids from the two populations of Rutabaga-BF × Hi-Q flowered earlier than the *B. napus* parent Hi-Q. Among these, the test hybrids CO.BC.TH.29, CO.BC.TH.18, CO.FF.TH.30 and CO.BC.TH.19 showed the greatest negative MPH (-4.7 to -5.7%) and CPH (-2.1 to -5.3%) for this trait. In the case of the Rutabaga-BF × A07-26NR cross, 18 test hybrids of the BC_1- and F_2-derived lines flowered earlier than A07-26NR, and the lowest level of MPH (- 4.3 to – 5.6%) and CPH (-2.0 to -4.1%) for this trait was recorded for the test hybrids RR.BC.TH.31, RR.BC.TH.05, RR.BC.TH.32 and RR.BC.TH.24 (Tables A3.8, A3.9, A3.10 and A3.11).

Table 3.2: Analysis of variance for days to flowering (DTF) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q/A07-26NR (CPH) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used for analysis of variance.

Source	DTF of test hybrid					MPH for DTF					CPH for DTF				
	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]
Environment[1]	2		39.63			2		3.27			2		4.65		
Replication	2		0.04			2		0			2		0		
Block	16		1.3			16		0.68			16		0.98		
Cross[2] [C]	1	155.19	21.46	5.78	0.0174 *	1	35.27	0.6	0.04	0.8367	1	48.04	11.99	0.7	0.4069
Population[3] [P]	1	489.05	0.09	0.02	0.8761	1	152.7	100.04	7.14	0.0084 **	1	209.56	2.81	0.16	0.6857
C × P[4]	1	488.66	5.14	1.39	0.2397	1	174.6	16.94	1.21	0.2731	1	232.91	12.22	0.71	0.399
Test hybrid (C × P)[5]	89	500.49	7.31	1.97	3×10^{-6} ***	89	498.9	18.93	1.35	0.0257 *	89	506.7	42.28	2.47	3.2×10^{-10} ***
Residual	503		3.71			499		14.02			503		17.12		
Total	615					611					615				

[1] Year/site of the test hybrid field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the paternal inbred lines of the test hybrids were developed.
[3] Types of population indicate from where, the BC_1 or F_2, the paternal inbred lines of the test hybrids were developed.
[4] Combination of cross and type of population.
[5] Test hybrid is nested in C × P.
[6] Due to unbalanced data for days to flowering, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

Table 3.3: Days to flowering (DTF) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used to calculate the pooled lsmeans values of the test hybrids, and to calculate the MPH and CPH for the trait.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	Test hybrid (day)		MPH (%)		CPH (%)	
		Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	47.1 - 52.2	49.3 ± 3.7 a	-5.2 - 2.5	-2.5 ± 1.2 a	-5.3 - 6.5	-0.1 ± 1.4 a
	F	47.7 - 52.1	49.5 ± 3.7 a	-5.7 - 3.3	-2.0 ± 1.1 a	-3.7 - 8.9	0.5 ± 1.4 a
Rut-BF × 26NR[3]	BC	47.1 - 50.2	48.9 ± 3.7 a	-6.9 - 0.8	-2.9 ± 1.2 a	-4.1 - 6.5	-0.1 ± 1.4 a
	F	46.8 - 50.3	48.7 ± 3.7 a	-3.8 - 1.6	-1.6 ± 1.1 a	-4.4 - 3.1	-0.3 ± 1.4 a
Hi-Q[4]			48.9 ± 3.7 a				
A07-26NR[4]			48.8 ± 3.7 a				
Rut-BF × Hi-Q		47.1 - 52.2	49.5 ± 3.7 a	-5.7 - 3.3	-2.3 ± 1.2 a	-5.3 - 8.9	0.3 ± 1.4 a
Rut-BF × 26NR		46.8 - 50.3	48.7 ± 3.7 b	-6.9 - 1.6	-2.3 ± 1.1 a	-4.4 - 6.5	-0.2 ± 1.3 a
BC[5]		47.1 - 52.5	48.8 ± 3.7 a	-6.9 - 2.5	-2.7 ± 1.1 b	-5.3 - 6.5	-0.2 ± 1.4 a
F[6]		46.8 - 52.1	48.9 ± 3.7 a	-5.7 - 3.3	-1.8 ± 1.1 a	-4.4 - 8.9	0.1 ± 1.4 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

About 12% of the test hybrids of the BC_1- and F_2-derived lines of Rutabaga-BF × Hi-Q, and 14.3% and 19.0% test hybrids, respectively, of the BC_1- and F_2-derived inbred lines of Rutabaga-BF × A07-26NR flowered about two days earlier than the spring canola parent. The majority of the test hybrids of the BC_1- and F_2-derived lines of the two crosses (56 to 53.8% of Rutabaga-BF × Hi-Q and 71.4 to 76.2% of Rutabaga-BF × A07-26NR) was similar to the spring canola parent for days to flowering (Figure 3.2).

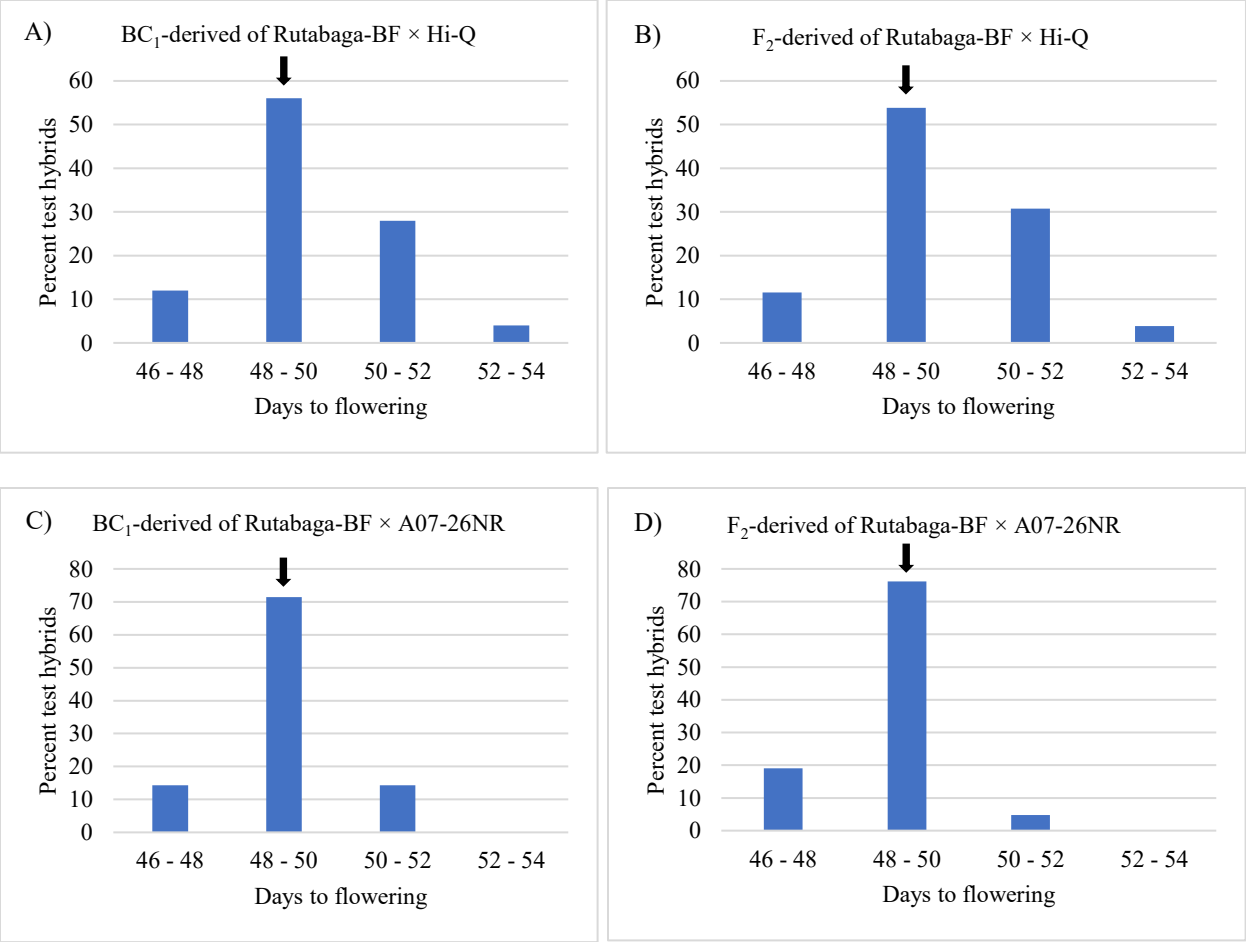

Figure 3.2: Frequency distribution of the test hybrids of the BC_1- and F_2-derived inbred lines of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses of *B. napus* for days to flowering. Pooled data of the 2014, 2015 and 2016 field trials for days to flowering was used to categorize the test hybrids of the BC_1- and F_2-derived inbred lines of Rutabaga-BF × Hi-Q (graphs A and B) and Rutabaga-BF × A07-26NR (graphs C and D). The arrows show the position of the spring *B. napus* canola parent (Hi-Q or A07-26NR).

3.3.2 Days to maturity

Analysis of variance for days to maturity of the test hybrids, and MPH and CPH are presented in Table 3.4. Significant variation between the test hybrids of the inbred lines of the two crosses was found ($p < 0.01$). Variation among the test hybrids as well as for the level of MPH and CPH was also significant ($p < 0.01$ and $p < 0.001$) for this trait.

Among the test hybrids of the inbred lines of the two crosses, test hybrids of Rutabaga-BF ×

Table 3.4: Analysis of variance for days to maturity (DTM) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q/A07-26NR (CPH) of the test hybrid for this trait. Three (2014, 2015 and 2016) years data was used for analysis of variance.

Source	DTM of test hybrid					MPH for DTM					CPH for DTM				
	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]
Environment[1]	2		43.87			2		1.2			2		2.92		
Replication	2		0.7			2		0.65			2		0.18		
Block	16		5.12			16		1.89			16		0.48		
Cross[2] [C]	1	186.83	112.26	9.42	0.0025 **	1	82.91	4.23	0.53	0.4671	1	27.83	6.13	0.57	0.4557
Population[3] [P]	1	500.69	31.09	2.61	0.1069	1	422.2	7.75	0.98	0.3237	1	123.6	6.49	0.6	0.4407
C × P[4]	1	500.13	6.7	0.56	0.4536	1	425.9	45.73	5.77	0.0168 *	1	140.11	28.99	2.71	0.1022
Test hybrid (C × P)[5]	89	496.94	18.4	1.54	0.0023 **	89	477	11.87	1.5	0.0044 **	89	491.62	19.05	1.78	7.1×10^{-5} ***
Residual	500		11.92			487		7.93			498		10.71		
Total	612					599					610				

[1] Year/site of the test hybrid field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the paternal inbred lines of the test hybrids were developed.
[3] Types of population indicate from where, the BC_1 or F_2, the paternal inbred lines of the test hybrids were developed.
[4] Combination of cross and type of population.
[5] Test hybrid is nested in C × P.
[6] Due to unbalanced data for days to maturity, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

Hi-Q inbred lines took significantly longer time to mature than the test hybrids of the inbred lines of Rutabaga-BF × A07-26NR (101.6 ± 3.9 vs. 100.4 ± 3.9 days); however, no significant difference was found between the test hybrids of the two types of inbred lines (F_2- and BC_1-derived). No significant difference could be detected between the test hybrids of the BC_1- and F_2-derived lines as well as between the lines of the two crosses for MPH and CPH. The four test hybrid populations of the four inbred line populations were statistically similar for days to maturity as well as displayed similar level of CPH; however, significant difference among these four populations was found for MPH (Table 3.5), as also evident from ANOVA (Table 3.4). MPH in all four populations was negative – suggesting the possibility of developing early maturing hybrid cultivars by use of the inbred lines used in this study.

Table 3.5: Days to maturity (DTM) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used to calculate the pooled lsmeans values of the test hybrids, and to calculate the MPH and CPH for the trait.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	Test hybrid (day)		MPH (%)		CPH (%)	
		Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	99.2 - 105.8	102.2 ± 3.9 a	-3.9 - 2.6	-1.6 ± 1.0 ab	-3.6 - 3.9	0.4 ± 1.1 a
	F	98.2 - 106.5	101.4 ± 3.9 a	-6.0 - 2.3	-2.0 ± 1.0 ab	-3.6 - 3.5	0.2 ± 1.1 a
Rut-BF × 26NR[3]	BC	98.4 - 104.1	100.6 ± 3.9 a	-4.5 - 0.4	-2.1 ± 0.9 b	-2.9 - 2.7	-0.4 ± 1.1 a
	F	98.1 - 102.9	100.3 ± 3.9 a	-2.7 - 1.0	-0.9 ± 0.9 a	-2.1 - 3.4	0.3 ± 1.1 a
Hi-Q[4]			100.9 ± 3.9 a				
A07-26NR[4]			100.0 ± 3.9 a				
Rut-BF × Hi-Q		98.2 - 106.5	101.6 ± 3.9 a	-6.0 - 2.6	-1.9 ± 0.9 a	-3.6 - 3.9	0.3 ± 1.0 a
Rut-BF × 26NR		98.1 - 104.1	100.4 ± 3.9 b	-4.5 - 1.0	-1.4 ± 0.9 a	-2.9 - 3.4	0.0 ± 1.0 a
BC[5]		98.4 - 105.8	100.9 ± 4.2 a	-4.5 - 2.6	-1.5 ± 0.8 a	- 3.6 - 3.9	0.0 ± 1.0 a
F[6]		98.1 - 106.5	100.4 ± 4.2 a	-6.0 - 2.3	-1.4 ± 0.8 a	-3.6 - 3.5	0.2 ± 1.0 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Thirty-three and 14 test hybrids, respectively, of the inbred lines of Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR matured earlier than the spring canola parent. All these test hybrids, displayed negative MPH, and the majority also showed negative CPH. Of these test hybrids, the lowest negative MPH and CPH was observed for the test hybrids CO.FF.TH.19 (-6.0 and -3.6%) and CO.BC.TH.32 (-3.7 and -3.6%) of the Rutabaga-BF × Hi-Q cross, and for RR.BC.TH.28 (-4.5 and -2.1%), RR.BC.TH.26 (-3.9 and -2.9%) and RR.BC.TH.32 (-3.6 and -2.2%) of the Rutabaga-BF × A07-26NR cross (Tables A3.8, A3.9, A3.10 and A3.11).

3.3.3 Plant height

Significant variation in plant height was found between the test hybrid populations derived from the inbred lines of the two crosses ($p < 0.001$). Variation for MPH and CPH observed in the test hybrid population was significant ($p < 0.05$), and variation for CPH due to the crosses was also significant for this trait ($p < 0.01$) (Table 3.6).

Test hybrid population of the inbred lines of Rutabaga-BF × Hi-Q was significantly taller than the test hybrid population of the inbred lines of Rutabaga-BF × A07-26NR (124.2 ± 10.7 vs. 115.8 ± 10.7 cm); however, the two test hybrid populations based on the F_2- and BC_1-derived inbred lines were statistically similar, and also not significantly different from their spring canola parent Hi-Q or A07-26NR. No significant difference was found between the test hybrid populations of the inbred lines of the two crosses for MPH; however, the difference due to the cross was significant for CPH (1.1 ± 1.9 vs. -1.0 ± 1.9%). Similar level of MPH and CPH was found for the BC_1- and F_2-derived inbred lines. The level of MPH detected among the four test hybrid populations of the two crosses were also statistically similar (Table 3.7).

As compared to the spring *B. napus* canola parent, 42 test hybrids of the inbred lines of Rutabaga-BF

Table 3.6: Analysis of variance of plant height of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q/A07-26NR (CPH) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used for analysis of variance.

Source	Plant height of test hybrid					MPH for plant height					CPH for plant height				
	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]
Environment[1]	2		350.84			2		13.4			2		8.92		
Replication	2		10.47			2		0.34			2		0		
Block	16		2.9			16		1.93			16		0.41		
Cross[2] [C]	1	53.97	6025.8	89.3	4.8×10^{-13} ***	1	30.74	112.12	2.02	0.1655	1	25.77	607.62	8.12	0.0085 **
Population[3][P]	1	200.39	19.2	0.29	0.5938	1	115.8	45.51	0.82	0.3673	1	43.09	0.92	0.01	0.9121
C × P[4]	1	224.02	266.5	3.95	0.048 *	1	135.5	0.34	0.01	0.9377	1	56.14	273.06	3.65	0.0612
Test hybrid (C × P)[5]	89	506.58	129.6	1.92	6.4×10^{-6} ***	89	497.4	73.36	1.32	0.0361 *	89	493.32	113.47	1.52	0.0033 **
Residual	501		67.41			501		55.57			501		74.8		
Total	613					613					613				

[1] Year/site of the test hybrid field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the paternal inbred lines of the test hybrids were developed.
[3] Types of population indicate from where, the BC_1 or F_2, the paternal inbred lines of the test hybrids were developed.
[4] Combination of cross and type of population.
[5] Test hybrid is nested in C × P.
[6] Due to unbalanced data for plant height, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

× Hi-Q and 19 test hybrids of the inbred lines of Rutabaga-BF × A07-26NR had shorter plant height. The greatest negative MPH and CPH for this trait was found for the test hybrids CO.BC.TH.25, CO.FF.TH.15, CO.FF.TH.29, CO.BC.TH.08 and CO.FF.TH.18 of Rutabaga-BF × Hi-Q (-2.9 to - 11.0% for MPH and -5.7 to -7.7% for CPH) and RR.BC.TH.18, RR.FF.TH.27, RR.FF.TH.20 and RR.BC.TH.28 of the Rutabaga-BF × A07-26NR cross (- 3.1 to -7.2% for MPH and -2.5 to -5.8% for CPH) (Tables A3.8, A3.9, A3.10 and A3.11).

Table 3.7: Plant height (cm) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used to calculate the pooled lsmeans values of the test hybrids, and to calculate the MPH and CPH for the trait.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	Test hybrid (cm)		MPH (%)		CPH (%)	
		Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	115.4 - 131.1	122.8 ± 10.8 a	-5.3 - 6.3	-0.3 ± 2.3 a	-7.6 - 10.3	-2.1 ± 2.0 b
	F	116.9 - 137.0	125.1 ± 10.7 a	-11 - 6.9	-0.6 ± 2.3 a	-7.7 - 11.9	-0.1 ± 1.9 ab
Rut-BF × 26NR[3]	BC	107.4 - 125.2	116.6 ± 10.8 b	-7.2 - 6.7	1.2 ± 2.3 a	-5.5 - 11.2	2.1 ± 2.0 a
	F	106.9 - 127.5	115.2 ± 10.7 b	-5.3 - 6.4	0.2 ± 2.3 a	-5.8 - 7.8	0.4 ± 2.0 ab
Hi-Q[4]			123.6 ± 10.8 a				
A07-26NR[4]			114.8 ± 10.8 b				
Rut-BF × Hi-Q		115.4 - 137.0	124.2 ± 10.7 a	-11.0 - 6.9	-0.4 ± 2.2 a	-7.7 - 11.9	-1.0 ± 1.9 b
Rut-BF × 26NR		106.9 - 127.5	115.8 ± 10.7 b	-7.2 - 6.7	0.7 ± 2.2 a	-5.8 - 11.2	1.1 ± 1.9 a
BC[5]		107.4 - 131.1	117.2 ± 11.7 a	-7.2 - 6.7	0.6 ± 2.2 a	-7.6 - 11.2	-0.1 ± 1.9 a
F[6]		106.9 - 137.0	118.5 ± 11.7 a	-11.0 - 6.9	-0.1 ± 2.2 a	-7.7 - 11.9	0.1 ± 1.9 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

3.3.4 Seed yield

Analysis of variance for seed yield of the test hybrid populations is presented in Table 3.8.

Table 3.8: Analysis of variance for seed yield of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q/A07-26NR (CPH) for this trait. Two (2015 and 2016) years data was used for analysis of variance.

Source	Seed yield of test hybrid					MPH for seed yield					CPH for seed yield				
	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]
Environment[1]	1		925451.00			1		0.00			1		42.24		
Replication	1		2838.00			1		0.00			1		3.36		
Block	16		273465.00			16		5.06			16		6.0×10^{-14}		
Cross[2] [C]	1	93.84	471595.00	1.24	0.2666	1	29.49	5298.90	18.71	0.0002 ***	1	253.22	4829.20	15.69	9.7×10^{-5} ***
Population[3] [P]	1	251.10	496278.00	1.31	0.2527	1	72.16	4729.90	16.70	0.0001 ***	1	253.14	3321.40	10.79	0.0012 **
C × P[4]	1	250.96	38499.00	0.10	0.7498	1	99.81	1455.20	5.13	0.0256 *	1	253.03	534.20	1.74	0.1889
Test hybrid (C × P)[5]	89	234.75	587572.00	1.56	0.0045 **	89	224.66	384.10	1.36	0.0376 *	89	253.03	345.20	1.12	0.2453
Residual	226		377648.00			217		283.16			237		307.80		
Total	336					327					347				

[1] Year/site of the test hybrid field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the paternal inbred lines of the test hybrids were developed.
[3] Types of population indicate from where, the BC_1 or F_2, the paternal inbred lines of the test hybrids were developed.
[4] Combination of cross and type of population.
[5] Test hybrid is nested in C × P.
[6] Due to unbalanced data for seed yield, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

Significant variation for seed yield was found within the whole population; however, variation between the test hybrids due to the cross and type of population (BC_1- and F_1-derived) was not significant. Variation due to the cross and type of population was significant for MPH and CPH for this trait (Table 3.8). Significant variation for MPH, but not for CPH, was found in the whole test hybrid population.

Mean seed yield of the two test hybrid populations of the inbred lines of the two crosses were statistically similar; however, the test hybrids of the BC_1- and F_2-derived lines of Rutabaga-BF × Hi-Q, on average, gave significantly greater seed yield than the spring canola parent (Table 3.9). Greater proportion of the test hybrids of this cross also gave high seed yield as compared to the test hybrids of the inbred lines of Rutabaga-BF × A07-26NR (Figure 3.3). MPH and CPH for seed yield was positive in all populations. On average, higher levels of MPH and CPH for seed yield was observed for the inbred lines derived from Rutabaga-BF × Hi-Q as compared to the inbred lines of Rutabaga-BF × A07-26NR (24.1 ± 1.9 vs. 12.7 ± 4.9% for MPH, and 12.6 ± 1.8 vs. 4.3 ± 5.0% for CPH). As compared to the F_2-derived lines, the BC_1-derived lines gave significantly greater MPH and CPH (Table 3.9). The lowest level MPH for seed yield were found in the F_2-derived lines of Rutabaga-BF × A07-26NR (7.0 ± 2.2%).

Almost all (50) test hybrids of the inbred lines of Rutabaga-BF × Hi-Q and 36 test hybrids of the inbred lines of Rutabaga-BF × A07-26NR exhibited positive MPH for seed yield, ranging from 3.7 to 48.0%, and 0.3 to 57.4%, respectively. Most of these test hybrids also showed positive CPH (1.4 to 37.5% for Rutabaga-BF × Hi-Q and 1.3 to 28.7% in Rutabaga-BF × A07-26NR) for seed yield. Forty-seven and 24 test hybrids of the inbred lines of the two crosses out-yielded the respective spring canola parent for seed yield.

The test hybrids CO.FF.TH.18, CO.BC.TH.32 and CO.FF.TH.24 were highest-yielding ones of Rutabaga-BF × Hi-Q giving seed yield of 5987.1, 5814.8 and 5755.7 kg/ha and exhibiting 29.6, 44.7 and 26.7% MPH and 15.9, 31.7 and 29.2% CPH. In case of Rutabaga-BF × A07-26NR, the test hybrids RR.FF.TH.13, RR.BC.TH.33 and RR.BC.TH.26 gave the highest seed yield (5975.1, 5876.8 and 5554.9 kg/ha) and displayed 30.0, 29.8 and 19.1% MPH and 12.0, 28.7 and 3.9% CPH (Tables A3.8, A3.9, A3.10 and A3.11).

Table 3.9: Seed yield (kg/ha) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) of the test hybrids for this trait. Two (2015 and 2016) years data was used to calculate the pooled lsmeans values of the test hybrids, and to calculate the MPH and CPH for the trait.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	Test hybrid (kg/ha)		MPH (%)		CPH (%)	
		Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	4697.8 - 5814.8	5253.0 ± 667.5 a	3.7 - 48.0	25.6 ± 2.2 a	1.7 - 37.5	14.3 ± 5.2 a
	F	4184.4 - 5987.1	5121.2 ± 669.1 a	-0.4 - 37.4	21.7 ± 2.2 a	-7.8 - 29.2	10.6 ± 5.2 a
Rut-BF × 26NR[3]	BC	4423.8 - 5876.8	4969.4 ± 665.2 ab	-4.1 - 57.4	19.2 ± 2.4 a	-17.9 - 28.7	8.7 ± 5.3 a
	F	4039.8 - 5975.1	4848.5 ± 664.3 ab	-4.7 - 37.0	7.0 ± 2.2 b	-14.3 - 13.1	0.2 ± 5.2 b
Hi-Q[4]			4642.3 ± 675.3 b				
A07-26NR[4]			4816.6 ± 671.6 ab				
Rut-BF × Hi-Q		4184.4 - 5987.1	5198.1 ± 662.6 a	-0.4 - 48.0	24.1 ± 1.9 a	-7.8 - 37.5	12.7 ± 4.9 a
Rut-BF × 26NR		4039.8 - 5975.1	4903.7 ± 657.9 a	-4.7 - 57.4	12.6 ± 1.8 b	-17.9 - 28.7	4.3 ± 5.0 b
BC[5]		4423.8 - 5876.8	5063.4 ± 713.2 a	-4.1 - 57.4	22.5 ± 2.3 a	-17.9 - 37.5	11.9 ± 5.2 a
F[6]		4039.8 - 5987.1	4924.8 ± 713.3 a	-4.7 - 37.4	13.7 ± 2.3 b	-14.3 - 29.2	5.5 ± 5.2 b

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

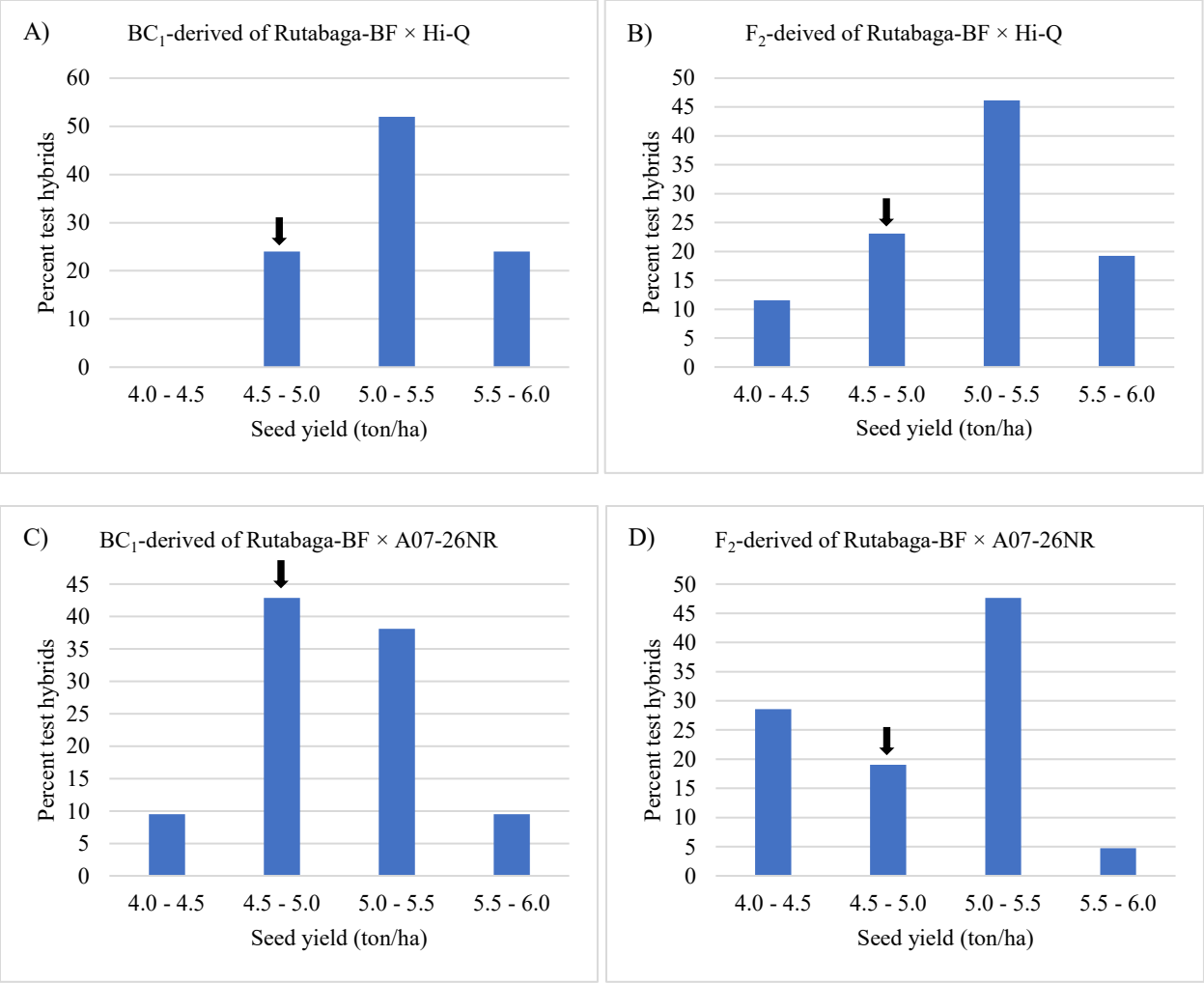

Figure 3.3: Frequency distribution of the test hybrids of the BC_1- and F_2-derived inbred lines of the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses of *B. napus* for seed yield. Pooled data of the 2015 and 2016 field trials for seed yield was used to categorize the test hybrids of the BC_1- and F_2-derived inbred lines of Rutabaga-BF × Hi-Q (graphs A and B) and Rutabaga-BF × A07-26NR (graphs C and D). The arrows show the position of the spring *B. napus* canola parent (Hi-Q or A07-26NR).

3.3.5 Seed oil content

Significant variation was found among the test hybrids for seed oil content ($p < 0.001$). The effects of the cross and the type of population were not significant; however, the interaction between the cross and population was significant for this trait (Table 3.10). Significant variation for MPH and CPH was found within the test hybrid population, and variation due to the cross was

Table 3.10: Analysis of variance for seed oil content of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q/A07-26NR (CPH) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used for analysis of variance.

Source	Seed oil content of test hybrid					MPH for seed oil content					CPH for seed oil content				
	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]
Environment[1]	2		2.43			2		0.54			2		7.46E-15		
Replication	2		0.05			2		0			2		0		
Block	16		1.29			16		0.23			16		0.32		
Cross[2][C]	1	368.97	0.01	0.01	0.9235	1	41.56	47.18	6.12	0.0176 *	1	115.96	256.49	29.9	2.7×10^{-7} ***
Population[3] [P]	1	514.2	0.78	0.55	0.4594	1	130.1	2.11	0.27	0.6016	1	186.37	41.73	4.86	0.0287 *
C × P[4]	1	514.26	12.67	8.9	0.003 **	1	158.6	3.39	0.44	0.5082	1	222.71	62.99	7.33	0.0073 **
Test hybrid (C × P)[5]	89	496.9	8.07	5.67	$< 2\times10^{-16}$ ***	89	494	10.5	1.36	0.023 *	89	504.55	39.88	4.64	$< 2.2\times10^{-16}$ ***
Residual	498		1.42			487		7.71			494		8.59		
Total	610					599					606				

[1] Year/site of the test hybrid field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the paternal inbred lines of the test hybrids were developed.
[3] Types of population indicate from where, the BC_1 or F_2, the paternal inbred lines of the test hybrids were developed.
[4] Combination of cross and type of population.
[5] Test hybrid is nested in C × P.
[6] Due to unbalanced data for seed oil content, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

also significant for MPH and CPH. The type of population (F_2- or BC_1-derived) did not exhibit a significant effect on MPH; however, the level of CPH was significantly affected by the type of population for this trait (Table 3.10).

No significant differences could be found between the test hybrids of the inbred lines of the two crosses as well as between the test hybrids of the BC_1- and F_2-derived lines for mean seed oil content. In case of the Rutabaga-BF × Hi-Q cross, the difference between the two test hybrid populations of the F_2- and BC_1-derived lines and Hi-Q was not significant, while the test hybrids of the BC_1-derived lines of Rutabaga-BF × A07-26-NR had significantly lower oil than A07-26NR. Average MPH and CPH of the test hybrids of the inbred lines of the Rutabaga-BF × Hi-Q cross were 0.9 ± 0.5 and -0.2 ± 0.3%, respectively; these values were statistically greater than the MPH and CPH values (0.2 ± 0.5 and -1.7 ± 0.3%) of the Rutabaga-BF × A07-26-NR cross. While comparing the test hybrid populations of the F_2- and BC_1-derived lines, no significant difference could be found; however, the difference between these two test hybrid populations was significant for CPH (Table 3.11).

Seed oil content of 25 and 14 test hybrids, respectively, of the inbred lines of Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR surpassed the seed oil content of the spring canola parent. In these test hybrids, the level of MPH varied from 0.4 to 4.1% and from 0.3 to 2%, and CPH varied from 0.3 to 5.7% and 0.2 to 1.4%, respectively, for the Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR crosses. The test hybrids CO.FF.TH.18, CO.BC.TH.08 and CO.FF.TH.40 of the inbred lines of Rutabaga-BF × Hi-Q had the greatest seed oil content (49.9, 49.4 and 49.2 %) and showed 2.0 to 4.1% MPH and 4.4 to 5.7% CPH for this trait. In case of Rutabaga-BF × A07-26NR, the test hybrids RR.BC.TH.28, RR.FF.TH.23, RR.FF.TH.19 and RR.FF.TH.20 had 48.6, 48.5, 48.4 and

Table 3.11: Seed oil content (%) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used to calculate the pooled lsmeans values of the test hybrids, and to calculate the MPH and CPH for the trait.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	Test hybrid (%)		MPH (%)		CPH (%)	
		Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	45.3 - 49.4	47.2 ± 1.0 ab	-1.7 - 3.7	1.0 ± 0.5 a	-4.6 - 4.3	-0.2 ± 0.4 a
	F	44.8 - 49.9	46.9 ± 1.0 b	-1.4 - 4.1	0.9 ± 0.5 a	-5.7 - 5.7	-0.2 ± 0.3 a
Rut-BF × 26NR[3]	BC	43.9 - 48.6	46.9 ± 1.0 b	-2.5 - 2.0	0.4 ± 0.5 a	-8.0 - 0.9	-2.4 ± 0.4 b
	F	45.2 - 48.5	47.4 ± 1.0 ab	-2.6 - 1.9	0.0 ± 0.5 a	-4.8 - 1.4	-0.9 ± 0.4 a
Hi-Q[4]			47.2 ± 1.0 ab				
A07-26NR[4]			47.8 ± 1.0 a				
Rut-BF × Hi-Q		44.8 - 49.9	47.0 ± 1.0 a	-1.7 - 4.1	0.9 ± 0.5 a	-5.7 - 5.7	-0.2 ± 0.3 a
Rut-BF × 26NR		43.9 - 48.6	47.1 ± 1.0 a	-2.6 - 2.0	0.2 ± 0.5 b	-8.0 - 1.4	-1.7 ± 0.3 b
BC[5]		43.9 - 49.4	47.1 ± 1.0 a	-2.5 - 3.7	0.5 ± 0.5 a	-8.0 - 4.3	-1.4 ± 0.3 b
F[6]		44.8 - 49.9	47.1 ± 1.0 a	- 2.6 - 4.1	0.5 ± 0.5 a	-5.7 - 5.7	-0.7 ± 0.3 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

48.3% seed oil content and displayed 0.7, 0.3, 0.5 and 1.0% MPH, and 0.8, 0.7, 0.2 and 1.4% CPH (Tables A3.12, A3.13, A3.14 and A3.15).

3.3.6 Seed protein content

Significant variation ($p < 0.001$) was found between the two test hybrid populations of the F_2- and BC_1-derived inbred lines and within the whole test hybrid population of the two crosses, while no significant variation was found between the two test hybrid populations based on the inbred lines of the two crosses for seed protein content. However, significant variation between the two test hybrid populations of the two crosses including the two populations based on F_2- and BC_1-

derived lines was detected for both MPH and CPH (Table 3.12). Interaction between the cross and the type of population (F_2- and BC_1-derived) was significant ($p < 0.05$) for seed protein content of the test hybrids as well as for CPH for this trait (Table 3.12).

Among the two test hybrid populations developed based on the F_2- or BC_1- derived lines, the test hybrids of the F_2-derived lines, had higher seed protein content as well as exhibited significantly greater MPH and CPH than the test hybrids of the BC_1-derived lines (Table 3.13). No significant difference between the test hybrids due to the cross was found for seed protein content; however, the test hybrids of the inbred lines of Rutabaga-BF × A07-26NR exhibited greater MPH and CPH than the test hybrids of the inbred lines of Rutabaga-BF × Hi-Q. In case of the Rutabaga-BF × Hi-Q cross, mean seed protein content of the test hybrids of the F_2-derived population was significantly higher than the test hybrids of the BC_1-derived population; however, these two test hybrid populations were not significantly different from Hi-Q for this trait. In case of the Rutabaga-BF × A07-26NR cross, mean seed protein content of the two test hybrid populations were also similar to A07-26NR. The two test hybrid populations of Rutabaga-BF × Hi-Q, on average, gave negative MPH; in contrast, the test hybrids of the F_2-derived lines of Rutabaga-BF × A07-26NR gave a positive MPH for seed protein content (Table 3.13).

As compared to the check spring canola parent, 19 and 30 test hybrids of the BC_1- and F_2-derived inbred lines of Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR exhibited higher seed protein content. The best test hybrids of the inbred lines of Rutabaga-BF × Hi-Q were CO.FF.TH.38, CO.FF.TH.35 and CO.FF.TH.33; these test hybrids had 28.9, 28.3 and 28.3% seed protein content and exhibited 2.2, 3.7 and 1.2% MPH and 7.8, 6.7 and 5.2% CPH, respectively. The best test hybrids of the inbred lines of Rutabaga-BF × A07-26NR were RR.BC.TH.27,

Table 3.12: Analysis of variance for seed protein content of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q/A07-26NR (CPH) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used for analysis of variance.

Source	Seed protein content of test hybrid					MPH for seed protein content					CPH for seed protein content				
	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]
Environment[1]	2		1.67			2		0.63			2		0.8		
Replication	2		0.01			2		0			2		0		
Block	16		0.67			16		0.18			16		0.65		
Cross[2] [C]	1	287.65	0.56	0.55	0.4609	1	36.16	204.22	15.7	0.0003 ***	1	66.17	711.85	38.3	4.3×10^{-8} ***
Population[3] [P]	1	512.59	19.22	18.7	1.9×10^{-5} ***	1	79.72	173.87	13.4	0.0005 ***	1	191.01	196.42	10.6	0.0014 **
C × P[4]	1	512.11	9.4	9.12	0.0027 **	1	103.3	7.81	0.6	0.4404	1	223.61	145.69	7.85	0.0055 **
Test hybrid (C × P)[5]	89	497.38	5.24	5.08	$< 2.2\times10^{-16}$ ***	89	494.9	16.22	1.25	0.0774	89	504.8	73.59	3.96	$< 2.2\times10^{-16}$ ***
Residual	498		1.03			487		13.02			494		18.57		
Total	610					599					606				

[1] Year/site of the test hybrid field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the paternal inbred lines of the test hybrids were developed.
[3] Types of population indicate from where, the BC_1 or F_2, the paternal inbred lines of the test hybrids were developed.
[4] Combination of cross and type of population.
[5] Test hybrid is nested in C × P.
[6] Due to unbalanced data for seed protein content, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

RR.BC.TH.31 and RR.FF.TH.35 having 28.8, 28.4 and 28.3% seed protein content and exhibiting1.2, 0.7 and 3.1% MPH and 9.2, 8.2 and 7% CPH (Tables A3.12, A3.13, A3.14 and A3.15).

Table 3.13: Seed protein content (%) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used to calculate the pooled lsmeans values of the test hybrids, and MPH and CPH for the trait.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	Test hybrid (%)		MPH (%)		CPH (%)	
		Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	23.5 - 28.0	26.4 ± 0.8 b	-7.6 - 1.4	-1.7 ± 0.6 b	-11.0 - 4.7	-0.9 ± 0.8 b
	F	25.9 - 28.9	27.1 ± 0.8 a	-3.4 - 3.7	-0.9 ± 0.6 b	-3.1 - 7.8	1.0 ± 0.7 a
Rut-BF × 26NR[3]	BC	25.0 - 28.8	26.7 ± 0.8 ab	-3.3 - 1.9	-0.7 ± 0.6 ab	-4.3 - 9.8	2.5 ± 0.8 a
	F	26.0 - 28.3	26.8 ± 0.8 ab	-2.6 - 3.1	0.6 ± 0.6 a	-1.6 - 8.1	2.4 ± 0.7 a
Hi-Q[4]			26.9 ± 0.8 ab				
A07-26NR[4]			26.2 ± 0.8 b				
Rut-BF × Hi-Q		23.5 - 28.9	26.9 ± 0.9 a	-7.6 - 3.7	-1.1 ± 0.5 b	-11.0 - 7.8	0.2 ± 0.6 b
Rut-BF × 26NR		25.0 - 28.8	26.7 ± 0.8 a	-3.3 - 3.1	0.0 ± 0.5 a	-4.3 - 9.8	2.4 ± 0.6 a
BC[5]		23.5 - 28.8	26.5 ± 0.8 b	-7.6 - 1.9	-0.8 ± 0.4 b	-11 - 9.8	1.2 ± 0.7 b
F[6]		25.9 - 28.9	27.0 ± 0.8 a	-3.4 - 3.7	0.0 ± 0.4 a	-3.1 - 8.1	2.1 ± 0.7 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

3.3.7 Seed glucosinolate content

Analysis of variance (Table 3.14) showed that significant variation existed within the whole test hybrid population as well as between the two test hybrid populations partitioned based on the inbred lines of the two crosses for seed glucosinolate content ($P < 0.001$), while no significant

Table 3.14: Analysis of variance for seed glucosinolate content of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q/A07-26NR (CPH) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used for analysis of variance.

Source	Seed glucosinolate content of test hybrid					MPH for seed glucosinolate content					CPH for seed glucosinolate content				
	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]	df	Error df[6]	MS	F value	p value[7]
Environment[1]	2		0.13			2		14.94			2		11.77		
Replication	2		0.01			2		0.05			2		0		
Block	16		1.01			16		0			16		1.99		
Cross[2] [C]	1	130.8	339.97	83.8	8.9×10^{-16} ***	1	498.8	190.13	1.92	0.166	1	23.7	2324.03	20.2	0.0002 ***
Population[3][P]	1	421.81	1.36	0.34	0.5629	1	499.2	5.773	0.06	0.8091	1	62.42	476.94	4.15	0.0458 *
C × P[4]	1	428.17	49.76	12.3	0.0005 ***	1	498.9	75.12	0.76	0.3837	1	78.64	120.19	1.05	0.3095
Test hybrid (C × P)[5]	89	495.52	27.98	6.9	$< 2.2\times10^{-16}$ ***	89	498.9	230.56	2.33	4.5×10^{-9} ***	89	475.6	576.12	5.02	$< 2.2\times10^{-16}$ ***
Residual	498		4.06			482		98.81			478		114.85		
Total	610					594					590				

[1] Year/site of the test hybrid field trials.
[2] Original cross (Rutabaga-BF × Hi-Q or Rutabaga-BF × A07-26NR) from which the paternal inbred lines of the test hybrids were developed.
[3] Types of population indicate from where, the BC_1 or F_2, the paternal inbred lines of the test hybrids were developed.
[4] Combination of cross and type of population.
[5] Test hybrid is nested in C × P.
[6] Due to unbalanced data for seed glucosinolate content, Satterwaite's synthesis method from 'lmerTest' package of the software program R (Kuznetsova et al., 2016) was applied to calculate denominator degrees of freedom.
[7] *, ** and *** = $p < 0.05$, 0.01 and 0.001, respectively.

variation was found between the two test hybrid populations partitioned based on the type (F_2- or BC_1-derived) of inbred lines. No significant variation was found for MPH due to cross or population difference; however, significant variation within the whole test hybrid population was found for this phenomenon. In the case of CPH, significant variation was found within the whole test hybrid population.

Seed glucosinolate content of the test hybrid population of the inbred lines of Rutabaga-BF × A07-26NR (16.2 ± 0.4 µmol/g) was significantly higher as compared to Rutabaga-BF × Hi-Q (14.1 ± 0.5 µmol/g); however, in both cases, mean seed glucosinolate content of the test hybrids of the BC_1- and F_2-derived lines were similar to the spring canola parent Hi-Q or A07-26NR. All four

Table 3.15: Seed glucosinolate content (µmol/g) of the test hybrids, produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) of the test hybrids for this trait. Three (2014, 2015 and 2016) years data was used to calculate the pooled lsmeans values of the test hybrids, and to calculate the MPH and CPH for the trait.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	Test hybrid (µmol/g)		MPH (%)		CPH (%)	
		Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE
Rut-BF × Hi-Q[2]	BC	11.2 - 22.7	13.7 ± 0.5 c	-12.1 - 41.8	-3.5 ± 2.6 a	-17.6 - 56.0	-2.1 ± 2.6 b
	F	12.1 - 20.9	14.3 ± 0.5 bc	-9.8 - 9.5	-3.3 ± 2.5 a	-13.1 - 11.7	-1.8 ± 2.5 b
Rut-BF × 26NR[3]	BC	13.8 - 24.4	16.6 ± 0.5 a	-16.4 - 8.6	-3.7 ± 2.6 a	-13.8 - 48.0	3.5 ± 2.6 a
	F	14.0 - 17.2	15.8 ± 0.5 a	-11.0 - 3.8	-3.5 ± 2.6 a	-10.3 - 11.4	1.9 ± 2.6 ab
Hi-Q[4]			13.8 ± 0.6 bc				
A07-26NR[4]			15.5 ± 0.6 ab				
Rut-BF × Hi-Q		11.2 - 22.7	14.1 ± 0.5 b	-12.1 - 41.8	-3.4 ± 2.5 a	-17.6 - 56.0	-1.9 ± 2.4 b
Rut-BF × 26NR		13.8 - 24.4	16.2 ± 0.4 a	-16.4 - 8.6	-3.6 ± 2.5 a	-13.8 - 48.0	2.7 ± 2.4 a
BC[5]		11.2 - 24.4	15.6 ± 0.7 a	-16.4 - 41.8	-3.6 ± 2.5 a	-17.6 - 56.0	1.2 ± 2.3 a
F[6]		12.1 - 20.9	15.4 ± 0.7 a	-11.0 - 9.5	-3.4 ± 2.5 a	-13.1 - 11.7	0.1 ± 2.3 b

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

populations of the two crosses exhibited negative MPH for seed glucosinolate content. In the case of CPH, only the test hybrids of the inbred lines of Rutabaga-BF × Hi-Q exhibited negative heterosis, while the test hybrids of the inbred lines of Rutabaga-BF × A07-26NR exhibited positive heterosis for this trait. As compared to the test hybrids of the F_2-derived lines (0.1 ± 2.3%), the test hybrids of the BC_1-derived lines (1.2 ± 2.3%) exhibited significantly greater CPH; no significant difference was found between these two groups of test hybrids for MPH (Table 3.15).

Thirty-nine and 17 test hybrids of the BC_1- and F_2-derived lines of Rutabaga-BF × Hi-Q and Rutabaga-BF × A07-26NR had lower seed glucosinolate content than the spring canola parent. The test hybrids CO.BC.TH.18, CO.BC.TH.21 and CO.BC.TH.33 of the inbred lines of Rutabaga-BF × Hi-Q had 11.2, 11.7 and 11.8 µmol/g seed glucosinolate content and exhibited the lowest negative MPH (-9.7, -5.9 and -4.9%) and CPH (-17.6, -10.8 and -10.9%). In case of the test hybrids of the inbred lines of Rutabaga-BF × A07-26NR, RR.BC.TH.19, RR.BC.TH.26 and RR.FF.TH.19 had 13.8, 13.9 and 14 µmol/g seed glucosinolate content and exhibited -8.5, -9.8 and -10.9% MPH and -13.8, -12.7 and -10.3% CPH (Tables A3.12, A3.13, A3.14 and A3.15).

3.3.8 Correlation between seed yield of the inbred lines and heterosis

No significant correlation was detected between seed yield of the inbred lines and seed yield of the test hybrids ($r = 0.21^{n.s.}$ for Rutabaga-BF × Hi-Q and $r = 0.06^{n.s.}$ for Rutabaga-BF × A07-26-NR). Correlation between seed yield of the inbred lines and MPH ($r = 0.01^{n.s.}$ and $r = -0.19^{n.s.}$) and CPH ($r = 0.12^{n.s.}$ and $r = -0.10^{n.s.}$) was also not significant.

Correlation between genetic divergence of the inbred lines from their spring *B. napus* canola parent (Hi-Q or A07-26NR) and MPH for seed yield was almost zero ($r = -0.08^{n.s.}$) in the case of the Rutabaga-BF × Hi-Q cross, while it was strong and positive (r = 0.56 ***) in the Rutabaga-BF

× A07-26-NR cross. In the Rutabaga-BF × A07-26-NR cross, this correlation was much stronger for the F_2-derived population than the BC_1-derived population (r = 0.59 *** vs. r = 0.04 $^{n.s.}$). Correlation between genetic distance of the inbred lines from the spring canola parent and CPH for seed yield was not consistent in the two crosses (r = -0.35 * for Rutabaga-BF × Hi-Q and r = 0.38* for Rutabaga-BF × A07-26NR).

The best three high-yielding test hybrids CO.FF.TH.18, CO.BC.TH.32 and CO.FF.TH.24 of Rutabaga-BF × Hi-Q exhibited a negative MPH and CPH for days to flowering and maturity, and a positive MPH and CPH for seed oil content. These test hybrids were similar to Hi-Q for seed protein and glucosinolate content. With the Rutabaga-BF × A07-26NR cross, the three highest yielding test hybrids RR.FF.TH.13, RR.BC.TH.33 and RR.BC.TH.26 were similar to A07-26NR for days to flowering and maturity, seed oil and glucosinolate content; however, these test hybrids had higher seed protein content (Tables A3.8, A3.9, A3.10, A3.11, A3.12, A3.13, A3.14 and A3.15).

3.4 Discussion

The increased productivity in some of the field crops in the world has been achieved, to a great extent, from the exploitation of heterosis (for review, see Schnable and Springer, 2013; for review, see Groszmann et al., 2013). In Canada, hybrid *B. napus* cultivars have made a significant contribution to the increased production of this crop in the 2000's (McVetty et al., 2016; Morrison et al., 2016). According to Fu and Gugel (2010), a decline in genetic diversity in this crop has occurred over several years of breeding. Several researchers, such as Grant and Beversdorf (1985) and Lefort-Buson et al. (1987a), reported that genetic divergence between the parents is needed to achieve a high level of heterosis in hybrid cultivars. Therefore, success for exploitation of the

phenomenon 'heterosis' for continued improvement of seed yield in hybrid cultivars would depend on increasing the level of divergence of the favorable alleles in this crop (for review, see Rahman, 2013).

Introgression of favorable alleles and creation of a heterotic gene pool in a breeding population is important in a hybrid breeding program (Gehringer et al., 2007; for review, see Rahman, 2013). The primary gene pools of spring *B. napus*, such as winter and semi-winter *B. napus*, have been used to diversify the genetic base of this crop and to understand the value of these gene pools for heterosis in spring *B. napus* canola (Quijada et al., 2004; Udall et al., 2004; Qian et al., 2007; Rahman et al., 2016). However, very little is known about the value of Rutabaga for broadening the diversity of alleles for different agronomic and seed quality traits in spring *B. napus* canola, and exploitation of this gene pool for increasing the level of heterosis in hybrid cultivars.

In the present study, by use of the spring *B. napus* canola inbred lines derived from the Rutabaga × spring canola crosses, positive MPH for seed yield was detected in 92.5% of the test hybrids; where the majority of the test hybrids showed positive CPH for this trait. As a result, more than 55% of the test hybrids surpassed seed yields of the spring *B. napus* canola parents. The test hybrids of the inbred lines of Rutabaga-BF × Hi-Q gave similar seed yields as the test hybrids of the inbred lines of Rutabaga-BF × A07-26NR despite seed yield of this inbred population being significantly lower among the two populations. The high level of seed yield in this test hybrid population apparently resulted from the higher level of MPH and CPH, and this might have resulted from the favorable heterotic alleles introgressed from Rutabaga-BF into the inbred lines of Rutabaga-BF × Hi-Q.

Radoev et al. (2008), reported that diverse types of dominance and epistasis interaction are

involved in the control of heterosis in *B. napus*. According to Lefort-Buston et al. (1987b), Ali et al. (1995) and Ahmad et al. (2011), genetic divergence between the parents correlates well with heterosis for seed yield in *B. napus*. In contrast, Diers et al. (1996) and Qian et al. (2007) argued that heterosis in *B. napus* cannot be precisely predicted based on parental genetic distance alone. In the present study, correlation between genetic divergence of the inbred lines of Rutabaga-BF × Hi-Q and MPH was not significant for seed yield. This result, as well as the results reported by Diers et al. (1996) and Qian et al. (2007) indicate that not all but only a few of the exotic alleles contribute to MPH for seed yield. Quijada et al. (2006) and Rahman et al. (2016) also found high heterosis in test hybrids derived from crossing spring *B. napus* to the spring *B. napus* lines carrying alleles of winter *B. napus*.

The test hybrid populations of the inbred lines of Rutabaga × spring canola showed about 2% negative MPH for days to flowering. The occurrence of negative MPH for this trait apparently resulted from the dominance effect or epistatic interaction (Long et al., 2007) of the genes. Rahman et al. (2016) also reported negative MPH for days to flowering in *B. napus* test hybrids carrying *B. oleracea* alleles. Therefore, it can be assumed that advantageous alleles for earliness of flowering can be found in Rutabaga which can contribute to a negative heterosis for this trait.

Non-significant MPH for days to maturity suggests that the non-additive type of gene effect was predominantly involved in the control of this trait. For plant height, the test hybrid population of Rutabaga-BF × Hi-Q displayed, on average, a negative MPH while the population of Rutabaga-BF × A07-26NR showed a positive MPH. Mid-parent heterosis for plant height has also been reported by Rahman et al. (2016) in a test hybrid population developed based on spring *B. napus* canola inbred lines, carrying alleles of winter *B. napus* canola, crossed to spring *B. napus* canola.

On average, very negligible heterosis was detected for seed oil content in the two hybrid populations; however, about 2% of the test hybrids showed about 2 - 4% positive heterosis for this trait. In case of seed protein content, the opposite direction of MPH, as compared to seed oil content, was found; this apparently resulted from a strong negative correlation between these two traits in *B. napus* (Grami et al., 1977; Zhao et al., 2006). Similar MPH for seed oil content (0.5%) and negative MPH for seed protein content (-1.1%) has also been reported by Rahman et al. (2016) while working with spring *B. napus* inbred lines carrying winter *B. napus* canola alleles. The greater positive MPH for seed oil content in the test hybrid population of Rutabaga-BF × Hi-Q as compared to the test hybrid population of Rutabaga-BF × A07-26NR could be due to introgression of a greater number of favorable alleles from Rutabaga into the inbred lines of the former cross, as evident from the extent of Rutabaga alleles detected in this inbred population (Table 2.19). Seed glucosinolate content in the test hybrid population varied from 11.2 to 24.4 µmol/g; slightly negative MPH was detected in all test hybrid populations.

The greater MPH for days to flowering and seed yield observed in the test hybrid population based on the BC_1-derived inbred lines as compared to the F_2-derived inbred lines (Tables 3.3 and 3.13) might have resulted from the new and favorable allelic combination created by the Rutabaga and *B. napus* cvs. Hi-Q and A07-26NR alleles in the BC_1-derived inbred lines. According to Falk (2010), the frequently observed poor performance of the inbred lines derived from crosses involving genetically diverse germplasm is due to the disruption of favorable allele combinations of the elite lines, which has been created over cycles of breeding. This disruption is, theoretically, expected to occur to a greater extent in the F_2-derived inbred lines than the BC_1-derived lines; this might have also contributed to this increased MPH in the test hybrids of the BC_1-derived inbred lines.

In conclusion, the allelic diversity introgressed from Rutabaga into spring *B. napus* canola contributed to heterosis for seed yield in this oilseed crop. This study demonstrated the potential value of this gene pool for use in the breeding of high-yielding hybrid spring *B. napus* canola cultivars.

Chapter 4

General discussion and conclusions

4.1 General discussion

The *Brassica* oilseed is the second largest oilseed crop in the world (FAOSTAT, 2016a). This crop is one of the most important elements of the Canadian agricultural economy (Canola Council of Canada, 2016d). Among the different *Brassica* oilseed crop species, *B. napus* canola is the most important one in the world (Hayward, 2012). In Canada, the spring type of *B. napus* canola constitutes more than 95% of the total canola acreage, and about 90% of the Canadian canola is exported as seed or as oil and meal to other countries (Canola Council of Canada, 2016d). Annual production of this crop in Canada has been escalating in the past decade sharply, reflecting the global demand for this crop (McVetty et al., 2016). Further increase in the production of *B. napus* canola mainly relies on the development of new cultivars with higher seed yield (Hayward, 2012); for this, divergence in the breeding materials is needed (for review, see Rahman, 2013). The limited genetic variability present today in the gene pool of this crop has primarily resulted from the bottleneck of selection for canola quality type (Bus et al., 2011); this type of genetically narrow gene pool is also considered a hindrance for the development of genetically distinct heterotic groups for exploitation of the phenomenon known as heterosis in hybrid cultivars (Bonnema, 2012).

Attempts have been made to broaden the genetic base of the spring *B. napus* canola by the use of the genetically distinct germplasms of its primary gene pool, such as winter and semi-winter *B. napus* (Quijada et al., 2004; Qian et al., 2007; Kebede et al., 2010), as well as the secondary gene pool, such as *B. rapa* and *B. oleracea* (Qian et al., 2005; Bennett et al., 2012; Rahman et al., 2011).

The resynthesized *B. napus*, created from crossing of the two parental diploid species (secondary gene pools), *B. rapa* and *B. oleracea*, has also been used to broaden the genetic base of the spring *B. napus* gene pool (Girke et al., 2012a). The use of the diploid parental species or resynthesized *B. napus* for the improvement of *B. napus* canola often introduces several undesirable traits due to linkage disequilibrium (Qian et al., 2005; Jesske et al., 2013; Rahman et al., 2015); intensive breeding is, therefore, needed for use of the alleles of these allied species in the breeding of spring *B. napus* canola (Jesske et al., 2013; for review, see Rahman, 2013).

The potential of the primary gene pools, such as winter and semi-winter forms of *B. napus*, for broadening the genetic base of spring *B. napus* canola has been demonstrated by several researchers (Quijada et al., 2004; Qian et al., 2007; Kebede et al., 2010; Rahman and Kebede, 2012; Rahman et al., 2016). The potential of the Rutabaga (*B. napus* var. *napobrassica*) gene pool for broadening the genetic base of spring *B. napus* canola, however, has not been studied well. This variant of *B. napus* is quite distinct from spring canola (Bus et al., 2011) and has showed the potential of contributing clubroot resistance trait for the development of clubroot resistant oilseed *B. napus* (Lüders et al., 2011; Rahman et al., 2014; Hasan and Rahman, 2016).

In the present study, genetic diversity analysis revealed that the unique alleles of Rutabaga can diversify both the A and C genomes of spring *B. napus* canola. The genetic base of the C genome in spring *B. napus* is known to be narrower than the A genome (Bus et al., 2011). In this regard, the Rutabaga gene pool has demonstrated the potential of broadening the genetic base of this genome of spring *B. napus* through contributing new alleles.

Wide variation was found in both BC_1- and F_2-derived inbred lines of the Rutabaga × spring *B. napus* canola crosses for both allelic diversity, and agronomic and seed quality traits. Obviously,

the Rutabaga alleles, present in the inbred lines, have contributed to the improvement of some of the important traits, such as seed yield and oil content. This is evident from the fact that a number of lines performed better than the spring canola parent and also had higher oil content. This could have also resulted from novel non-allelic interaction of the Rutabaga and spring canola alleles of the A and C genomes. Advantageous non-allelic interaction of exotic and native *B. napus* alleles in *B. napus* inbred lines carrying genome contents of *B. rapa* has been reported by Fu et al. (2012). Rutabaga may have also contributed the co-localized desired and undesired QTL alleles or contributed pleiotropic alleles in the spring canola lines derived from the Rutabaga × spring canola crosses. This is evident from strong negative correlations found between seed yield and days to flowering or seed oil and protein content. Introduction of favorable alleles of Rutabaga, that are independent of other alleles, might also have occurred as some of the high-yielding inbred lines also showed early flowering and increased seed oil and protein contents. Independent QTL alleles for oil and protein content have also been reported by Zhao et al. (2006).

Introgression of Rutabaga alleles in spring *B. napus* canola inbred lines has contributed to superior performance of the test hybrids. These alleles may have also resulted in a new heterotic group; however, this needs to be confirmed by producing test hybrids by crossing with different genetically distinct canola lines. Exotic alleles of winter and semi-winter types *B. napus* contributing to heterosis in spring canola have been demonstrated by Quijada et al. (2004), Udall et al. (2004), Qian et al. (2007) and Rahman et al. (2016). More than 90% of the test hybrids of the inbred lines in this study showed positive MPH for seed yield. This might have resulted from the introduction of advantageous alleles of Rutabaga, which in combination with spring canola alleles exerting non-additive effects (e.g. overdominance) to the phenomenon heterosis. The

contribution of non-additive effect of the genes to heterosis in *B. napus* has been reported by several researchers (Long et al., 2007; Radoev et al., 2008; Rahman et al., 2016).

4.2 Conclusions

The following conclusions were drawn from the present study:

- The genetic base of spring *B. napus* canola can be broadened using the alleles of the A and C genomes of Rutabaga. Introgression of allelic diversity from Rutabaga into spring *B. napus* canola was confirmed by use of SSR markers.
- Genetically distinct spring *B. napus* canola lines can be developed from both F_2- and BC_1-derived populations of Rutabaga × spring *B. napus* canola crosses.
- Favorable combinations of Rutabaga and spring *B. napus* canola alleles can be achieved due to independent assortment of the alleles, and this can result in improved agronomic and seed quality traits, such as high seed yield and increased seed oil content.
- Allelic diversity introgressed from Rutabaga can contribute to heterosis in spring *B. napus* canola. This was evident from the fact that the majority of the test hybrids of the F_2- and BC_1-derived inbred lines displayed positive MPH for seed yield, and about 50% of the test hybrids surpassed seed yield of the *B. napus* parent (Hi-Q or A07-26NR).

4.3 Future research

The inbred lines developed in this research can be used to produce test hybrids with other spring *B. napus* canola lines to evaluate the level of heterosis that can be achieved for different

agronomic and seed quality traits by using the Rutabaga gene pool in breeding, as well as to assess the general combining ability (GCA) of these lines. By use of SNP markers, QTL mapping of the genome regions involved in the control of agronomic and seed quality traits, can be performed, as well as the genomic regions contributing to heterosis can be identified.

Breeding for clubroot resistance is one of the most important objectives of canola breeding today (Rahman et al., 2014). The Rutabaga parent used in this study is known to be resistant to this disease (Hasan and Rahman, 2016); therefore, some of the inbred lines developed in this project are expected to carry resistance to clubroot disease. These lines can be evaluated for clubroot resistance to identify the resistant lines.

The spring canola populations derived from the Rutabaga × *B. napus* spring canola crosses, on average, flowered and matured later than the spring canola parent; further breeding would be needed to improve these traits.

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Appendices

Table A2.1: List of SSR markers used for genotyping of the BC_1- and F_2-derived inbred lines of the Rutabaga-BF × Hi-Q cross.

Row	Primer name	Linkage group	Primer number	Source	Total alleles	Alleles specific to Rutabaga-BF
1	sN11641	A01, C1	257	AAFC	3	1
2	Na14F11	A01	343	BBSRC microsatellite programme	2	1
3	CB10369	A01	626	Celera AgGen Brassica consortium	3	1
4	NRC-929	A01	929	NRC-Saskatoon	2	0
5	sN0990 (bNP)	A01	2001	AAFC	2	1
6	A01_10214	A01	2711	Canola Program (The University of Alberta)	3	1
7	sN3761	A02,C2	315	AAFC	6	0
8	MR 144	A02	916	IPB (University of Goettingen)	5	2
9	sN3672R	A02	2009	AAFC	3	2
10	A02_497	A02	2712	Canola Program (The University of Alberta)	1	1
11	A02_6263	A02	2714	Canola Program (The University of Alberta)	2	1
12	A02_13002	A02	2717	Canola Program (The University of Alberta)	2	1
13	Na12-E02	A03	461	Ukcrop.net	2	1
14	1188F	A03	1188	Canola Program (The University of Alberta)	2	1
15	A03_6890	A03	2721	Canola Program (The University of Alberta)	2	1
16	KB59N07	A03	2821	10.1270/jsbbs.63.116	4	2
17	B4732	A03	2826	10.1270/jsbbs.63.116	2	1
18	A03_12778	A03	2850	B.rapa Chifu assembly v1.5	2	0
19	sN0412(a)	A04,C4	217	AAFC	3	2
20	SN11516	A04,C4	273	AAFC	4	2
21	BRMS-195	A04	963	Suwabe et al (2006) Genetics.(173) 309-319	2	1
22	sN0786	A04	2491	AAFC	2	1
23	sN8093I	A04	2495	AAFC	5	0
24	A04_2310	A04	2725	Canola Program (The University of Alberta)	2	1
25	A04_5262	A04	2727	Canola Program (The University of Alberta)	3	1
26	BRAS072A	A05	355	Celera AgGen Brassica Consortium	4	2
27	CB10080	A05	357	Celera AgGen Brassica Consortium	2	1
28	BnGMS293	A05	559	Cheng et al TAG 118:1121-1131	2	1
29	A05_4730	A05	2732	Canola Program (The University of Alberta)	2	1
30	A05_10574	A05	2735	Canola Program (The University of Alberta)	4	1
31	sN1958 (bNM)	A06	2025	AAFC	2	1
32	sN1939 F(b)	A06	2026	AAFC	3	1
33	sN1503 (bNP)	A06	2036	AAFC	3	1
34	sN3678 (a)	A06	2041	AAFC	2	1

Table A2.1: Continued.

Row	Primer name	Linkage group	Primer number	Source	Total alleles	Alleles specific to Rutabaga-BF
35	sS1949 (a)	A06	2044	AAFC	3	1
36	sN3603 (a)	A06	2053	AAFC	2	1
37	A06_2300	A06	2737	Canola Program (The University of Alberta)	2	1
38	sN12131	A07	92	AAFC	3	1
39	sN2318R	A07	111	AAFC	3	1
40	sR0282R	A07	281	AAFC	2	1
41	sNRA59	A07	285	AAFC	2	1
42	BRMS-036	A07	394	Suwabe ert al. Theor Appl Genet (2002) 104:1092–1098	4	1
43	CB10278	A07	483	Celera AgGen Brassica Consortium	3	0
44	A07_5462	A07	2743	Canola Program (The University of Alberta)	2	1
45	sR3688	A08	288	AAFC	3	1
46	BRMS-070	A08	971	Suwabe et al (2006) Genetics.(173) 309-319	2	1
47	sNRF19	A08	2336	AAFC	2	1
48	sN0809	A08	2343	AAFC	2	1
49	sN10840	A08	2352	AAFC	2	1
50	A08_4614	A08	2561	Canola Program (The University of Alberta)	2	1
51	A08_7860	A08	2752	Canola Program (The University of Alberta)	2	1
52	A08_4450	A08	2766	Canola Program (The University of Alberta)	3	1
53	NRC-778	A09	778	NRC-Saskatoon	2	1
54	NRC-864	A09	864	NRC-Saskatoon	2	1
55	NRC-869	A09	869	NRC-Saskatoon	3	1
56	NRC-1008	A09	1008	NRC-Saskatoon	2	1
57	NRC-1029	A09	1029	NRC-Saskatoon	3	1
58	NRC-1048	A09	1048	NRC-Saskatoon	2	1
59	A09_17847	A09	2759	Canola Program (The University of Alberta)	2	1
60	CB10079A	A10	441	Celera AgGen Brassica Consortium	4	1
61	Na12-H04	A10	637	Celera AgGen Brassica consortium	2	1
62	A10_109	A10	2760	Canola Program (The University of Alberta)	2	1
63	A10_2396	A10	2761	Canola Program (The University of Alberta)	2	1
64	A10_5336	A10	2763	Canola Program (The University of Alberta)	4	1
65	A10_7834	A10	2765	Canola Program (The University of Alberta)	4	2
66	sS2136	C1	2277	AAFC	2	1
67	sN1958	C1	2294	AAFC	2	1
68	sN12790	C1	2302	AAFC	2	1
69	sS1725	C1	2307	AAFC	4	1
70	sS2268 (bNP)	C2	2065	AAFC	2	1
71	sORB56 (aNP)	C2	2079	AAFC	2	1
72	sN12639	C3	103	AAFC	2	1

Table A2.1: Continued.

Row	Primer name	Linkage group	Primer number	Source	Total alleles	Alleles specific to Rutabaga-BF
73	BnGMS273	C3	659	Cheng et al TAG 118:1121-1131	2	1
74	BoGMS0819	C3	1082	Li et al. 2010 Mol Breeding	2	1
75	Na10D11	C5	983	BBSRC microsatellite programme	2	1
76	Ol10B02	C5	988	BBSRC microsatellite programme	2	1
77	sN2046R	C5	2446	AAFC	2	1
78	sN12572	C5	2464	AAFC	2	1
79	CB10234	C6	735	Celera AgGen Brassica consortium	2	1
80	sR0472	C6	2362	AAFC	2	1
81	sN11904	C6	2365	AAFC	2	1
82	MD 50	C7	889	IPB (University of Goettingen)	2	1
83	BnGMS4	C8	2180	Cheng et al. 2009 TAG	2	1
84	BoGMS0468	C8	2244	Li et al. 2011 Mol Breed	2	1
85	BoGMS0631	C8	2245	Li et al. 2011 Mol Breed	2	1
86	CB10064	C9	914	Celera AgGen Brassica Consortium	2	1
87	BoGMS1283	C9	2258	Li et al. 2011 Mol Breed	2	1

Table A2.2: List of SSR markers used for genotyping of the BC_1- and F_2-derived inbred lines of the Rutabaga-BF × A07-26NR cross.

Row	Primer name	Linkage group	Primer number	Source	Total alleles	Alleles specific to Rutabaga-BF
1	Na14F11	A01	343	BBSRC microsatellite programme	4	1
2	CB10369	A01	626	Celera AgGen Brassica consortium	3	1
3	NRC-929	A01	929	NRC-Saskatoon	3	1
4	sN0990 (bNP)	A01	2001	AAFC	7	2
5	A01_6237	A01	2708	Canola Program (The University of Alberta)	5	2
6	sN3761	A02,C2	315	AAFC	3	2
7	BRMS-082	A02	956	Suwabe et al (2006) Genetics.(173) 309-319	2	1
8	sN3672R	A02	2009	AAFC	3	2
9	A02_497	A02	2712	Canola Program (The University of Alberta)	1	1
10	A02_6263	A02	2714	Canola Program (The University of Alberta)	2	1
11	A02_13002	A02	2717	Canola Program (The University of Alberta)	5	2
12	sNRG67	A03	160	AAFC	3	1
13	Na12-E02	A03	461	Ukcrop.net	3	2
14	1188F	A03	1188	Canola Program (The University of Alberta)	3	1
15	A03_5090	A03	2720	Canola Program (The University of Alberta)	3	1
16	A03_12095	A03	2722	Canola Program (The University of Alberta)	3	1
17	B4732	A03	2826	Canola Program (The University of Alberta)	1	0
18	SN11516	A04,C4	273	AAFC	4	2
19	CB10045B	A04	438	Celera AgGen Brassica Consortium	2	1
20	sN11639	A04	2487	AAFC	2	1
21	sN0786	A04	2491	AAFC	2	1
22	A04_2310	A04	2725	Canola Program (The University of Alberta)	3	1
23	A04_5262	A04	2727	Canola Program (The University of Alberta)	4	2
24	CB10080	A05	357	Celera AgGen Brassica Consortium	3	1
25	BnGMS293	A05	559	Cheng et al TAG 118:1121-1131	3	2
26	sN12572	A05	2464	AAFC	2	0
27	A05_1116	A05	2730	Canola Program (The University of Alberta)	3	1
28	A05_4730	A05	2732	Canola Program (The University of Alberta)	3	0
29	A05_10574	A05	2735	Canola Program (The University of Alberta)	2	1
30	sN1939 F(b)	A06	2026	AAFC	3	1
31	sN1503 (bNP)	A06	2036	AAFC	3	1
32	sN3678 (a)	A06	2041	AAFC	3	1
33	sN3760a	A06	2051	AAFC	2	1
34	BnGMS205	A06	2191	Cheng et al. 2009 TAG	4	2

Table A2.2: Continued.

Row	Primer name	Linkage group	Primer number	Source	Total alleles	Alleles specific to Rutabaga-BF
35	A06_4740	A06	2738	Canola Program (The University of Alberta)	2	1
36	sN12131	A07	92	AAFC	3	2
37	sN2318R	A07	111	AAFC	3	1
38	sN4026	A07	128	AAFC	1	1
39	sNRA59	A07	285	AAFC	3	1
40	BRMS-018	A07	495	Suwabe ert al. Theor Appl Genet (2002) 104:1092–1098	5	1
41	A07_5462	A07	2743	Canola Program (The University of Alberta)	4	1
42	sR3688	A08	288	AAFC	3	1
43	BRMS-070	A08	971	Suwabe et al (2006) Genetics.(173) 309-319	2	1
44	sNRF19	A08	2336	AAFC	2	1
45	sN10692I	A08	2338	AAFC	2	1
46	sN0809	A08	2343	AAFC	2	1
47	sR0841	A08	2348	AAFC	3	2
48	NRC-767	A09	767	NRC-Saskatoon	3	1
49	NRC-778	A09	778	NRC-Saskatoon	2	1
50	NRC-864	A09	864	NRC-Saskatoon	2	1
51	NRC-1029	A09	1029	NRC-Saskatoon	3	2
52	NRC-1048	A09	1048	NRC-Saskatoon	4	1
53	A09_17847	A09	2759	Canola Program (The University of Alberta)	2	1
54	CB10524	A10	487	Celera AgGen Brassica Consortium	1	1
55	Na12-H04	A10	637	Celera AgGen Brassica consortium	2	1
56	A10_109	A10	2760	Canola Program (The University of Alberta)	2	1
57	A10_2396	A10	2761	Canola Program (The University of Alberta)	3	1
58	A10_5336	A10	2763	Canola Program (The University of Alberta)	2	1
59	BoGMS0789	C1	2226	Li et al. 2011 Mol Breed	4	2
60	sN1958	C1	2294	AAFC	5	2
61	sN12790	C1	2302	AAFC	2	1
62	sS1725	C1	2307	AAFC	6	3
63	sN11675	C1	2310	AAFC	4	2
64	sR1464	C1	2327	AAFC	3	1
65	BRAS011	C2	723	Celera AgGen Brassica consortium	5	1
66	Ol13-G05	C2	763	BBRC	2	1
67	sN3732	C2	2060	AAFC	2	1
68	sN1937 (aNP)	C2	2063	AAFC	3	2
69	sORB56 (aNP)	C2	2079	AAFC	4	1
70	sN11730	C3	233	AAFC	2	1
71	sN1844(a)	C3	236	AAFC	2	1
72	BnGMS575	C3	657	Cheng et al TAG 118:1121-1131	2	1

Table A2.2: Continued.

Row	Primer name	Linkage group	Primer number	Source	Total alleles	Alleles specific to Rutabaga-BF
73	BnGMS273	C3	659	Cheng et al TAG 118:1121-1131	4	3
74	BnGMS305	C3	662	Cheng et al TAG 118:1121-1131	3	1
75	BoGMS0616	C3	1086	Li et al. 2010 Mol Breeding	2	1
76	sN0276	C4	82	AAFC	2	1
77	CB10493	C4	994	Celera AgGen Brassica Consortium	1	0
78	MR 229	C4	999	IPB (University of Goettingen)	5	2
79	MR 36	C4	1001	IPB (University of Goettingen)	2	1
80	BoGMS1131	C4	2234	Li et al. 2011 Mol Breed	2	1
81	Na10D11	C5	983	BBSRC microsatellite programme	3	2
82	sORB17	C5	2445	AAFC	4	2
83	sN2046R	C5	2446	AAFC	2	1
84	sNRC18	C5	2451	AAFC	2	1
85	sORF31	C5	2456	AAFC	3	1
86	sORH47	C5	2457	AAFC	2	1
87	CB10234	C6	735	Celera AgGen Brassica consortium	2	1
88	sR0472	C6	2362	AAFC	2	1
89	sN1451	C6	2363	AAFC	2	0
90	sN11904	C6	2365	AAFC	4	2
91	sORB31	C6	2367	AAFC	2	1
92	sN12743J	C6	2379	AAFC	7	2
93	BRMS-042	C7	396	Suwabe ert al. Theor Appl Genet (2002) 104:1092–1098	2	1
94	MD 50	C7	889	IPB (University of Goettingen)	4	1
95	sN12940	C7	2385	AAFC	2	1
96	CB10139	C8	489	Celera AgGen Brassica Consortium	1	0
97	BnGMS4	C8	2180	Cheng et al. 2009 TAG	4	2
98	BoGMS0468	C8	2244	Li et al. 2011 Mol Breed	5	4
99	BoGMS0631	C8	2245	Li et al. 2011 Mol Breed	1	1
100	CB10064	C9	914	Celera AgGen Brassica Consortium	4	1
101	BnGMS43	C9	2182	Cheng et al. 2009 TAG	3	1
102	BnGMS213	C9	2193	Cheng et al. 2009 TAG	2	1
103	BnGMS625	C9	2220	Cheng et al. 2009 TAG	3	1
104	BoGMS0624	C9	2256	Li et al. 2011 Mol Breed	2	1
105	BoGMS1283	C9	2258	Li et al. 2011 Mol Breed	3	1

Table A3.1: Days to flowering (DTF) of the test hybrids (TH), produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) for this trait in 2014, 2015 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	2014			2015			2016		
		TH (days)	MPH (%)	CPH (%)	TH (days)	MPH (%)	CPH (%)	TH (days)	MPH (%)	CPH (%)
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Rut-BF × Hi-Q[2]	BC	45.1 ± 0.4 a	-3.5 ± 0.7 a	-1.4 ± 1.2 a	57.0 ± 0.6 a	-4.1 ± 0.7 b	-0.1 ± 1.0 a	47.7 ± 0.3 a	0.3 ± 0.5 ab	2.8 ± 0.6 ab
	F	45.1 ± 0.3 a	-4.0 ± 0.5 a	-0.8 ± 1.2 a	57.2 ± 0.7 a	-3.0 ± 0.7 ab	-1.4 ± 1.1 a	48.2 ± 0.3 a	1.7 ± 0.5 a	4.5 ± 0.6 a
Rut-BF × 26NR[3]	BC	45.0 ± 0.3 a	-3.9 ± 0.5 a	-2.5 ± 1.6 a	56.9 ± 0.7 a	-2.1 ± 0.7 ab	1.1 ± 1.1 a	45.1 ± 0.3 b	-2.5 ± 0.5 c	0.8 ± 0.6 b
	F	45.1 ± 0.3 a	-2.9 ± 0.6 a	-3.9 ± 1.5 a	57.6 ± 0.7 a	-0.3 ± 0.7 a	1.1 ± 1.1 a	45.1 ± 0.3 b	-0.6 ± 0.5 b	1.1 ± 0.6 b
Hi-Q[4]		45.9 ± 0.6 a			57.5 ± 0.8 a			46.2 ± 0.4 b		
A07-26NR[4]		46.4 ± 0.6 a			56.8 ± 0.9 a			44.8 ± 0.4 b		
Rut-BF × Hi-Q		45.1 ± 0.2 a	-3.7 ± 0.4 a	-1.4 ± 0.6 a	57.1 ± 0.4 a	-3.6 ± 0.6 b	-0.7 ± 0.6 a	47.9 ± 0.2 a	1.0 ± 0.3 a	3.7 ± 0.4 a
Rut-BF × 26NR		45.1 ± 0.2 a	-3.4 ± 0.5 a	-2.8 ± 0.7 a	57.3 ± 0.5 a	-1.2 ± 0.6 a	1.1 ± 0.7 a	45.1 ± 0.3 b	-1.6 ± 0.4 b	1.0 ± 0.5 b
BC[5]		45.1 ± 0.2 a	-3.6 ± 0.5 a	-2.2 ± 0.6 a	56.9 ± 0.4 a	-3.2 ± 0.7 a	0.4 ± 0.8 a	46.5 ± 0.4 a	-0.9 ± 0.5 b	1.9 ± 0.5 a
F[6]		45.1 ± 0.2 a	-3.6 ± 0.5 a	-1.9 ± 0.5 a	57.5 ± 0.4 a	-1.7 ± 0.8 a	-0.2 ± 0.9 a	46.7 ± 0.4 a	0.6 ± 0.5 a	2.9 ± 0.5 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Table A3.2: Days to maturity (DTM) of the test hybrids (TH), produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) for this trait in 2014, 2015 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	2014			2015			2016		
		TH (days)	MPH (%)	CPH (%)	TH (days)	MPH (%)	CPH (%)	TH (days)	MPH (%)	CPH (%)
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Rut-BF × Hi-Q[2]	BC	95.5 ± 0.7 a	-1.6 ± 0.7 a	-1.3 ± 0.7 a	107.3 ± 2.0 a	-1.3 ± 0.6 a	-0.4 ± 0.6 a	108.9 ± 0.7 a	1.5 ± 0.5 a	3.4 ± 0.5 a
	F	95.4 ± 0.5 a	-2.0 ± 0.7 a	-1.3 ± 0.6 a	105.5 ± 2.1 a	-2.1 ± 0.6 a	-0.6 ± 0.7 a	108.7 ± 0.7 a	1.2 ± 0.5 a	3.0 ± 0.5 a
Rut-BF × 26NR[3]	BC	93.8 ± 0.6 a	-1.8 ± 0.7 a	-1.1 ± 0.6 a	106.4 ± 2.1 a	0.0 ± 0.6 a	-0.1 ± 0.7 a	102.7 ± 0.8 bc	-1.9 ± 0.5 b	0.7 ± 0.6 b
	F	94.3 ± 0.6 a	-1.1 ± 0.7 a	-0.6 ± 0.6 a	106.3 ± 2.1 a	-0.1 ± 0.6 a	0.5 ± 0.7 a	102.9 ± 0.8 bc	-0.6 ± 0.5 b	1.4 ± 0.6 ab
Hi-Q[4]		96.6 ± 1.1 a			107.1 ± 2.2 a			105.6 ± 0.9 b		
A07-26NR[4]		94.6 ± 1.1 a			106.1 ± 2.3 a			102.0 ± 1.0 c		
Rut-BF × Hi-Q		95.4 ± 0.5 a	-1.8 ± 0.6 a	-1.3 ± 0.5 a	106.5 ± 1.9 a	-1.7 ± 0.5 b	-0.5 ± 0.4 a	108.8 ± 0.7 a	1.3 ± 0.4 a	3.2 ± 0.4 a
Rut-BF × 26NR		94.0 ± 0.5 a	-1.4 ± 0.6 a	-0.9 ± 0.5 a	106.3 ± 2.0 a	-0.1 ± 0.5 a	0.2 ± 0.4 a	102.8 ± 0.7 b	-1.3 ± 0.4 b	1.1 ± 0.4 b
BC[5]		95.0 ± 0.6 a	-1.7 ± 0.6 a	-1.2 ± 0.5 a	106.9 ± 1.9 a	-0.7 ± 0.6 a	-0.3 ± 0.5 a	106.0 ± 1.0 a	-0.2 ± 0.5 a	2.1 ± 0.5 a
F[6]		94.7 ± 0.6 a	-1.6 ± 0.6 a	-1.0 ± 0.5 a	105.9 ± 1.9 a	-1.1 ± 0.7 a	-0.1 ± 0.5 a	106.0 ± 1.0 a	0.4 ± 0.5 a	2.3 ± 0.5 b

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Table A3.3: Plant height (cm) of the test hybrids (TH), produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) for this trait in 2014, 2015 and 2016 trials.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	2014			2015			2016		
		TH (cm)	MPH (%)	CPH (%)	TH (cm)	MPH (%)	CPH (%)	TH (cm)	MPH (%)	CPH (%)
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Rut-BF × Hi-Q[2]	BC	119.4 ± 3.2 bc	-3.4 ± 2.6 a	-5.9 ± 2.3 c	108.8 ± 2.0 a	-2.9 ± 1.2 a	-3.7 ± 1.6 a	144.7 ± 1.4 b	5.8 ± 0.6 a	3.4 ± 0.7 b
	F	123.3 ± 3.1 b	-0.1 ± 2.5 a	-1.0 ± 1.8 bc	107.3 ± 2.2 a	-4.3 ± 1.2 a	-4.5 ± 1.6 a	149.0 ± 1.4 a	6.2 ± 0.6 a	6.0 ± 0.7 a
Rut-BF × 26NR[3]	BC	112.2 ± 4.0 bcd	3.6 ± 3.6 a	4.1 ± 2.1 a	101.4 ± 2.3 a	-1.0 ± 1.3 a	-0.3 ± 1.8 a	137.2 ± 1.5 c	1.9 ± 0.7 b	1.9 ± 0.8 b
	F	108.5 ± 3.6 d	-1.8 ± 3.2 a	-0.3 ± 2.3 ab	103.5 ± 2.3 a	-1.1 ± 1.3 a	-0.5 ± 1.7 a	137.0 ± 1.5 c	2.4 ± 0.7 b	1.8 ± 0.8 b
Hi-Q[4]		126.3 ± 4.1 ab			113.2 ± 3.1 a			140.4±1.8 bc		
A07-26NR[4]		109.6 ± 4.0 cd			103.3 ± 3.5 a			135.2 ± 1.9 c		
Rut-BF × Hi-Q		121.4 ± 2.4 a	-1.9 ± 1.3 a	-2.9 ± 1.4 a	108.1 ± 1.4 a	-3.6 ± 0.9 a	-4.1 ± 1.0 b	146.9 ± 1.3 a	6.0 ± 0.4 a	4.7 ± 0.5 a
Rut-BF × 26NR		110.9 ± 2.5 b	1.7 ± 1.5 a	1.8 ± 1.6 a	102.5 ± 1.5 a	-1.1 ± 1.0 a	-0.4 ± 1.1 a	137.1 ± 1.3 b	2.2 ± 0.5 b	1.9 ± 0.6 b
BC[5]		117.5 ± 3.5 a	-1.2 ± 2.1 a	-3.4 ± 2.6 a	105.8 ± 1.9 a	-2.1 ± 1.0 a	-2.2 ± 1.4 a	141.4 ± 1.7 a	4.0 ± 0.6 a	2.7 ± 0.6 a
F[6]		116.4 ± 3.3 a	0.2 ± 1.9 a	0.8 ± 2.4 a	105.4 ± 2.1 a	-2.8 ± 1.0 a	-2.6 ± 1.4 a	143.1 ± 1.7 a	4.3 ± 0.6 a	4.0 ± 0.6 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Table A3.4: Seed yield (kg/ha) of the test hybrids (TH), produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) for this trait in 2015 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	2015			2016		
		TH (kg/ha)	MPH (%)	CPH (%)	TH(kg/ha)	MPH (%)	CPH (%)
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Rut-BF × Hi-Q[2]	BC	4195.5 ± 235.6 ab	27.6 ± 3.2 a	12.3 ± 3.4 a	5485.5 ± 174.7 bc	23.2 ± 2.2 a	16.6 ± 2.3 a
	F	4441.2 ± 237.5 a	23.3 ± 3.1 a	9.4 ± 3.4 a	5282.5 ± 175.4 c	20.3 ± 2.2 ab	12.1 ± 2.3 a
Rut-BF × 26NR[3]	BC	3438.8 ± 242.1 c	16.6 ± 3.7 ab	2.6 ± 3.7 ab	6218.9 ± 191.5 a	20.8 ± 2.5 ab	14.6 ± 2.6 a
	F	3664.9 ± 239.7 bc	-0.3 ± 3.1 b	-10.3 ± 3.5 b	6048.2 ± 188.9 ab	13.8 ± 2.4 b	10.6 ± 2.5 a
Hi-Q[4]		3865.4 ± 294.2 abc			4774.5 ± 204.2 d		
A07-26NR[4]		3709.7 ± 311.2 abc			5860.1 ± 212.5 abc		
Rut-BF × Hi-Q		4310.7 ± 228.7 a	25.6 ± 3.6 a	10.9 ± 2.9 a	5386.4 ± 160.9 b	21.7 ± 1.6 a	14.3 ± 1.7 a
Rut-BF × 26NR		3553.7 ± 232.5 b	7.5 ± 3.9 b	-4.1 ± 3.0 b	6127.8 ± 170.4 a	17.2 ± 1.7 a	12.5 ± 1.9 a
BC[5]		3968.8 ± 281.6 a	22.9 ± 5.1 a	8.2 ± 4.4 a	5833.4 ± 178.9 a	22.1 ± 1.7 a	15.7 ± 1.7 a
F[6]		3969.1 ± 291.0 a	11.5 ± 5.4 a	-0.3 ± 4.6 a	5628.2 ± 178.6 a	17.3 ± 1.7 b	11.4 ± 1.7 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Table A3.5: Seed oil content (%) of the test hybrids (TH), produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) for this trait in 2014, 2015 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	2014			2015			2016		
		TH (%)	MPH (%)	CPH (%)	TH (%)	MPH (%)	CPH (%)	TH (%)	MPH (%)	CPH (%)
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Rut-BF × Hi-Q[2]	BC	46.1 ± 0.3 ab	1.5 ± 0.6 a	-0.6 ± 0.7 a	45.3 ± 0.2 a	2.7 ± 0.4 a	0.6 ± 0.7 a	47.9 ± 0.4 b	-0.9 ± 0.3 a	-0.8 ± 0.4 a
	F	45.9 ± 0.2 b	1.2 ± 0.4 a	-0.4 ± 0.5 a	45.2 ± 0.2 a	1.5 ± 0.4 ab	0.8 ± 0.7 a	47.8 ± 0.4 b	0.0 ± 0.3 a	-0.7 ± 0.4 a
Rut-BF × 26NR[3]	BC	45.9 ± 0.3 b	0.7 ± 0.5 a	-2.1 ± 0.5 a	44.1 ± 0.2 a	0.2 ± 0.5 b	-3.6 ± 0.7 b	49.8 ± 0.4 a	0.1 ± 0.3 a	-1.5 ± 0.5 a
	F	46.9 ± 0.3 a	0.1 ± 0.5 a	-0.4 ± 0.6 a	44.5 ± 0.2 a	-0.3 ± 0.5 b	-2.1 ±0.7 ab	50.2 ± 0.4 a	-0.1 ± 0.3 a	-0.6 ± 0.5 a
Hi-Q[4]		46.2 ± 0.6 ab			45.0 ± 0.5 a			48.3 ± 0.4 b		
A07-26NR[4]		46.6 ± 0.6 ab			45.6 ± 0.5 a			50.6 ± 0.4 a		
Rut-BF × Hi-Q		46.0 ± 0.3 a	1.3 ± 0.3 a	-0.6 ± 0.5 a	45.2 ± 0.1 a	2.1 ± 0.4 a	0.7 ± 0.5 a	47.9 ± 0.4 b	-0.4 ± 0.2 a	-0.7 ± 0.3 a
Rut-BF × 26NR		46.4 ± 0.3 a	0.4 ± 0.3 a	-1.0 ± 0.5 a	44.3 ± 0.2 b	-0.1 ± 0.4 b	-2.9 ± 0.5 b	50.0 ± 0.4 a	0.0 ± 0.2 a	-1.0 ± 0.4 a
BC[5]		46.0 ± 0.3 a	1.0 ± 0.5 a	-1.4 ± 0.6 a	44.8 ± 0.3 a	1.7 ± 0.6 a	-1.1 ± 1.0 a	48.8 ± 0.4 a	-0.4 ± 0.2 a	-1.1 ± 0.3 a
F[6]		46.3 ± 0.3 a	0.8 ± 0.4 a	-0.3 ± 0.5 a	44.9 ± 0.3 a	0.6 ± 0.7 a	-0.7 ± 1.1 a	49.0 ± 0.4 a	0.0 ± 0.2 a	-0.6 ± 0.3 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Table A3.6: Seed protein content (%) of the test hybrids (TH), produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH,%) for this trait in 2014, 2015 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	2014			2015			2016		
		TH (%)	MPH (%)	CPH (%)	TH (%)	MPH (%)	CPH (%)	TH (%)	MPH (%)	CPH (%)
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Rut-BF × Hi-Q[2]	BC	27.9 ± 0.3 a	-1.5 ± 1.0 a	1.5 ± 1.3 a	27.2 ± 0.2 b	-3.0 ± 0.7 b	-1.5 ± 1.0 b	25.7 ± 0.4 a	-0.4 ± 0.4 a	-1.6 ± 0.6 b
	F	28.4 ± 0.2 a	-1.3 ± 0.8 a	0.7 ± 1.1 a	28.1 ± 0.2 ab	-1.2 ± 0.7 ab	0.8 ± 1.2 ab	26.2 ± 0.4 a	-0.7 ± 0.4 a	0.4 ± 0.6 ab
Rut-BF × 26NR[3]	BC	28.3 ± 0.2 a	-2.8 ± 0.9 a	0.2 ± 1.4 a	28.1 ± 0.2 ab	0.3 ± 0.8 ab	4.3 ± 1.2 ab	24.4 ± 0.4 b	0.2 ± 0.5 a	2.0 ± 0.7 a
	F	27.8 ± 0.2 a	-1.4 ± 1.0 a	-0.6 ± 1.4 a	28.8 ± 0.2 a	2.8 ± 0.8 a	6.1 ± 1.2 a	24.5 ± 0.4 b	0.7 ± 0.4 a	2.3 ± 0.7 a
Hi-Q[4]		28.0 ± 0.5 a			27.8 ± 0.3 ab			26.1 ± 0.4 a		
A07-26NR[4]		28.4 ± 0.5 a			27.1 ± 0.4 b			23.9 ± 0.4 b		
Rut-BF × Hi-Q		28.2 ± 0.2 a	-1.4 ± 0.7 a	0.7 ± 0.5 a	27.6 ± 0.2 a	-2.2 ± 0.7 b	-0.5 ± 0.8 b	25.9 ± 0.3 a	-0.5 ± 0.3 b	-0.6 ± 0.4 b
Rut-BF × 26NR		28.0 ± 0.2 a	-2.1 ± 0.8 a	0.0 ± 0.5 a	28.4 ± 0.3 a	1.6 ± 0.8 a	5.2 ± 0.9 a	24.4 ± 0.3 b	0.5 ± 0.3 a	2.2 ± 0.5 a
BC[5]		28.1 ± 0.2 a	-2.4 ± 0.4 a	0.4 ± 0.6 a	27.6 ± 0.2 a	-1.7 ± 1.0 a	0.8 ± 1.5 a	25.0 ± 0.4 a	-0.1 ± 0.3 a	-0.1 ± 0.6 b
F[6]		28.2 ± 0.2 a	-1.2 ± 0.4 a	0.4 ± 0.5 a	28.4 ± 0.2 a	0.8 ± 1.1 a	3.4 ± 1.7 a	25.4 ± 0.4 a	0.0 ± 0.3 a	1.3 ± 0.6 a

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Table A3.7: Seed glucosinolate content (µmol/g) of the test hybrids (TH), produced from crossing of Hi-Q/A07-26NR to the inbred lines derived from F_2 and BC_1 of the Rutabaga-BF × Hi-Q/A07-26NR crosses, and mid-parent heterosis (MPH, %) and heterosis over Hi-Q/A07-26NR (CPH, %) for this trait in 2014, 2015 and 2016 field trials.

Cross/*B. napus* parent/type of population[1]	Test hybrid population[7]	2014			2015			2016		
		TH (µmol/g)	MPH (%)	CPH (%)	TH (µmol/g)	MPH (%)	CPH (%)	TH (µmol/g)	MPH (%)	CPH (%)
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Rut-BF × Hi-Q[2]	BC	13.6 ± 0.6 b	-8.9 ± 2.4 a	-9.4 ± 2.6 b	14.1 ± 0.6 c	-5.6 ± 1.9 a	-5.2 ± 2.6 a	14.2 ± 0.3 ab	4.4 ± 1.4 a	7.6 ± 1.6 a
	F	15.5 ± 0.4 b	-4.1 ± 1.8 a	-3.2 ± 2.0ab	14.1 ± 0.7 c	-6.0 ± 2.0 a	-4.5 ± 2.8 a	14.3 ± 0.3 ab	-0.2 ± 1.4ab	2.7 ± 1.6 a
Rut-BF × 26NR[3]	BC	17.2 ± 0.5 a	-7.2 ± 2.0 a	1.7 ± 2.1 a	17.8 ± 0.7 ab	-6.1 ± 2.2 a	2.4 ± 3.1 a	15.0 ± 0.3 a	2.1 ± 1.5 ab	6.2 ± 1.8 a
	F	16.4 ± 0.5 ab	-5.5 ± 2.2 a	0.1 ± 2.2 a	17.0 ± 0.7 bc	-4.3 ± 2.1 a	5.6 ± 3.0 a	14.3 ± 0.3 ab	-1.0 ± 1.5 b	1.5 ± 1.7 a
Hi-Q[4]		15.0 ± 1.1 b			14.7 ± 0.9 bc			13.3 ± 0.4 b		
A07-26NR[4]		16.6 ± 1.1 ab			16.4 ± 1.0 bc			14.0 ± 0.5 ab		
Rut-BF × Hi-Q		14.8 ± 0.5 a	-5.9 ± 1.8 a	-5.4 ± 1.9 a	14.1 ± 0.4 b	-5.8 ± 1.3 a	-4.9 ± 1.7 b	14.2 ± 0.2 a	2.2 ± 1.1 a	5.2 ± 1.1 a
Rut-BF × 26NR		16.8 ± 0.5 a	-6.3 ± 2.1 a	0.6 ± 2.1 a	17.4 ± 0.5 a	-5.2 ± 1.4 a	4.1 ± 1.9 a	14.6 ± 0.2 a	0.5 ± 1.2 a	3.8 ± 1.3 a
BC[5]		14.8 ± 0.8 a	-8.2 ± 1.3 a	-4.5 ± 3.0 a	15.7 ± 0.8 a	-5.9 ± 1.3 a	-2.0 ± 2.9 a	14.6 ± 0.2 a	3.4 ± 1.0 a	7.0 ± 1.2 a
F[6]		16.3 ± 0.7 a	-4.6 ± 1.1 a	-1.5 ± 2.6 a	15.5 ± 0.9 a	-5.2 ± 1.3 a	0.4 ± 3.1 a	14.3 ± 0.2 a	-0.6 ± 1.0 b	2.1 ± 1.2 b

[1] Original cross, *B. napus* parent and type of population (F_2 or BC_1) from which the paternal inbred lines of the test hybrids were developed.
[2] = Rutabaga-BF × Hi-Q and [3] = Rutabaga-BF × A07-26NR.
[4] *B. napus* parents of the test hybrids used as checks in the field trials.
[5] and [6] = Test hybrids of the BC_1- and F_2-derived inbred lines, respectively.
[7] Test hybrid population that were produced from the BC_1- and F_2-derived inbred lines.
Values within the group with different letters are significantly different, $p < 0.05$.

Table A3.8: Agronomic traits of the inbred lines (IN) derived from BC_1 of the Rutabaga-BF × Hi-Q cross and their test hybrids (TH), produced from the Hi-Q × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q (CPH) for these traits.

Inbred Line / Test Hybrid	DTF[1] (day) Mean ± SE		MPH	CPH	DTM[1] (day) Mean ± SE		MPH	CPH	Plant height (cm) Mean ± SE		MPH	CPH	Seed yield (kg/ha) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TC	(%)		IN	TH	(%)	
CO.BC.IN/TH.03[2]	52.6 ± 2.4	50.7 ± 3.7	-1.4 ± 2.2	5.0 ± 2.5	103.9 ± 4.2	104.1 ± 4.3	-1.1 ± 1.7	1.7 ± 2.0	130.8 ± 5.6	129.8 ± 11.7	2.5 ± 4.4	4.5 ± 4.7	3140.1 ± 290.0	5244.4 ± 745.7	32.9 ± 9.9	17.9 ± 10.0
CO.BC.IN/TH.04	53.7 ± 2.4	50.0 ± 3.7	-4.4 ± 2.2	-2.0 ± 2.5	105.5 ± 4.2	101.9 ± 4.3	-1.8 ± 1.7	0.1 ± 2.0	132.0 ± 5.7	128.8 ± 11.7	6.3 ± 4.4	6.9 ± 4.7	2913.3 ± 299.0	5026.9 ± 748.5	7.9 ± 12.3	15.4 ± 10.1
CO.BC.IN/TH.06	54.5 ± 2.4	48.8 ± 3.7	-3.6 ± 2.2	0.2 ± 2.5	104.5 ± 4.2	104.8 ± 4.3	-1.6 ± 1.7	1.4 ± 2.0	130.3 ± 5.6	131.1 ± 11.7	4.4 ± 4.4	10.3 ± 4.7	2939.7 ± 290.0	5506.9 ± 748.4	26.3 ± 8.6	13.5 ± 10.1
CO.BC.IN/TH.07	52.6 ± 2.4	50.8 ± 3.8	2.5 ± 2.2	2.9 ± 2.5	104.8 ± 4.2	105.8 ± 4.3	2.6 ± 1.7	3.9 ± 2.0	130.1 ± 5.6	128.2 ± 11.7	-1.4 ± 4.4	4.0 ± 4.7	3073.2 ± 290.1	4765.0 ± 750.8	3.7 ± 8.7	11.2 ± 10.1
CO.BC.IN/TH.08	54.3 ± 2.4	49.7 ± 3.7	-2.4 ± 2.2	3.0 ± 2.5	104.7 ± 4.2	101.9 ± 4.4	-2.9 ± 1.9	-1.4 ± 2.2	126.0 ± 5.6	116.1 ± 11.7	-3.3 ± 4.4	-5.7 ± 4.7	2921.4 ± 289.8	5250.0 ± 747.4	36.5 ± 8.5	6.5 ± 10.0
CO.BC.IN/TH.09	54.6 ± 2.4	50.5 ± 3.8	-3.4 ± 2.2	1.5 ± 2.5	105.3 ± 4.2	103.8 ± 4.3	-2.0 ± 1.7	-0.7 ± 2.0	128.6 ± 5.6	129.2 ± 11.7	-0.7 ± 4.4	-2.0 ± 4.7	2766.1 ± 289.8	4962.6 ± 750.8	27.7 ± 8.6	14.7 ± 10.1
CO.BC.IN/TH.16	53.0 ± 2.4	52.2 ± 3.7	1.5 ± 2.2	6.5 ± 2.5	104.3 ± 4.3	104.7 ± 4.3	0.0 ± 1.7	2.7 ± 2.0	126.2 ± 5.9	127.4 ± 11.7	4.8 ± 4.4	6.5 ± 4.7	2849.9 ± 306.9	5054.0 ± 767.3	48.0 ± 12.3	2.0 ± 11.3
CO.BC.IN/TH.18	49.8 ± 2.4	47.7 ± 3.7	-4.7 ± 1.8	-5.2 ± 2.1	101.4 ± 4.2	99.7 ± 4.1	-3.1 ± 1.4	-3.1 ± 1.6	123.7 ± 5.6	123.9 ± 11.4	-1.6 ± 3.6	-2.4 ± 3.7	3447.8 ± 290.1	5621.3 ± 747.4	38.6 ± 8.5	19.8 ± 11.3
CO.BC.IN/TH.19	51.7 ± 2.4	48.3 ± 3.7	-5.1 ± 1.9	-2.1 ± 2.0	103.9 ± 4.2	101.5 ± 4.1	-2.6 ± 1.4	-0.8 ± 1.6	123.2 ± 5.6	118.4 ± 11.4	-2.3 ± 3.6	-7.3 ± 3.7	2997.2 ± 289.9	4935.0 ± 748.7	23.2 ± 12.3	12.9 ± 10.1
CO.BC.IN/TH.20	53.3 ± 2.4	49.1 ± 3.7	-1.6 ± 1.8	-0.6 ± 2.0	102.6 ± 4.2	101.4 ± 4.1	-3.3 ± 1.4	-2.3 ± 1.6	117.3 ± 5.6	123.0 ± 11.4	0.7 ± 3.6	-2.0 ± 3.7	2871.0 ± 289.8	5647.9 ± 748.5	21.8 ± 8.6	27.5 ± 10.1
CO.BC.IN/TH.21	53.2 ± 2.4	48.2 ± 3.7	-1.2 ± 1.8	-0.6 ± 2.0	102.6 ± 4.2	100.8 ± 4.1	-0.9 ± 1.4	-0.7 ± 1.6	119.8 ± 5.6	120.0 ± 11.4	-0.9 ± 3.6	-1.6 ± 3.7	2776.9 ± 289.8	5379.7 ± 765.5	10.8 ± 8.5	6.4 ± 11.3
CO.BC.IN/TH.22	52.3 ± 2.4	48.8 ± 3.7	-2.5 ± 1.8	-0.7 ± 2.1	104.2 ± 4.2	101.8 ± 4.1	-1.2 ± 1.4	0.7 ± 1.6	118.5 ± 5.6	118.5 ± 11.4	-2.7 ± 3.6	-7.0 ± 3.7	3034.8 ± 290.0	5671.6 ± 748.4	35.7 ± 8.6	25.2 ± 10.1
CO.BC.IN/TH.23	54.0 ± 2.4	49.2 ± 3.7	-4.5 ± 1.8	-0.7 ± 2.0	104.2 ± 4.2	104.7 ± 4.1	0.1 ± 1.5	2.9 ± 1.6	123.4 ± 5.6	121.6 ± 11.4	0.2 ± 3.6	-2.8 ± 3.7	2948.7 ± 290.0	5204.9 ± 747.5	20.4 ± 8.5	4.9 ± 10.0
CO.BC.IN/TH.24	53.5 ± 2.4	49.1 ± 3.7	0.3 ± 1.8	2.1 ± 2.1	104.7 ± 4.2	100.9 ± 4.1	-2.0 ± 1.4	0.5 ± 1.6	128.2 ± 5.6	125.1 ± 11.4	2.3 ± 3.6	1.1 ± 3.7	3115.4 ± 290.4	5302.4 ± 772.2	15.1 ± 8.7	7.4 ± 11.3
CO.BC.IN/TH.25	53.2 ± 2.4	48.3 ± 3.7	-3.2 ± 1.8	-2.1 ± 2.0	103.9 ± 4.2	102.8 ± 4.1	-0.2 ± 1.4	1.9 ± 1.6	119.2 ± 5.6	115.4 ± 11.4	-5.3 ± 3.6	-7.6 ± 3.7	3041.9 ± 289.8	5127.1 ± 747.5	21.0 ± 8.5	14.7 ± 10.0
CO.BC.IN/TH.26	52.1 ± 2.4	48.5 ± 3.7	-3.5 ± 1.8	-1.8 ± 2.1	103.1 ± 4.2	100.1 ± 4.1	-3.9 ± 1.4	-0.6 ± 1.6	122.3 ± 5.6	117.5 ± 11.4	-0.5 ± 3.6	-4.9 ± 3.7	2992.3 ± 290.1	5129.6 ± 750.6	26.4 ± 9.9	15.5 ± 10.1
CO.BC.IN/TH.27	52.3 ± 2.4	49.7 ± 3.7	-1.8 ± 1.8	0.2 ± 2.0	102.1 ± 4.2	102.4 ± 4.1	-1.2 ± 1.4	2 .0± 1.6	124.7 ± 5.7	118.7 ± 11.4	0.0 ± 3.6	-4.4 ± 3.7	2837.2 ± 296.1	5474.2 ± 747.4	36.6 ± 9.9	18.2 ± 10.0
CO.BC.IN/TH.28	54.5 ± 2.4	50.1 ± 3.7	-3.5 ± 1.8	-1.2 ± 2.1	103.1 ± 4.2	102.6 ± 4.1	-0.1 ± 1.5	1.8 ± 1.6	122.5 ± 5.6	123.9 ± 11.4	0.7 ± 3.6	-4.1 ± 3.7	2882.0 ± 290.0	4766.9 ± 747.4	10.2 ± 9.9	2.4 ± 10.0
CO.BC.IN/TH.29	50.6 ± 2.4	47.1 ± 3.7	-5.2 ± 1.8	-5.3 ± 2.1	103.5 ± 4.2	100.1 ± 4.1	-2.0 ± 1.5	-1.4 ± 1.6	126.4 ± 5.7	124.5 ± 11.4	-2.3 ± 3.6	-4.8 ± 3.7	3121.0 ± 296.1	5065.9 ± 748.4	29.8 ± 10	9.5 ± 10.1
CO.BC.IN/TH.31	52.6 ± 2.4	48.3 ± 3.7	-4.1 ± 1.8	-1.5 ± 2.0	102.5 ± 4.2	100.2 ± 4.1	-1.9 ± 1.4	0.5 ± 1.6	122.5 ± 5.7	119.2 ± 11.4	-2.6 ± 3.6	-4.0 ± 3.7	2768.8 ± 296.2	5007.1 ± 748.3	20.0 ± 8.6	5.9 ± 10.1
CO.BC.IN/TH.32	52.9 ± 2.4	49.2 ± 3.7	-1.2 ± 1.8	0.0 ± 2.0	102.4 ± 4.2	99.2 ± 4.1	-3.7 ± 1.4	-3.6 ± 1.6	122.6 ± 5.6	123.7 ± 11.4	-0.6 ± 3.6	-2.7 ± 3.7	2849.4 ± 289.9	5814.8 ± 747.5	44.7 ± 8.5	31.7 ± 10.0
CO.BC.IN/TH.33	53.9 ± 2.4	49.3 ± 3.7	-2.0 ± 1.8	-0.3 ± 2.0	103.3 ± 4.2	103.9 ± 4.1	-0.9 ± 1.4	1.3 ± 1.6	120.4 ± 5.7	125.5 ± 11.4	3.2 ± 3.6	-3.5 ± 3.7	3022.4 ± 296.1	4899.9 ± 748.7	6.0 ± 8.6	1.7 ± 10.1
CO.BC.IN/TH.34	53.9 ± 2.4	51.0 ± 3.7	-2.7 ± 1.8	1.7 ± 2.0	104.0 ± 4.2	104.3 ± 4.1	-0.4 ± 1.5	2.5 ± 1.6	131.7 ± 5.7	125.4 ± 11.4	-1.7 ± 3.6	2.3 ± 3.7	3062.3 ± 296.1	5043.7 ± 750.8	36.6 ± 8.6	21.7 ± 10.1
CO.BC.IN/TH.36	53.3 ± 2.4	47.7 ± 3.7	-3.0 ± 1.8	0.0 ± 2.1	103.8 ± 4.2	101.2 ± 4.1	-2.0 ± 1.4	-1.8 ± 1.6	123.6 ± 5.6	120.1 ± 11.5	-2.5 ± 3.8	-5.6 ± 4.0	2977.2 ± 290.2	5562.1 ± 748.3	32.6 ± 8.6	37.5 ± 10.1
CO.BC.IN/TH.37	53.8 ± 2.4	50.0 ± 3.7	-1.0 ± 1.8	1.6 ± 2.1	103.7 ± 4.2	103.3 ± 4.1	-1.5 ± 1.4	0.7 ± 1.7	130.5 ± 5.6	124.9 ± 11.4	0.7 ± 3.7	0 .0 ± 3.8	2814.0 ± 289.7	4697.8 ± 752.8	26.1 ± 10.0	7.4 ± 11.3
Hi-Q	51.3 ± 2.4	49.0 ± 3.7			102.7 ± 4.2	102.5 ± 4			127.1 ± 5.3	128.3 ± 11.1			3377.9 ± 274.2	4642.3 ± 675.3		

[1] DTF = days to flowering and DTM = days to maturity.

[2] CO.BC.IN/TH = BC_1-derived inbred line (IN) of Rutabaga-BF × Hi-Q and its test hybrid (TH) of Hi-Q × IN. For example, CO.BC.IN/TH.03 shows data of the inbred line CO.BC.IN.03 (see Table A3.16 for details) and its test hybrid.

Table A3.9: Agronomic traits of the inbred lines (IN) derived from F_2 of the Rutabaga-BF × Hi-Q cross and their test hybrids (TH), produced from the Hi-Q × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q (CPH) for these traits.

Inbred Line / Test Hybrid	DTF[1] (day) Mean ± SE		MPH	CPH	DTM[1] (day) Mean ± SE		MPH	CPH	Plant height (cm) Mean ± SE		MPH	CPH	Seed yield (kg/ha) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TH	(%)		IN	TH	(%)	
CO.FF.IN/TH.13[2]	52.4 ± 2.4	49.0 ± 3.7	-2.8 ± 1.8	-1.2 ± 2.0	102.6 ± 4.2	103.2 ± 4.1	0.2 ± 1.4	2.0 ± 1.6	128.1 ± 5.6	123.8 ± 11.4	-3.0 ± 3.6	-3.9 ± 3.7	2957.1 ± 289.8	4795.4 ± 750.9	13.1 ± 10.0	3.6 ± 10.1
CO.FF.IN/TH.15	52.5 ± 2.4	48.1 ± 3.7	-2.8 ± 1.8	-1.2 ± 2.0	102.8 ± 4.2	100.1 ± 4.1	-2.3 ± 1.4	-1.9 ± 1.6	128.5 ± 5.6	118.1 ± 11.4	-5.4 ± 3.6	-7.7 ± 3.7	2813.0 ± 289.8	4675.7 ± 767.7	23.8 ± 9.9	22.6 ± 11.3
CO.FF.IN/TH.16	52.7 ± 2.4	49.9 ± 3.7	-2.0 ± 1.8	-0.4 ± 2.0	103.5 ± 4.2	101.0 ± 4.1	-2.7 ± 1.5	-1.3 ± 1.7	128.1 ± 5.6	124.2 ± 11.4	1.1 ± 3.6	0.8 ± 3.7	2937.5 ± 289.7	4960.1 ± 754.1	26.4 ± 8.7	14.0 ± 10.1
CO.FF.IN/TH.17	50.2 ± 2.4	49.1 ± 3.7	0.2 ± 1.8	0.7 ± 2.0	101.7 ± 4.3	99.9 ± 4.1	-2 .0± 1.4	-0.1 ± 1.6	120.0 ± 5.9	126.6 ± 11.4	6.9 ± 3.6	4.1 ± 3.7	3216.2 ± 306.9	5085.3 ± 750.7	12.8 ± 8.6	9.2 ± 10.1
CO.FF.IN/TH.18	50.0 ± 2.4	48.1 ± 3.7	-1.1 ± 1.8	-1.3 ± 2.0	101.2 ± 4.3	99.5 ± 4.1	-2.9 ± 1.4	-0.6 ± 1.6	123.3 ± 5.9	116.9 ± 11.4	-2.9 ± 3.6	-6.6 ± 3.7	3411.4 ± 306.8	5987.1 ± 767.9	29.6 ± 8.6	15.9 ± 10.1
CO.FF.IN/TH.19	53.2 ± 2.4	50.4 ± 3.7	-2.3 ± 1.8	-0.1 ± 2.0	103.7 ± 4.2	101.0 ± 4.1	-6.0 ± 1.4	-3.6 ± 1.6	131.5 ± 5.6	124.8 ± 11.4	-4.0 ± 3.6	-1.3 ± 3.7	3014.0 ± 289.8	5724.3 ± 747.8	24.8 ± 8.5	9.2 ± 10.0
CO.FF.IN/TH.20	52.2 ± 2.4	48.9 ± 3.7	-3.5 ± 1.8	-1.2 ± 2.0	103.7 ± 4.2	100.9 ± 4.1	-2.1 ± 1.4	-0.6 ± 1.6	126.7 ± 5.6	125.5 ± 11.4	-0.3 ± 3.6	-0.2 ± 3.7	3030.7 ± 290.0	4834.0 ± 756.3	15.9 ± 8.9	8.5 ± 10.3
CO.FF.IN/TH.21	52.5 ± 2.4	49.8 ± 3.7	-2.8 ± 1.8	1.0 ± 2.0	104.6 ± 4.2	101.4 ± 4.1	-2.7 ± 1.4	-1.1 ± 1.6	137.3 ± 5.6	126.9 ± 11.4	-2.4 ± 3.6	2.4 ± 3.7	3101.7 ± 290.2	4265.4 ± 749.3	22.1 ± 9.9	-4.4 ± 11.3
CO.FF.IN/TH.22	52.5 ± 2.4	50.5 ± 3.7	-1.2 ± 1.8	2.8 ± 2.0	102.6 ± 4.3	103.1 ± 4.1	-0.2 ± 1.4	3.5 ± 1.6	131.9 ± 5.9	125.8 ± 11.4	-3.8 ± 3.6	-3.1 ± 3.7	3408.7 ± 306.8	5078.9 ± 749.1	25.8 ± 9.9	4.0 ± 11.3
CO.FF.IN/TH.23	52.5 ± 2.4	50.3 ± 3.7	-2.4 ± 1.9	0.5 ± 2.0	103.4 ± 4.2	103.9 ± 4.1	-0.8 ± 1.5	2.5 ± 1.6	134.5 ± 5.7	128.4 ± 11.4	-0.1 ± 3.6	2.0 ± 3.7	3115.5 ± 296.4	5173.9 ± 746.2	29.1 ± 8.5	12.7 ± 10.0
CO.FF.IN/TH.24	50.2 ± 2.4	48.6 ± 3.7	-2.6 ± 1.8	-1.3 ± 2.0	102.7 ± 4.3	101.6 ± 4.1	-1.8 ± 1.4	-1.2 ± 1.6	124.8 ± 5.7	123.0 ± 11.4	3.4 ± 3.6	0.5 ± 3.7	3282.2 ± 299.1	5755.7 ± 748.9	26.7 ± 9.9	29.2 ± 10.1
CO.FF.IN/TH.25	50.4 ± 2.4	49.2 ± 3.7	-1.7 ± 1.8	-1.3 ± 2.0	102.7 ± 4.2	101.1 ± 4.1	-1.3 ± 1.4	0.0 ± 1.6	124.4 ± 5.6	127.4 ± 11.4	1.0 ± 3.6	-1.8 ± 3.7	3329.1 ± 289.8	5614.8 ± 753.0	19.0 ± 10.0	14.8 ± 11.3
CO.FF.IN/TH.26	50.4 ± 2.4	48.0 ± 3.7	-2.2 ± 1.8	-2.4 ± 2.0	101.5 ± 4.2	99.1 ± 4.1	-3.4 ± 1.4	0.0 ± 1.6	127.3 ± 5.7	119.8 ± 11.4	2.3 ± 3.6	-0.1 ± 3.7	3403.0 ± 296.1	5217.6 ± 747.1	37.4 ± 8.6	21.0 ± 10.1
CO.FF.IN/TH.27	52.8 ± 2.4	49.3 ± 3.7	-1.4 ± 1.8	3.0 ± 2.0	101.9 ± 4.2	100.5 ± 4.1	-1.9 ± 1.4	-0.6 ± 1.6	129.9 ± 5.7	127.3 ± 11.4	6.2 ± 3.6	3.5 ± 3.7	3143.2 ± 296.4	4184.4 ± 751.5	-0.4 ± 8.6	-7.8 ± 10.1
CO.FF.IN/TH.28	55.2 ± 2.4	51.0 ± 3.7	-0.7 ± 1.8	3.3 ± 2.0	105.1 ± 4.3	99.8 ± 4.1	-5.2 ± 1.4	-0.5 ± 1.6	139.3 ± 5.7	128.0 ± 11.4	-4.3 ± 3.6	1.6 ± 3.7	3044.9 ± 299.0	5015.2 ± 754.1	27.1 ± 8.7	11.0 ± 10.1
CO.FF.IN/TH.29	52.4 ± 2.4	47.9 ± 3.7	-3.9 ± 1.8	-1.4 ± 2.1	103.0 ± 4.2	100.0 ± 4.1	-2.0 ± 1.4	-0.5 ± 1.7	136.4 ± 5.7	118.4 ± 11.4	-11.0 ± 3.6	-5.8 ± 3.8	3348.5 ± 296.2	5215.3 ± 752.1	21.3 ± 8.7	10.9 ± 10.1
CO.FF.IN/TH.30	51.0 ± 2.4	47.7 ± 3.7	-5.7 ± 1.8	-3.7 ± 2.0	101.5 ± 4.2	101.3 ± 4.1	-2.7 ± 1.5	-1.0 ± 1.7	125.3 ± 5.7	119.0 ± 11.4	-5.0 ± 3.6	-5.1 ± 3.7	3478.3 ± 296.0	5652.0 ± 751.0	17.0 ± 9.9	28.6 ± 10.1
CO.FF.IN/TH.31	50.9 ± 2.4	48.9 ± 3.7	-1.0 ± 1.8	0.0 ± 2.0	102.8 ± 4.2	101.9 ± 4.1	-0.8 ± 1.4	0.9 ± 1.6	123.7 ± 5.6	127.6 ± 11.4	2.3 ± 3.6	-2.2 ± 3.7	3317.8 ± 289.8	4476.2 ± 770.3	14.8 ± 10	2.9 ± 11.3
CO.FF.IN/TH.32	53.1 ± 2.4	50.8 ± 3.7	0.4 ± 1.8	4.8 ± 2.0	103.2 ± 4.2	103.8 ± 4.1	-0.5 ± 1.4	3.3 ± 1.6	135.2 ± 5.6	130.8 ± 11.4	0.9 ± 3.6	5.4 ± 3.7	2957.8 ± 290.2	4547.1 ± 747.8	23.5 ± 8.5	1.4 ± 10.0
CO.FF.IN/TH.33	54.9 ± 2.4	49.2 ± 3.7	-2.6 ± 1.8	-0.3 ± 2.0	104.9 ± 4.3	100.3 ± 4.1	-3.6 ± 1.4	-0.9 ± 1.6	137.2 ± 5.9	129.7 ± 11.4	-0.3 ± 3.6	3.1 ± 3.7	3326.1 ± 306.9	5122.1 ± 748.3	25.0 ± 8.7	10.2 ± 11.4
CO.FF.IN/TH.34	53.1 ± 2.4	50.7 ± 3.7	-0.7 ± 1.8	3.2 ± 2.0	103.5 ± 4.2	102.9 ± 4.1	-0.9 ± 1.4	0.7 ± 1.6	133.8 ± 5.6	123.6 ± 11.4	2.0 ± 3.6	1.5 ± 3.7	2981.3 ± 290.0	4902.1 ± 767.8	7.5 ± 8.5	-4.2 ± 10.0
CO.FF.IN/TH.35	54.9 ± 2.4	51.7 ± 3.7	3.3 ± 2.0	3.7 ± 2.0	102.8 ± 4.3	106.5 ± 4.1	2.3 ± 1.6	3.4 ± 1.7	132.0 ± 5.9	131.3 ± 11.4	-1.5 ± 3.6	3.2 ± 3.7	2965.4 ± 306.9	5098.2 ± 767.9	21.4 ± 12.3	-1.9 ± 13.5
CO.FF.IN/TH.36	54.8 ± 2.4	50.7 ± 3.7	-4.3 ± 1.8	2.8 ± 2.0	103.9 ± 4.3	101.3 ± 4.1	-0.8 ± 1.6	2.3 ± 1.6	138.0 ± 5.7	128.0 ± 11.4	-2.3 ± 3.6	0.2 ± 3.7	3209.6 ± 299.0	5310.4 ± 751.1	25.7 ± 8.6	14.1 ± 10.1
CO.FF.IN/TH.38	55.0 ± 2.4	52.1 ± 3.7	-0.7 ± 1.8	8.9 ± 2.1	103.1 ± 4.3	105.3 ± 4.1	-0.6 ± 1.5	2.0 ± 1.6	134.1 ± 5.9	137.0 ± 11.4	1.5 ± 3.6	11.9 ± 3.8	3210.8 ± 307.0	5221.2 ± 753.5	31.7 ± 8.7	9.5 ± 10.2
CO.FF.IN/TH.39	50.0 ± 2.4	47.9 ± 3.7	-3.2 ± 1.9	-3.3 ± 2.1	101.1 ± 4.3	98.2 ± 4.1	-2.1 ± 1.6	-2.9 ± 1.7	122.0 ± 5.9	117.9 ± 11.4	-0.8 ± 3.6	-2.6 ± 3.7	3159.6 ± 306.8	5087.4 ± 747.3	26.1 ± 9.9	17.7 ± 10.1
CO.FF.IN/TH.40	50.2 ± 2.4	48.9 ± 3.7	-1.4 ± 1.8	-1.9 ± 2.1	101.1 ± 4.3	99.7 ± 4.1	-2.9 ± 1.4	-2.3 ± 1.6	120.8 ± 5.7	124.1 ± 11.4	1.3 ± 3.6	-3.3 ± 3.7	3289.2 ± 299.2	5024.7 ± 751.2	17.9 ± 10.0	14.5 ± 11.3
Hi-Q	51.3 ± 2.4	49.0 ± 3.7			102.7 ± 4.2	102.5 ± 4.0			127.1 ± 5.3	128.3 ± 11.1			3377.9 ± 274.2	4642.3 ± 675.3		

[1] DTF = days to flowering and DTM = days to maturity.

[2] CO.FF.IN/TH = F_2-derived inbred line (IN) of Rutabaga-BF × Hi-Q and its test hybrid (TH) of Hi-Q × IN. For example, CO.FF.IN/TH.13 shows data of the inbred line CO.FF.IN.13 (see Table A3.16 for details) and its test hybrid.

Table A3.10: Agronomic traits of the inbred lines (IN) derived from BC_1 of the Rutabaga-BF × A07-26NR cross and their test hybrids (TH), produced from the A07-26NR × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over A07-26NR (CPH) for these traits.

Inbred Line / Test Hybrid	DTF[1] (day) Mean ± SE		MPH	CPH	DTM[1] (day) Mean ± SE		MPH	CPH	Plant height (cm) Mean ± SE		MPH	CPH	Seed yield (kg/ha) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TH	(%)		IN	TH	(%)	
RR.BC.IN/TH.01[2]	53.5 ± 2.4	49.2 ± 3.8	-3.9 ± 2.4	0.5 ± 2.7	104.3 ± 4.3	100.4 ± 4.4	-0.8 ± 1.9	2.7 ± 2.2	134.1 ± 5.9	116.0 ± 11.9	-4.4 ± 4.9	5.7 ± 5.3	3029.5 ± 306.8	5422.2 ± 765.5	31.0 ± 9.9	5.4 ± 11.3
RR.BC.IN/TH.03	50.4 ± 2.4	50.0 ± 3.7	0.3 ± 2.2	6.5 ± 2.5	100.7 ± 4.2	101.9 ± 4.3	0.4 ± 1.7	1.6 ± 2.0	115.9 ± 5.6	116.8 ± 11.7	4.8 ± 4.4	4.1 ± 4.7	3229.3 ± 289.9	4788.7 ± 745.8	12.8 ± 8.5	-1.0 ± 10.0
RR.BC.IN/TH.05	50.1 ± 2.4	47.5 ± 3.7	-4.3 ± 2.2	-2.3 ± 2.5	100.4 ± 4.2	99.7 ± 4.3	-0.4 ± 1.7	-2.0 ± 2.0	117.3 ± 5.6	117.8 ± 11.7	4.8 ± 4.4	1.6 ± 4.7	3089.4 ± 289.9	4971.5 ± 745.8	24.2 ± 8.5	16.7 ± 10.0
RR.BC.IN/TH.14	51.9 ± 2.4	50.0 ± 3.7	-0.4 ± 1.8	1.3 ± 2.0	102.8 ± 4.2	100.1 ± 4.1	-1.6 ± 1.4	-0.7 ± 1.6	116.9 ± 5.6	114.0 ± 11.4	0.5 ± 3.6	-1.4 ± 3.7	3120.3 ± 290.0	4673.1 ± 747.2	8.7 ± 8.6	4.5 ± 10.1
RR.BC.IN/TH.15	52.5 ± 2.4	49.1 ± 3.7	-2.8 ± 1.8	0.7 ± 2.0	102.4 ± 4.2	99.1 ± 4.1	-2.3 ± 1.4	-0.7 ± 1.6	117.9 ± 5.6	115.0 ± 11.4	-0.6 ± 3.6	-0.6 ± 3.7	3133.6 ± 289.9	5112.0 ± 766.3	10.6 ± 9.9	6.7 ± 11.3
RR.BC.IN/TH.17	50.2 ± 2.4	48.7 ± 3.7	-2.9 ± 1.8	0.2 ± 2.0	101.3 ± 4.2	104.1 ± 4.1	0.4 ± 1.4	1.8 ± 1.6	116.6 ± 5.6	121.6 ± 11.4	3.5 ± 3.6	4.9 ± 3.7	2932.1 ± 291.5	5095.5 ± 746.4	13.9 ± 12.2	1.9 ± 10.1
RR.BC.IN/TH.18	50.4 ± 2.4	48.8 ± 3.7	-4.1 ± 1.8	-0.3 ± 2.0	99.0 ± 4.2	99.4 ± 4.1	-3.2 ± 1.4	-1.3 ± 1.6	112.8 ± 5.6	111.9 ± 11.4	-7.2 ± 3.6	-3.7 ± 3.7	2903.2 ± 291.8	4868.3 ± 765.6	14.0 ± 9.9	6.7 ± 10.0
RR.BC.IN/TH.19	49.3 ± 2.4	47.5 ± 3.7	-3.1 ± 1.8	-4.1 ± 2.0	99.6 ± 4.2	99.6 ± 4.1	-2.7 ± 1.4	-2.5 ± 1.6	108.4 ± 5.7	107.4 ± 11.4	2.5 ± 3.6	-1.5 ± 3.7	3164.1 ± 296.0	5055.5 ± 802.0	30.5 ± 8.5	26.1 ± 10.0
RR.BC.IN/TH.21	50.1 ± 2.4	48.8 ± 3.7	-1.9 ± 1.8	-0.9 ± 2.0	99.3 ± 4.2	98.5 ± 4.1	-2.3 ± 1.4	-2.5 ± 1.6	115.9 ± 5.6	119.4 ± 11.4	2.0 ± 3.6	3.2 ± 3.7	2872.3 ± 289.8	4529.5 ± 767.5	18.2 ± 9.9	6.7 ± 11.2
RR.BC.IN/TH.22	50.8 ± 2.4	50.2 ± 3.7	0.8 ± 1.8	3.1 ± 2.0	99.9 ± 4.2	101.8 ± 4.1	-2.4 ± 1.4	-1.7 ± 1.6	114.7 ± 5.6	118.6 ± 11.4	2.4 ± 3.6	4.3 ± 3.7	3149.1 ± 291.7	5339.0 ± 747.3	29.1 ± 10.0	10.3 ± 11.3
RR.BC.IN/TH.23	51.6 ± 2.4	49.4 ± 3.7	-2.5 ± 1.8	1.4 ± 2.0	101.9 ± 4.3	101.9 ± 4.1	-0.6 ± 1.4	1.5 ± 1.6	118.9 ± 5.7	121.4 ± 11.4	6.7 ± 3.6	9.3 ± 3.7	3339.7 ± 298.9	4785.2 ± 802	35.1 ± 9.9	27.6 ± 10.0
RR.BC.IN/TH.24	51.5 ± 2.4	48.0 ± 3.7	-5.5 ± 1.8	-2.0 ± 2.0	102.0 ± 4.3	100.5 ± 4.1	-2.7 ± 1.4	-1.0 ± 1.6	119.6 ± 5.7	116.4 ± 11.4	2.9 ± 3.6	5.0 ± 3.7	3547.6 ± 299.0	5064.0 ± 767.1	27.0 ± 12.2	22.6 ± 10.1
RR.BC.IN/TH.25	51.8 ± 2.4	49.8 ± 3.7	-2.4 ± 1.8	1.4 ± 2.0	101.7 ± 4.2	101.4 ± 4.1	-1.7 ± 1.4	0.8 ± 1.6	122.4 ± 5.6	123.0 ± 11.4	6.6 ± 3.6	11.2 ± 3.7	3568.4 ± 290.0	5344.4 ± 747.6	12.3 ± 8.6	5.0 ± 10.0
RR.BC.IN/TH.26	50.2 ± 2.4	48.3 ± 3.7	-3.8 ± 1.8	-3.2 ± 2.0	101.3 ± 4.3	100.5 ± 4.1	-3.9 ± 1.4	-2.9 ± 1.6	115.1 ± 5.7	116.4 ± 11.4	1.9 ± 3.6	0.9 ± 3.7	3313.2 ± 298.9	5554.9 ± 766.5	19.1 ± 12.2	3.9 ± 13.4
RR.BC.IN/TH.27	49.7 ± 2.4	48.5 ± 3.7	-2.5 ± 1.8	-1.8 ± 2.0	100.7 ± 4.2	101.2 ± 4.1	-1.8 ± 1.4	-0.7 ± 1.6	112.8 ± 5.7	114.0 ± 11.4	-0.7 ± 3.6	-5.5 ± 3.7	3162.1 ± 296.0	4730.2 ± 745.9	17.2 ± 8.5	7.0 ± 10.0
RR.BC.IN/TH.28	51.5 ± 2.4	49.8 ± 3.7	-1.1 ± 1.8	2.4 ± 2.0	102.5 ± 4.2	98.4 ± 4.1	-4.5 ± 1.4	-2.1 ± 1.6	119.2 ± 5.7	112.3 ± 11.4	-3.1 ± 3.6	-2.5 ± 3.7	3115.5 ± 296.0	4472.2 ± 767.7	11.1 ± 8.6	1.5 ± 10.1
RR.BC.IN/TH.29	51.9 ± 2.4	49.9 ± 3.7	-0.2 ± 1.8	4.9 ± 2.0	102.2 ± 4.2	101.6 ± 4.1	-2.1 ± 1.4	0.9 ± 1.6	116.7 ± 5.6	125.2 ± 11.4	6.0 ± 3.6	9.5 ± 3.7	3196.7 ± 291.8	4423.8 ± 746.6	-4.1 ± 8.6	-17.9 ± 10.1
RR.BC.IN/TH.30	50.9 ± 2.4	49.2 ± 3.7	-4.2 ± 1.8	0.4 ± 2.0	103.1 ± 4.4	103.0 ± 4.1	-1.0 ± 1.4	1.6 ± 1.6	123.0 ± 6.5	114.8 ± 11.4	-5.7 ± 3.6	0.7 ± 3.7	3686.6 ± 343.8	5077.6 ± 803.8	57.4 ± 17.2	3.0 ± 10.1
RR.BC.IN/TH.31	49.7 ± 2.4	47.1 ± 3.7	-4.8 ± 1.8	-4.1 ± 2.0	99.6 ± 4.2	98.6 ± 4.1	-2.2 ± 1.4	-0.5 ± 1.6	111.8 ± 5.6	110.6 ± 11.4	0.9 ± 3.6	-2.3 ± 3.7	3501.3 ± 291.9	4874.9 ± 745.9	25.3 ± 8.5	18.0 ± 10.0
RR.BC.IN/TH.32	50.9 ± 2.4	48.0 ± 3.7	-5.6 ± 1.8	-2.2 ± 2.0	100.2 ± 4.2	99.1 ± 4.1	-3.6 ± 1.4	-2.2 ± 1.6	115.9 ± 5.7	116.1 ± 11.5	-1.7 ± 3.8	-0.8 ± 4	3000.3 ± 296.1	4706.6 ± 745.8	16.6 ± 8.5	5.5 ± 10.0
RR.BC.IN/TH.33	52.2 ± 2.4	48.8 ± 3.7	-6.9 ± 1.8	2.5 ± 2.0	102.0 ± 4.2	101.8 ± 4.1	-1.9 ± 1.5	0.3 ± 1.6	117.3 ± 5.7	119.5 ± 11.4	-0.7 ± 3.6	2.2 ± 3.7	3112.9 ± 296.2	5876.8 ± 805.4	29.8 ± 12.2	28.7 ± 13.4
A07-26NR	49.1 ± 2.4	48.8 ± 3.7			99.9 ± 4.2	100.6 ± 4			114.5 ± 5.3	115.8 ± 11.1			3675.5 ± 274.3	4816.6 ± 671.6		

[1] DTF = days to flowering and DTM = days to maturity.

[2] RR.BC.IN/TH = BC_1-derived inbred line (IN) of Rutabaga-BF × A07-26NR and its test hybrid (TH) of A07-26NR × IN. For example, RR.BC.IN/TH.01 shows data of the inbred line RR.BC.IN.01 (see Table A3.17 for details) and its test hybrid.

Table A3.11: Agronomic traits of the inbred lines (IN) derived from F_2 of the Rutabaga-BF × A07-26NR cross and their test hybrids (TH), produced from the A07-26NR × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over A07-26NR (CPH) for these traits.

Inbred Line / Test Hybrid	DTF[1] (day) Mean ± SE		MPH	CPH	DTM[1] (day) Mean ± SE		MPH	CPH	Plant height (cm) Mean ± SE		MPH	CPH	Seed yield (kg/ha) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TH	(%)		IN	TH	(%)	
RR.FF.IN/TH.13[2]	51.1 ± 2.4	49.4 ± 3.7	-1.8 ± 2.2	2.8 ± 2.5	102.0 ± 4.3	102.9 ± 4.3	0.2 ± 1.7	3.4 ± 2.0	121.8 ± 5.9	127.5 ± 11.7	4.0 ± 4.4	7.8 ± 4.7	3303.7 ± 307.0	5975.1 ± 746.8	30.0 ± 8.6	12.0 ± 10.1
RR.FF.IN/TH.17	52.6 ± 2.4	49.8 ± 3.7	-1.1 ± 1.8	2.6 ± 2.0	103.2 ± 4.3	101.8 ± 4.1	1.0 ± 1.4	1.9 ± 1.6	124.6 ± 5.7	119.2 ± 11.4	-3.1 ± 3.6	1.8 ± 3.7	3366.7 ± 299.0	5077.5 ± 767.3	5.2 ± 8.6	-5.0 ± 10.1
RR.FF.IN/TH.18	51.8 ± 2.4	48.8 ± 3.7	-3.3 ± 1.8	0.3 ± 2.0	103.4 ± 4.2	101.6 ± 4.1	-1.2 ± 1.5	0.8 ± 1.7	122.7 ± 5.7	124.4 ± 11.4	3.1 ± 3.6	4.7 ± 3.7	3247.6 ± 296.3	5270.5 ± 765.6	31.1 ± 9.9	13.1 ± 10.0
RR.FF.IN/TH.19	49.4 ± 2.4	47.8 ± 3.7	-3.3 ± 1.8	-1.6 ± 2.0	100.3 ± 4.2	98.8 ± 4.1	-2.7 ± 1.4	-1.1 ± 1.6	112.7 ± 5.7	114.5 ± 11.4	2.3 ± 3.6	2.5 ± 3.7	3471.8 ± 296.3	5144.6 ± 746.6	7.6 ± 8.6	1.3 ± 10.1
RR.FF.IN/TH.20	49.4 ± 2.4	47.4 ± 3.7	-3.8 ± 1.8	-3.2 ± 2.0	99.5 ± 4.2	98.3 ± 4.1	-1.9 ± 1.4	0.2 ± 1.6	115.4 ± 5.6	111.2 ± 11.4	-3.8 ± 3.6	-5.8 ± 3.7	3612.6 ± 291.9	4802.3 ± 745.7	3.7 ± 8.5	8.4 ± 10.0
RR.FF.IN/TH.21	49.2 ± 2.4	48.6 ± 3.7	-1.5 ± 1.8	-0.7 ± 2.0	99.2 ± 4.2	102.5 ± 4.1	-0.2 ± 1.4	1.4 ± 1.6	109.8 ± 5.7	116.7 ± 11.4	2.5 ± 3.6	-1.4 ± 3.7	3518.3 ± 296.1	4641.8 ± 749.0	-0.2 ± 8.6	-2.2 ± 10.1
RR.FF.IN/TH.22	49.7 ± 2.4	48.3 ± 3.7	-1.5 ± 1.8	0.1 ± 2.0	100.6 ± 4.2	100.1 ± 4.1	-1.0 ± 1.4	-0.6 ± 1.6	111.9 ± 5.6	117.4 ± 11.4	6.4 ± 3.6	6.0 ± 3.7	3436.8 ± 290.3	5027.9 ± 767.1	12.4 ± 8.6	6.6 ± 10.1
RR.FF.IN/TH.23	49.2 ± 2.4	47.2 ± 3.7	-2.3 ± 1.8	-3.9 ± 2.0	98.7 ± 4.2	101.7 ± 4.1	0.0 ± 1.4	0.4 ± 1.6	111.6 ± 5.6	114.4 ± 11.4	-0.7 ± 3.6	-2.7 ± 3.7	3511.9 ± 290.0	5198.6 ± 765.6	8.2 ± 8.5	10.5 ± 10.0
RR.FF.IN/TH.24	49.3 ± 2.4	49.4 ± 3.7	-1.2 ± 1.8	1.1 ± 2.0	100.4 ± 4.2	100.8 ± 4.1	0.3 ± 1.4	1.3 ± 1.6	112.1 ± 5.7	111.2 ± 11.4	-0.1 ± 3.6	-0.4 ± 3.7	3438.8 ± 296.0	4322.9 ± 765.1	8.8 ± 8.5	12.1 ± 10.0
RR.FF.IN/TH.25	49.4 ± 2.4	49.6 ± 3.7	0.6 ± 1.8	-0.9 ± 2.0	99.9 ± 4.2	101.3 ± 4.1	-1.9 ± 1.4	-1.2 ± 1.6	114.1 ± 5.6	111.5 ± 11.4	-0.4 ± 3.6	-3.8 ± 3.7	3394.1 ± 293.7	4039.8 ± 764.3	-3.6 ± 8.5	-7 ± 10.0
RR.FF.IN/TH.26	50.1 ± 2.4	48.9 ± 3.7	-0.2 ± 1.8	0.6 ± 2.0	100.1 ± 4.2	100.9 ± 4.1	-0.5 ± 1.4	0.7 ± 1.6	109.1 ± 5.6	111.9 ± 11.4	-0.3 ± 3.6	-0.9 ± 3.7	3443.9 ± 289.8	5080.9 ± 748.6	-1.2 ± 8.6	-8.4 ± 10.0
RR.FF.IN/TH.27	48.8 ± 2.4	49.1 ± 3.7	-1.3 ± 1.8	0.1 ± 2.0	100.4 ± 4.2	99.0 ± 4.1	-2.1 ± 1.4	0.3 ± 1.6	108.9 ± 5.7	111.1 ± 11.4	-5.3 ± 3.6	-3.2 ± 3.7	3655.4 ± 296.2	5322.1 ± 767.2	3.9 ± 8.6	2.4 ± 10.1
RR.FF.IN/TH.28	50.1 ± 2.4	48.3 ± 3.7	-1.6 ± 1.8	0.0 ± 2.0	100.5 ± 4.3	99.0 ± 4.1	-0.5 ± 1.4	0.9 ± 1.6	110.8 ± 5.9	115.6 ± 11.4	2.6 ± 3.6	3.3 ± 3.7	3469.6 ± 306.8	4416.8 ± 765.7	-1.9 ± 8.5	-5.3 ± 10.0
RR.FF.IN/TH.29	49.7 ± 2.4	50.3 ± 3.7	1.6 ± 1.8	3.1 ± 2.0	99.0 ± 4.2	100.7 ± 4.1	0.6 ± 1.4	1.4 ± 1.6	114.8 ± 5.7	117.3 ± 11.4	1.4 ± 3.6	1.7 ± 3.7	3521.0 ± 296.2	4660.4 ± 745.7	5.4 ± 8.5	-9.3 ± 10.0
RR.FF.IN/TH.30	49.9 ± 2.4	48.9 ± 3.7	-1.5 ± 1.8	0.4 ± 2.0	100.8 ± 4.2	100.2 ± 4.1	-0.4 ± 1.4	0.4 ± 1.6	112.8 ± 5.7	110.0 ± 11.4	-2.5 ± 3.6	-1.0 ± 3.7	3529.4 ± 299.2	4385.0 ± 765.7	3.4 ± 8.5	-10.8 ± 10.0
RR.FF.IN/TH.31	49.7 ± 2.4	48.3 ± 3.7	-1.3 ± 1.8	-0.2 ± 2.0	99.4 ± 4.2	99.4 ± 4.1	-0.5 ± 1.4	0.8 ± 1.6	114.0 ± 5.7	115.2 ± 11.4	1.1 ± 3.6	4.1 ± 3.7	3544.5 ± 296.1	5397.0 ± 804.5	0.5 ± 8.6	-4.6 ± 10.0
RR.FF.IN/TH.33	50.0 ± 2.4	48.7 ± 3.7	-1.7 ± 1.8	0.3 ± 2.0	100.4 ± 4.3	100.1 ± 4.1	-0.8 ± 1.4	0.7 ± 1.6	113.6 ± 5.9	116.7 ± 11.4	3.2 ± 3.6	1.7 ± 3.7	3509.4 ± 307.1	5344.6 ± 745.9	13.0 ± 8.5	7.6 ± 10.0
RR.FF.IN/TH.34	50.1 ± 2.4	48.9 ± 3.7	-1.1 ± 1.8	-0.9 ± 2.0	101.3 ± 4.3	100 ± 4.1	-1.5 ± 1.4	-2.1 ± 1.6	114.6 ± 5.9	117.7 ± 11.4	2.6 ± 3.6	1.3 ± 3.7	3380.2 ± 306.9	4809.6 ± 767.3	0.3 ± 10.0	-1.7 ± 11.3
RR.FF.IN/TH.35	51.4 ± 2.4	48.8 ± 3.7	-1.3 ± 1.8	2.2 ± 2.0	102.5 ± 4.3	99.0 ± 4.1	-2.1 ± 1.4	0.1 ± 1.6	124.9 ± 5.9	116.6 ± 11.4	-1.5 ± 3.6	2.1 ± 3.7	3279.2 ± 306.8	5181.7 ± 747.4	37.0 ± 9.9	4.4 ± 11.3
RR.FF.IN/TH.37	50.2 ± 2.4	48.3 ± 3.7	-2.3 ± 1.8	-1.8 ± 2.0	100.4 ± 4.3	98.7 ± 4.1	-2.3 ± 1.4	-1.2 ± 1.6	114.4 ± 5.9	116.2 ± 11.4	-2.0 ± 3.6	-3.9 ± 3.7	3737.1 ± 307.1	4293.6 ± 745.9	-4.7 ± 8.6	-5.8 ± 10.1
RR.FF.IN/TH.38	49.1 ± 2.4	46.8 ± 3.7	-3.2 ± 1.8	-4.4 ± 2.0	101.0 ± 4.3	98.1 ± 4.1	-2.4 ± 1.4	-1.8 ± 1.6	108.2 ± 5.9	106.9 ± 11.5	-2.4 ± 3.8	-1.4 ± 4.0	2770.0 ± 306.9	4126.7 ± 747.1	7.1 ± 8.6	-14.3 ± 10.1
A07-26NR	49.1 ± 2.4	48.8 ± 3.7			99.9 ± 4.2	100.6 ± 4			114.5 ± 5.3	115.8 ± 11.1			3675.5 ± 274.3	4816.6 ± 671.6		

[1] DTF = days to flowering and DTM = days to maturity.

[2] RR.FF.IN/TH = F_2-derived inbred line (IN) of Rutabaga-BF × A07-26NR and its test hybrid (TH) of A07-26NR × IN. For example, RR.FF.IN/TH.13 shows data of the inbred line RR.FF.IN.13 (see Table A3.17 for details) and its test hybrid.

Table A3.12: Seed quality traits of the inbred lines (IN) derived from BC_1 of the Rutabaga-BF × Hi-Q cross and their test hybrids (TH), produced from the Hi-Q × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q (CPH) for these traits.

Inbred Line / Test Hybrid	Seed oil content (%) Mean ± SE		MPH	CPH	Seed protein content (%) Mean ± SE		MPH	CPH	Seed glucosinolate content (µmol/g) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TH	(%)	
CO.BC.IN/TH.03[1]	47.7 ± 0.8	48.6 ± 1.1	0.1 ± 1.5	0.3 ± 1.5	24.2 ± 1.2	25.1 ± 0.9	-0.9 ± 1.9	-2.5 ± 2.3	16.1 ± 0.8	12.9 ± 1.0	-2.9 ± 5.5	-1.3 ± 5.8
CO.BC.IN/TH.04	48.0 ± 0.9	47.7 ± 1.1	2.7 ± 1.5	2.3 ± 1.5	23.8 ± 1.2	25.8 ± 0.9	-5.2 ± 1.9	-5.7 ± 2.3	14.5 ± 0.9	13.0 ± 1.0	-6.1 ± 5.5	-7.5 ± 5.8
CO.BC.IN/TH.06	46.2 ± 0.8	47.7 ± 1.1	0.8 ± 1.5	0.5 ± 1.5	25.4 ± 1.2	26.2 ± 0.9	-1.3 ± 1.9	-2.4 ± 2.3	12.4 ± 0.8	12.1 ± 1.1	0.3 ± 5.5	-0.1 ± 5.8
CO.BC.IN/TH.07	48.2 ± 0.8	49.0 ± 1.1	0.9 ± 1.5	3.4 ± 1.5	24.0 ± 1.2	25.2 ± 0.9	-1.9 ± 1.9	-5.3 ± 2.3	23.5 ± 0.8	22.7 ± 1.1	41.8 ± 6.2	56.0 ± 6.6
CO.BC.IN/TH.08	47.1 ± 0.8	49.4 ± 1.1	3.3 ± 1.5	4.3 ± 1.5	23.8 ± 1.2	23.5 ± 0.9	-7.6 ± 1.9	-11.0 ± 2.3	16.0 ± 0.8	14.5 ± 1.0	-0.3 ± 5.5	10.3 ± 5.8
CO.BC.IN/TH.09	45.9 ± 0.8	46.5 ± 1.1	0.3 ± 1.5	-1.4 ± 1.5	24.6 ± 1.2	25.9 ± 0.9	-1.2 ± 1.9	-2.9 ± 2.3	13.5 ± 0.8	12.4 ± 1.1	-1.4 ± 5.5	-7.1 ± 5.8
CO.BC.IN/TH.16	48.4 ± 0.9	48.3 ± 1.2	0.6 ± 1.7	0.8 ± 1.7	23.3 ± 1.2	25.3 ± 1.0	0.4 ± 2.2	-4.3 ± 2.6	13.9 ± 0.9	13.5 ± 1.2	6.4 ± 6.2	5.6 ± 6.6
CO.BC.IN/TH.18	47.4 ± 0.8	48.4 ± 1.0	1.5 ± 1.2	1.2 ± 1.1	23.8 ± 1.2	24.9 ± 0.9	-3.1 ± 1.5	-4.4 ± 1.8	15.4 ± 0.8	11.2 ± 0.8	-9.7 ± 4.4	-17.6 ± 4.6
CO.BC.IN/TH.19	48.0 ± 0.8	47.5 ± 1.0	0.6 ± 1.2	0.6 ± 1.1	23.8 ± 1.2	25.5 ± 0.9	-0.4 ± 1.5	-3.0 ± 1.8	13.7 ± 0.8	13.2 ± 0.8	-0.3 ± 4.4	3.3 ± 4.6
CO.BC.IN/TH.20	46.2 ± 0.8	47.0 ± 1.0	0.2 ± 1.2	-0.2 ± 1.1	25.0 ± 1.2	26.1 ± 0.9	-2.5 ± 1.5	-2.3 ± 1.8	14.5 ± 0.8	12.7 ± 0.8	-1.8 ± 4.4	-2.2 ± 4.6
CO.BC.IN/TH.21	46.4 ± 0.8	46.9 ± 1.0	-0.9 ± 1.2	-1.9 ± 1.1	24.6 ± 1.2	25.7 ± 0.9	-2.4 ± 1.5	-2.0 ± 1.7	13.0 ± 0.8	11.7 ± 0.8	-5.9 ± 4.4	-10.8 ± 4.6
CO.BC.IN/TH.22	45.9 ± 0.8	47.4 ± 1.0	2.9 ± 1.2	0.4 ± 1.2	24.8 ± 1.2	26.1 ± 0.9	-1.9 ± 1.5	-2.4 ± 1.8	14.7 ± 0.8	12.4 ± 0.8	-7.3 ± 4.4	-7.9 ± 4.6
CO.BC.IN/TH.23	45.7 ± 0.8	46.5 ± 1.0	0.3 ± 1.2	-2.3 ± 1.1	25.6 ± 1.2	26.7 ± 0.9	-1.1 ± 1.5	0.2 ± 1.7	14.6 ± 0.8	12.3 ± 0.8	-3.8 ± 4.4	-8.3 ± 4.6
CO.BC.IN/TH.24	45.7 ± 0.8	47.1 ± 1.0	0.6 ± 1.2	-1.8 ± 1.2	26.9 ± 1.2	27.0 ± 0.9	0.0 ± 1.5	4.5 ± 1.8	16.1 ± 0.8	13.3 ± 0.8	-2.9 ± 4.4	0.0 ± 4.6
CO.BC.IN/TH.25	46.1 ± 0.8	48.3 ± 1.0	3.7 ± 1.2	2.8 ± 1.1	27.1 ± 1.2	27.5 ± 0.9	-1.2 ± 1.5	2.6 ± 1.7	15.2 ± 0.8	13.4 ± 0.8	-2.9 ± 4.4	1.3 ± 4.6
CO.BC.IN/TH.26	46.2 ± 0.8	47.0 ± 1.0	1.4 ± 1.2	-0.1 ± 1.2	27.0 ± 1.2	27.6 ± 0.9	-3.4 ± 1.6	2.4 ± 1.8	15.2 ± 0.8	13.2 ± 0.8	-6.8 ± 4.7	-1.4 ± 4.6
CO.BC.IN/TH.27	45.9 ± 0.9	46.5 ± 1.0	1.1 ± 1.2	-1.2 ± 1.1	27.1 ± 1.2	28.0 ± 0.9	-1.0 ± 1.5	4.7 ± 1.8	15.1 ± 0.9	13.7 ± 0.8	-3.6 ± 4.4	1.9 ± 4.6
CO.BC.IN/TH.28	45.8 ± 0.8	46.4 ± 1.0	-0.2 ± 1.2	-2.2 ± 1.2	25.0 ± 1.2	26.4 ± 0.9	-0.8 ± 1.5	-0.6 ± 1.8	13.3 ± 0.8	12.7 ± 0.8	-2.6 ± 4.4	-8.3 ± 4.6
CO.BC.IN/TH.29	47.2 ± 0.9	47.0 ± 1.0	1.0 ± 1.2	0.0 ± 1.2	24.2 ± 1.2	25.9 ± 0.9	-3.1 ± 1.5	-3.9 ± 1.8	15.4 ± 0.9	15.5 ± 0.8	0.7 ± 4.4	10.6 ± 4.6
CO.BC.IN/TH.31	46.4 ± 0.9	47.4 ± 1.0	2.1 ± 1.2	0.3 ± 1.1	27.2 ± 1.2	27.5 ± 0.9	-2.7 ± 1.5	2.4 ± 1.8	15.7 ± 0.9	13.0 ± 0.8	-9.7 ± 4.4	-3.5 ± 4.6
CO.BC.IN/TH.32	46.9 ± 0.8	47.8 ± 1.0	1.1 ± 1.2	0.3 ± 1.1	26.8 ± 1.2	27.1 ± 0.9	-2.7 ± 1.5	2.3 ± 1.7	17.3 ± 0.8	13.7 ± 0.8	-8.3 ± 4.4	0.6 ± 4.6
CO.BC.IN/TH.33	46.1 ± 0.9	47.2 ± 1.0	-0.3 ± 1.2	-1.3 ± 1.1	24.9 ± 1.2	25.9 ± 0.9	-0.8 ± 1.5	-0.3 ± 1.8	13.3 ± 0.9	11.8 ± 0.8	-4.9 ± 4.4	-10.9 ± 4.6
CO.BC.IN/TH.34	47.8 ± 0.9	48.1 ± 1.0	1.7 ± 1.2	2.1 ± 1.1	23.5 ± 1.2	25.8 ± 0.9	-1.1 ± 1.5	-3.1 ± 1.8	14.0 ± 0.9	12.9 ± 0.8	-12.1 ± 4.4	-8.5 ± 4.6
CO.BC.IN/TH.36	44.5 ± 0.8	45.3 ± 1.1	-0.8 ± 1.2	-4.6 ± 1.2	26.3 ± 1.2	27.7 ± 0.9	1.4 ± 1.6	3.6 ± 1.9	14.3 ± 0.8	12.9 ± 0.9	-6.4 ± 4.7	-6.6 ± 4.9
CO.BC.IN/TH.37	46.4 ± 0.8	45.8 ± 1.1	-1.7 ± 1.3	-3.4 ± 1.3	24.5 ± 1.2	26.3 ± 0.9	-0.4 ± 1.6	-0.9 ± 1.9	13.1 ± 0.8	12.7 ± 0.9	-3.7 ± 4.7	-5.3 ± 4.9
Hi-Q	47.0 ± 0.8	47.2 ± 1.0			25.1 ± 1.1	26.9 ± 0.8			14.3 ± 0.7	13.8 ± 0.6		

[1] CO.BC.IN/TH = BC_1-derived inbred line (IN) of Rutabaga-BF × Hi-Q and its test hybrid (TH) of Hi-Q × IN. For example, CO.BC.IN/TH.03 shows data of the inbred line CO.BC.IN.03 (see Table A3.16 for details) and its test hybrid.

Table A3.13: Seed quality traits of the inbred lines (IN) derived from F_2 of the Rutabaga-BF × Hi-Q cross and their test hybrids (TH), produced from the Hi-Q × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over Hi-Q (CPH) for these traits.

Inbred Line / Test Hybrid	Seed oil content (%) Mean ± SE		MPH	CPH	Seed protein content (%) Mean ± SE		MPH	CPH	Seed glucosinolate content (µmol/g) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TH	(%)	
CO.FF.IN/TH.13[1]	44.9 ± 0.8	45.4 ± 1.0	0.0 ± 1.2	-5.0 ± 1.1	25.0 ± 1.2	26.8 ± 0.9	-0.2 ± 1.5	2.1 ± 1.7	14.9 ± 0.8	14.4 ± 0.8	3.1 ± 4.4	4.4 ± 4.6
CO.FF.IN/TH.15	47.4 ± 0.8	47.1 ± 1.0	0.5 ± 1.2	0.7 ± 1.1	25.0 ± 1.2	26.5 ± 0.9	-1.8 ± 1.5	-1.6 ± 1.7	25.5 ± 0.8	20.9 ± 0.8	-2.0 ± 4.4	6.2 ± 6.6
CO.FF.IN/TH.16	44.4 ± 0.8	45.5 ± 1.0	1.2 ± 1.2	-3.7 ± 1.1	25.9 ± 1.2	26.5 ± 0.9	-1.9 ± 1.5	-0.6 ± 1.7	14.9 ± 0.8	12.9 ± 0.8	-7.8 ± 4.4	-8.2 ± 4.6
CO.FF.IN/TH.17	48.6 ± 0.9	48.9 ± 1.0	1.5 ± 1.2	3.5 ± 1.1	25.3 ± 1.2	26.2 ± 0.9	-1.0 ± 1.5	-0.6 ± 1.8	15.0 ± 0.9	13.2 ± 0.8	-0.6 ± 4.4	-1.3 ± 4.6
CO.FF.IN/TH.18	48.2 ± 0.9	49.9 ± 1.0	4.1 ± 1.2	5.7 ± 1.1	25.5 ± 1.2	26.0 ± 0.9	-0.8 ± 1.5	-1.8 ± 1.7	15.0 ± 0.9	13.6 ± 0.8	1.1 ± 4.4	1.7 ± 4.6
CO.FF.IN/TH.19	44.9 ± 0.8	45.1 ± 1.0	-0.4 ± 1.2	-3.7 ± 1.1	25.8 ± 1.2	26.6 ± 0.9	-2.2 ± 1.5	-1.6 ± 1.7	13.9 ± 0.8	12.8 ± 0.8	-2.9 ± 4.4	-5.2 ± 4.6
CO.FF.IN/TH.20	47.2 ± 0.8	46.2 ± 1.0	-0.5 ± 1.2	-1.2 ± 1.1	24.8 ± 1.2	27.3 ± 0.9	-1.0 ± 1.5	1.6 ± 1.8	15.7 ± 0.8	12.6 ± 0.8	-7.4 ± 4.4	-8.1 ± 4.6
CO.FF.IN/TH.21	47.0 ± 0.8	46.5 ± 1.0	-1.0 ± 1.2	-2.5 ± 1.1	25.1 ± 1.2	26.7 ± 0.9	-0.5 ± 1.5	1.7 ± 1.7	20.3 ± 0.8	20.7 ± 0.8	3.8 ± 4.7	-13.1 ± 5.8
CO.FF.IN/TH.22	47.0 ± 0.9	48.0 ± 1.0	1.8 ± 1.2	2.3 ± 1.1	25.9 ± 1.2	26.7 ± 0.9	-0.6 ± 1.5	-0.7 ± 1.7	17.8 ± 0.9	14.9 ± 0.8	-1.4 ± 4.4	11.3 ± 4.6
CO.FF.IN/TH.23	45.2 ± 0.9	46.8 ± 1.0	1.8 ± 1.1	-0.3 ± 1.1	26.1 ± 1.2	27.2 ± 0.9	-2.4 ± 1.5	0.6 ± 1.7	16.7 ± 0.9	15.1 ± 0.8	-3.8 ± 4.4	11.7 ± 4.6
CO.FF.IN/TH.24	47.9 ± 0.9	47.5 ± 1.0	0.4 ± 1.2	-0.6 ± 1.1	26.5 ± 1.2	27.2 ± 0.9	-1.3 ± 1.5	3.9 ± 1.7	15.4 ± 0.9	13.1 ± 0.8	-9.6 ± 4.4	-4.8 ± 4.6
CO.FF.IN/TH.25	47.7 ± 0.8	48.0 ± 1.0	2.0 ± 1.2	2.0 ± 1.1	26.6 ± 1.2	27.3 ± 0.9	-1.6 ± 1.5	2.7 ± 1.7	14.7 ± 0.8	13.9 ± 0.8	-3.3 ± 4.4	1.9 ± 4.6
CO.FF.IN/TH.26	47.2 ± 0.9	47.9 ± 1.0	2.3 ± 1.2	2.4 ± 1.1	26.4 ± 1.2	27.1 ± 0.9	-2.4 ± 1.5	1.5 ± 1.7	15.0 ± 0.9	13.5 ± 0.8	-7.5 ± 4.4	-0.5 ± 4.6
CO.FF.IN/TH.27	45.5 ± 0.9	45.9 ± 1.0	-0.8 ± 1.2	-2.3 ± 1.1	26.1 ± 1.2	27.6 ± 0.9	0.0 ± 1.5	2.4 ± 1.7	14.7 ± 0.9	15.0 ± 0.8	9.5 ± 4.4	5.8 ± 4.6
CO.FF.IN/TH.28	44.8 ± 0.9	46.6 ± 1.0	-0.1 ± 1.2	-1.8 ± 1.1	26.5 ± 1.2	27.9 ± 0.9	2.5 ± 1.5	4.4 ± 1.7	16.1 ± 0.9	13.2 ± 0.8	-7.3 ± 4.4	-8.0 ± 4.6
CO.FF.IN/TH.29	47.1 ± 0.9	48.0 ± 1.0	1.7 ± 1.2	2.2 ± 1.2	24.8 ± 1.2	26.1 ± 0.9	-1.8 ± 1.5	-3.1 ± 1.8	17.3 ± 0.9	12.2 ± 0.8	-5.4 ± 4.4	-8.5 ± 4.6
CO.FF.IN/TH.30	47.8 ± 0.9	48.4 ± 1.0	2.9 ± 1.2	3.1 ± 1.1	25.6 ± 1.2	26.5 ± 0.9	-3.4 ± 1.5	-1.7 ± 1.7	13.7 ± 0.9	12.1 ± 0.8	-9.0 ± 4.4	-7.1 ± 4.6
CO.FF.IN/TH.31	47.5 ± 0.8	47.4 ± 1.1	0.5 ± 1.2	0.4 ± 1.2	26.6 ± 1.2	27.1 ± 0.9	-2.2 ± 1.6	1.6 ± 1.9	15.1 ± 0.8	13.5 ± 0.9	-5.5 ± 4.7	-0.4 ± 4.9
CO.FF.IN/TH.32	46.8 ± 0.8	47.4 ± 1.0	1.0 ± 1.2	1.8 ± 1.1	25.8 ± 1.2	26.9 ± 0.9	-2.2 ± 1.5	-2.3 ± 1.7	15.8 ± 0.8	12.7 ± 0.8	-9.8 ± 4.4	-7.3 ± 4.6
CO.FF.IN/TH.33	44.6 ± 0.9	46.1 ± 1.0	0.2 ± 1.2	-2.6 ± 1.1	26.7 ± 1.2	28.3 ± 0.9	1.2 ± 1.5	5.2 ± 1.8	15.8 ± 0.9	12.8 ± 0.8	-7.3 ± 4.4	-9.3 ± 4.6
CO.FF.IN/TH.34	44.9 ± 0.8	45.1 ± 1.0	-0.7 ± 1.2	-5.4 ± 1.1	26.1 ± 1.2	26.7 ± 0.9	-1.6 ± 1.5	1.6 ± 1.7	15.5 ± 0.8	13.2 ± 0.8	-1.6 ± 4.4	-2.0 ± 4.6
CO.FF.IN/TH.35	45.4 ± 0.9	46.8 ± 1.0	2.7 ± 1.3	-0.4 ± 1.2	26.4 ± 1.2	28.3 ± 0.9	3.7 ± 1.7	6.7 ± 1.9	15.3 ± 0.9	14.0 ± 0.8	1.3 ± 5.0	2.6 ± 4.9
CO.FF.IN/TH.36	45.1 ± 0.9	46.6 ± 1.0	0.2 ± 1.2	-1.9 ± 1.1	26.3 ± 1.2	27.7 ± 0.9	1.5 ± 1.5	4.7 ± 1.7	16.3 ± 0.9	12.5 ± 0.8	-6.1 ± 4.4	-6.7 ± 4.6
CO.FF.IN/TH.38	44.5 ± 0.9	44.8 ± 1.0	-1.4 ± 1.2	-5.7 ± 1.2	26.7 ± 1.2	28.9 ± 0.9	2.2 ± 1.5	7.8 ± 1.8	15.6 ± 0.9	13.6 ± 0.8	2.3 ± 4.4	1.7 ± 4.6
CO.FF.IN/TH.39	48.1 ± 0.9	48.9 ± 1.0	2.9 ± 1.2	4.4 ± 1.1	25.6 ± 1.2	26.2 ± 0.9	-2.0 ± 1.6	-2.9 ± 1.7	15.4 ± 0.9	14.1 ± 0.8	0.0 ± 4.7	0.4 ± 4.6
CO.FF.IN/TH.40	48.8 ± 0.9	49.2 ± 1.0	2.0 ± 1.2	4.4 ± 1.1	25.0 ± 1.2	25.9 ± 0.9	-2.0 ± 1.5	-2.6 ± 1.8	15.1 ± 0.9	12.9 ± 0.8	-3.8 ± 4.4	-2.4 ± 4.6
Hi-Q	47.0 ± 0.8	47.2 ± 1.0			25.1 ± 1.1	26.9 ± 0.8			14.3 ± 0.7	13.8 ± 0.6		

[1] CO.FF.IN/TH = F_2-derived inbred line (IN) of Rutabaga-BF × Hi-Q and its test hybrid (TH) of Hi-Q × IN. For example, CO.FF.IN/TH.13 shows data of the inbred line CO.FF.IN.13 (see Table A3.16 for details) and its test hybrid.

Table A3.14: Seed quality traits of the inbred lines (IN) derived from BC_1 of the Rutabaga-BF × A07-26NR cross and their test hybrids (TH), produced from the A07-26NR × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over A07-26NR (CPH) for these traits.

Inbred Line / Test Hybrid	Seed oil content (%) Mean ± SE		MPH	CPH	Seed protein ontent (%) Mean ± SE		MPH	CPH	Seed glucosinolate ontent (µmol/g) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TH	(%)	
RR.BC.IN/TH.01[1]	46.6 ± 0.9	46.7 ± 1.2	0.7 ± 1.7	-4.4 ± 1.7	25.0 ± 1.2	27.2 ± 1.0	-0.1 ± 2.2	6.5 ± 2.6	16.9 ± 0.9	16.9 ± 1.2	-1.3 ± 6.2	15.5 ± 6.6
RR.BC.IN/TH.03	48.6 ± 0.8	47.2 ± 1.1	1.1 ± 1.5	-1.1 ± 1.5	24.6 ± 1.2	27.1 ± 0.9	-0.3 ± 1.9	4.3 ± 2.3	15.3 ± 0.8	15.8 ± 1.0	-10.8 ± 5.5	9.4 ± 5.8
RR.BC.IN/TH.05	48.6 ± 0.8	47.2 ± 1.1	0.7 ± 1.5	-0.5 ± 1.5	23.4 ± 1.2	26.8 ± 0.9	0.5 ± 1.9	1.3 ± 2.3	13.4 ± 0.8	15.3 ± 1.0	-8.0 ± 5.5	-4.7 ± 5.8
RR.BC.IN/TH.14	47.0 ± 0.8	46.7 ± 1.0	0.8 ± 1.2	-2.9 ± 1.1	25.5 ± 1.2	27.1 ± 0.9	-1.5 ± 1.5	3.5 ± 1.8	17.4 ± 0.8	16.9 ± 0.8	-5.7 ± 4.4	10.3 ± 4.6
RR.BC.IN/TH.15	48.4 ± 0.8	47.4 ± 1.0	-1.2 ± 1.2	-1.4 ± 1.1	24.2 ± 1.2	25.6 ± 0.9	-0.7 ± 1.5	-1.7 ± 1.7	15.3 ± 0.8	15.3 ± 0.8	1.6 ± 4.4	3.7 ± 4.6
RR.BC.IN/TH.17	48.9 ± 0.8	48.2 ± 1.0	0.7 ± 1.2	-1.1 ± 1.1	22.8 ± 1.2	25.4 ± 0.9	0.0 ± 1.5	-0.7 ± 1.7	17.1 ± 0.8	16.2 ± 0.8	-7.5 ± 4.4	6.6 ± 4.6
RR.BC.IN/TH.18	48.0 ± 0.8	47.9 ± 1.0	1.9 ± 1.2	-0.6 ± 1.1	23.5 ± 1.2	25.3 ± 0.9	-2.6 ± 1.5	-3.2 ± 1.7	14.7 ± 0.8	14.6 ± 0.8	-8.2 ± 4.4	-5.2 ± 4.6
RR.BC.IN/TH.19	46.4 ± 0.9	46.0 ± 1.0	-0.3 ± 1.2	-3.9 ± 1.1	27.1 ± 1.2	28.0 ± 0.9	-0.9 ± 1.5	6.8 ± 1.7	13.8 ± 0.9	13.8 ± 0.8	-8.5 ± 4.4	-13.8 ± 4.6
RR.BC.IN/TH.21	49.1 ± 0.8	47.6 ± 1.0	1.2 ± 1.2	0.2 ± 1.1	22.4 ± 1.2	25.2 ± 0.9	-3.3 ± 1.5	-4.3 ± 1.7	16.2 ± 0.8	16.7 ± 0.8	5.9 ± 4.4	7.0 ± 4.6
RR.BC.IN/TH.22	48.5 ± 0.8	48.0 ± 1.0	1.1 ± 1.2	0.9 ± 1.1	23.1 ± 1.2	25.8 ± 0.9	-1.4 ± 1.5	-2.5 ± 1.7	20.2 ± 0.8	20.0 ± 0.8	6.5 ± 4.4	25.6 ± 4.6
RR.BC.IN/TH.23	46.7 ± 0.9	46.4 ± 1.0	-0.2 ± 1.2	-3.9 ± 1.1	24.9 ± 1.2	27.2 ± 0.9	0.6 ± 1.6	4.4 ± 1.7	19.5 ± 0.9	23.7 ± 0.8	8.6 ± 5.5	30.8 ± 7.9
RR.BC.IN/TH.24	46.3 ± 0.9	46.4 ± 1.0	1.8 ± 1.2	-3.7 ± 1.1	25.3 ± 1.2	27.1 ± 0.9	-2.9 ± 1.6	5.5 ± 1.7	16.1 ± 0.9	14.3 ± 0.8	-9.5 ± 4.7	-4.8 ± 4.6
RR.BC.IN/TH.25	46.4 ± 0.8	43.9 ± 1.0	-2.5 ± 1.2	-8.0 ± 1.1	24.7 ± 1.2	28.2 ± 0.9	1.9 ± 1.5	6.3 ± 1.8	24.3 ± 0.8	24.4 ± 0.8	8.6 ± 4.4	48.0 ± 5.3
RR.BC.IN/TH.26	47.8 ± 0.9	47.5 ± 1.0	-0.4 ± 1.2	-0.8 ± 1.2	24.2 ± 1.2	25.8 ± 0.9	-1.4 ± 1.6	-0.9 ± 1.9	14.7 ± 0.9	13.9 ± 0.8	-9.8 ± 4.7	-12.7 ± 4.9
RR.BC.IN/TH.27	47.1 ± 0.9	45.1 ± 1.0	-1.3 ± 1.2	-5.1 ± 1.1	26.3 ± 1.2	28.8 ± 0.9	1.2 ± 1.5	9.2 ± 1.7	13.8 ± 0.9	14.7 ± 0.8	-2.9 ± 4.4	-7.1 ± 4.6
RR.BC.IN/TH.28	49.3 ± 0.9	48.6 ± 1.0	0.7 ± 1.2	0.8 ± 1.2	23.3 ± 1.2	25.5 ± 0.9	0.0 ± 1.6	-0.6 ± 1.9	15.3 ± 0.9	14.7 ± 0.8	-9.5 ± 4.7	-7.3 ± 4.9
RR.BC.IN/TH.29	47.4 ± 0.8	47.9 ± 1.0	0.6 ± 1.2	-0.5 ± 1.1	24.7 ± 1.2	26.2 ± 0.9	-0.4 ± 1.5	1.2 ± 1.7	16.3 ± 0.8	16.4 ± 0.8	1.9 ± 4.4	4.6 ± 4.6
RR.BC.IN/TH.30	47.4 ± 1.0	46.7 ± 1.0	1.1 ± 1.2	-3.5 ± 1.1	23.9 ± 1.2	26.4 ± 0.9	-1.5 ± 1.6	2.7 ± 1.7	14.2 ± 1.2	14.5 ± 0.8	-4.2 ± 4.7	-4.4 ± 4.6
RR.BC.IN/TH.31	47.4 ± 0.8	46.2 ± 1.0	0.5 ± 1.2	-3.8 ± 1.1	26.2 ± 1.2	28.4 ± 0.9	0.7 ± 1.5	8.2 ± 1.7	15.4 ± 0.8	15.8 ± 0.8	-7.4 ± 4.4	0.2 ± 4.6
RR.BC.IN/TH.32	48.4 ± 0.9	48.1 ± 1.1	2.0 ± 1.2	0.0 ± 1.2	23.3 ± 1.2	25.0 ± 0.9	-3.2 ± 1.6	-3.5 ± 1.9	16.5 ± 0.9	16.3 ± 0.9	-16.4 ± 5.0	-7.3 ± 5.2
RR.BC.IN/TH.33	45.8 ± 0.9	45.4 ± 1.1	-0.6 ± 1.3	-6.9 ± 1.3	25.9 ± 1.2	28.2 ± 0.9	0.7 ± 1.7	9.8 ± 2.0	17.4 ± 0.9	17.2 ± 0.9	-8.7 ± 5.0	10.7 ± 5.3
A07-26NR	49.2 ± 0.8	47.8 ± 1.0			23.7 ± 1.1	26.2 ± 0.8			14.5 ± 0.7	15.5 ± 0.6		

[1] RR.BC.IN/TH = BC_1-derived inbred line (IN) of Rutabaga-BF × A07-26NR and its test hybrid (TH) of A07-26NR × IN. For example, RR.BC.IN/TH.01 shows data of the inbred line RR.BC.IN.01 (see Table A3.17 for details) and its test hybrid.

Table A3.15: Seed quality traits of the inbred lines (IN) derived from F_2 of the Rutabaga-BF × A07-26NR cross and their test hybrids (TH), produced from the A07-26NR × inbred line crosses, and mid-parent heterosis (MPH) and heterosis over A07-26NR (CPH) for these traits.

Inbred Line / Test Hybrid	Seed oil content (%) Mean ± SE		MPH	CPH	Seed protein content (%) Mean ± SE		MPH	CPH	Seed glucosinolate content (µmol/g) Mean ± SE		MPH	CPH
	IN	TH	(%)		IN	TH	(%)		IN	TH	(%)	
RR.FF.IN/TH.13[1]	46.4 ± 0.9	46.0 ± 1.1	-0.1 ± 1.5	-4.6 ± 1.5	26.1 ± 1.2	27.9 ± 0.9	2.6 ± 1.9	8.1 ± 2.3	16.1 ± 0.9	15.0 ± 1.0	-7.0 ± 5.5	1.9 ± 5.8
RR.FF.IN/TH.17	46.5 ± 0.9	46.8 ± 1.0	1.2 ± 1.2	-1.7 ± 1.1	25.4 ± 1.2	26.3 ± 0.9	-1.8 ± 1.5	0.7 ± 1.7	15.4 ± 0.9	14.7 ± 0.8	-4.3 ± 4.4	-2.1 ± 4.6
RR.FF.IN/TH.18	46.8 ± 0.9	46.2 ± 1.0	1.4 ± 1.2	-4.3 ± 1.1	25.9 ± 1.2	27.7 ± 0.9	0.0 ± 1.5	7.3 ± 1.7	16.0 ± 0.9	16.1 ± 0.8	-0.9 ± 4.4	6.6 ± 4.6
RR.FF.IN/TH.19	50.0 ± 0.9	48.4 ± 1.0	0.5 ± 1.2	0.2 ± 1.1	23.7 ± 1.2	26.1 ± 0.9	-0.6 ± 1.5	0.9 ± 1.7	14.7 ± 0.9	14.0 ± 0.8	-10.9 ± 4.4	-10.3 ± 4.6
RR.FF.IN/TH.20	49.8 ± 0.8	48.3 ± 1.0	1.0 ± 1.1	1.4 ± 1.1	23.9 ± 1.2	26.0 ± 0.8	-2.6 ± 1.5	-1.6 ± 1.7	14.7 ± 0.8	14.4 ± 0.8	-11.0 ± 4.4	-7.1 ± 4.6
RR.FF.IN/TH.21	49.8 ± 0.9	47.3 ± 1.0	-1.4 ± 1.2	-1.6 ± 1.1	23.9 ± 1.2	27.0 ± 0.9	1.7 ± 1.5	3.3 ± 1.8	15.1 ± 0.9	15.7 ± 0.8	-5.7 ± 4.4	0.3 ± 4.6
RR.FF.IN/TH.22	49.9 ± 0.8	48.2 ± 1.0	-0.9 ± 1.2	0.2 ± 1.1	23.4 ± 1.2	26.3 ± 0.9	1.8 ± 1.6	0.5 ± 1.8	15.0 ± 0.8	16.6 ± 0.8	0.9 ± 4.7	7.4 ± 4.6
RR.FF.IN/TH.23	50.4 ± 0.8	48.5 ± 1.0	0.3 ± 1.2	0.7 ± 1.1	23.4 ± 1.2	26.1 ± 0.9	-0.6 ± 1.5	0.8 ± 1.7	14.4 ± 0.8	14.7 ± 0.8	-7.8 ± 4.4	-4.6 ± 4.6
RR.FF.IN/TH.24	49.8 ± 0.9	47.8 ± 1.0	-0.2 ± 1.2	0.2 ± 1.1	23.9 ± 1.2	26.7 ± 0.9	0.7 ± 1.5	1.2 ± 1.7	15.5 ± 0.9	16.8 ± 0.8	-4.5 ± 4.4	4.2 ± 4.6
RR.FF.IN/TH.25	49.5 ± 0.8	46.6 ± 1.0	-2.6 ± 1.2	-2.0 ± 1.1	24.0 ± 1.2	27.6 ± 0.9	2.4 ± 1.5	3.6 ± 1.7	14.7 ± 0.8	15.4 ± 0.8	-8.4 ± 4.4	-5.7 ± 4.6
RR.FF.IN/TH.26	50.2 ± 0.8	48.2 ± 1.0	0.4 ± 1.2	0.4 ± 1.1	23.4 ± 1.2	26.5 ± 0.9	0.6 ± 1.5	1.1 ± 1.8	14.6 ± 0.8	14.7 ± 0.8	-6.6 ± 4.4	-3.5 ± 4.6
RR.FF.IN/TH.27	49.7 ± 0.9	48.2 ± 1.0	1.0 ± 1.2	0.6 ± 1.1	23.7 ± 1.2	26.3 ± 0.9	0.0 ± 1.5	0.8 ± 1.7	14.6 ± 0.9	16.0 ± 0.8	1.0 ± 4.4	4.2 ± 4.6
RR.FF.IN/TH.28	49.8 ± 0.9	47.2 ± 1.0	-1.0 ± 1.1	-1.3 ± 1.1	23.4 ± 1.2	27.0 ± 0.9	3.0 ± 1.5	2.8 ± 1.7	14.9 ± 0.9	15.4 ± 0.8	-2.7 ± 4.4	4.7 ± 4.6
RR.FF.IN/TH.29	49.8 ± 0.9	47.3 ± 1.0	0.0 ± 1.1	-0.7 ± 1.1	23.5 ± 1.2	27.2 ± 0.8	1.6 ± 1.5	3.4 ± 1.7	14.6 ± 0.9	16.5 ± 0.8	2.1 ± 4.4	10.1 ± 4.6
RR.FF.IN/TH.30	49.3 ± 0.9	47.3 ± 1.0	-0.7 ± 1.2	-2.0 ± 1.1	23.9 ± 1.2	26.7 ± 0.9	1.3 ± 1.5	3.2 ± 1.7	14.9 ± 0.9	15.6 ± 0.8	-0.5 ± 4.4	3.5 ± 4.6
RR.FF.IN/TH.31	49.1 ± 0.9	48.1 ± 1.0	1.1 ± 1.2	-0.5 ± 1.1	24.5 ± 1.2	27.1 ± 0.9	1.4 ± 1.5	4.8 ± 1.8	15.0 ± 0.9	16.5 ± 0.8	0.5 ± 4.4	11.4 ± 4.6
RR.FF.IN/TH.33	48.9 ± 0.9	47.0 ± 1.0	-1.1 ± 1.2	-1.8 ± 1.1	24.4 ± 1.2	27.0 ± 0.9	1.7 ± 1.5	3.5 ± 1.7	15.2 ± 0.9	17.2 ± 0.8	3.8 ± 4.4	9.3 ± 4.6
RR.FF.IN/TH.34	49.2 ± 0.9	47.9 ± 1.0	0.9 ± 1.2	0.5 ± 1.1	24.1 ± 1.2	26.5 ± 0.9	0.8 ± 1.5	1.6 ± 1.8	15.4 ± 0.9	16.5 ± 0.8	-0.9 ± 4.4	5.1 ± 4.6
RR.FF.IN/TH.35	46.2 ± 0.9	45.2 ± 1.0	-1.0 ± 1.2	-4.8 ± 1.1	25.7 ± 1.2	28.3 ± 0.9	3.1 ± 1.5	7.0 ± 1.7	15.3 ± 0.9	15.6 ± 0.8	-3.4 ± 4.4	-1.2 ± 4.6
RR.FF.IN/TH.37	49.2 ± 0.9	47.8 ± 1.0	1.9 ± 1.2	1.3 ± 1.1	24.2 ± 1.2	26.7 ± 0.9	-0.8 ± 1.5	-0.3 ± 1.7	15.4 ± 0.9	16.0 ± 0.8	-0.7 ± 4.4	4.3 ± 4.6
RR.FF.IN/TH.38	46.5 ± 0.9	47.1 ± 1.0	1.2 ± 1.2	-1.6 ± 1.1	25.6 ± 1.2	26.9 ± 0.9	-0.3 ± 1.5	4.1 ± 1.7	18.0 ± 0.9	15.9 ± 0.8	-7.4 ± 4.4	1.6 ± 4.6
A07-26NR	49.2 ± 0.8	47.8 ± 1.0			23.7 ± 1.1	26.2 ± 0.8			14.5 ± 0.7	15.5 ± 0.6		

[1] RR.FF.IN/TH = F_2-derived inbred line (IN) of Rutabaga-BF × A07-26NR and its test hybrid (TH) of A07-26NR × IN. For example, RR.FF.IN/TH.13 shows data of the inbred line RR.FF.IN.13 (see Table A3.17 for details) and its test hybrid.

Table A3.16: List of the inbred lines derived from the Rutabaga-BF × Hi-Q cross and their test hybrids (produced by crossing Hi-Q to the inbred lines).

Row	BC_1-derived inbred lines/test hybrids				Row	F_2-derived inbred lines/test hybrids			
	Inbred line		Test hybrid			Inbred line		Test hybrid	
	Code	S/N*	Code	S/N*		Code	S/N*	Code	S/N*
1	CO.BC.IN.03	1CA1866.231-A1096	CO.BC.TH.03	TC1866.231	1	CO.FF.IN.13	1CA1866.189-A1096	CO.FF.TH.13	TC1866.189
2	CO.BC.IN.04	1CA1866.235-A1096	CO.BC.TH.04	TC1866.235	2	CO.FF.IN.15	1CA1866.196-A1096	CO.FF.TH.15	TC1866.196
3	CO.BC.IN.06	1CA1866.238-A1096	CO.BC.TH.06	TC1866.238	3	CO.FF.IN.16	1CA1204.235-A1096	CO.FF.TH.16	TC1204.235
4	CO.BC.IN.07	1CA1866.232-A1096	CO.BC.TH.07	TC1866.232	4	CO.FF.IN.17	1CA1866.203-A1096	CO.FF.TH.17	TC1866.203
5	CO.BC.IN.08	1CA1866.234-A1096	CO.BC.TH.08	TC1866.234	5	CO.FF.IN.18	1CA1866.206-A1096	CO.FF.TH.18	TC1866.206
6	CO.BC.IN.09	1CA1866.239-A1096	CO.BC.TH.09	TC1866.239	6	CO.FF.IN.19	1CA1204.236-A1096	CO.FF.TH.19	TC1204.236
7	CO.BC.IN.16	1CA1866.233-A1096	CO.BC.TH.16	TC1866.233	7	CO.FF.IN.20	1CA1866.188-A1096	CO.FF.TH.20	TC1866.188
8	CO.BC.IN.18	1CA1866.211-A1096	CO.BC.TH.18	TC1866.211	8	CO.FF.IN.21	1CA1866.195-A1096	CO.FF.TH.21	TC1866.195
9	CO.BC.IN.19	1CA1866.215-A1096	CO.BC.TH.19	TC1866.215	9	CO.FF.IN.22	1CA1866.197-A1096	CO.FF.TH.22	TC1866.197
10	CO.BC.IN.20	1CA1866.217-A1096	CO.BC.TH.20	TC1866.217	10	CO.FF.IN.23	1CA1866.199-A1096	CO.FF.TH.23	TC1866.199
11	CO.BC.IN.21	1CA1866.218-A1096	CO.BC.TH.21	TC1866.218	11	CO.FF.IN.24	1CA1866.200-A1096	CO.FF.TH.24	TC1866.200
12	CO.BC.IN.22	1CA1866.219-A1096	CO.BC.TH.22	TC1866.219	12	CO.FF.IN.25	1CA1866.201-A1096	CO.FF.TH.25	TC1866.201
13	CO.BC.IN.23	1CA1866.222-A1096	CO.BC.TH.23	TC1866.222	13	CO.FF.IN.26	1CA1866.202-A1096	CO.FF.TH.26	TC1866.202
14	CO.BC.IN.24	1CA1866.223-A1096	CO.BC.TH.24	TC1866.223	14	CO.FF.IN.27	1CA1204.234-A1096	CO.FF.TH.27	TC1204.234
15	CO.BC.IN.25	1CA1866.224-A1096	CO.BC.TH.25	TC1866.224	15	CO.FF.IN.28	1CA1866.204-A1096	CO.FF.TH.28	TC1866.204
16	CO.BC.IN.26	1CA1866.225-A1096	CO.BC.TH.26	TC1866.225	16	CO.FF.IN.29	1CA1866.205-A1096	CO.FF.TH.29	TC1866.205
17	CO.BC.IN.27	1CA1866.226-A1096	CO.BC.TH.27	TC1866.226	17	CO.FF.IN.30	1CA1866.208-A1096	CO.FF.TH.30	TC1866.208
18	CO.BC.IN.28	1CA1866.229-A1096	CO.BC.TH.28	TC1866.229	18	CO.FF.IN.31	1CA1866.209-A1096	CO.FF.TH.31	TC1866.209
19	CO.BC.IN.29	1CA1866.236-A1096	CO.BC.TH.29	TC1866.236	19	CO.FF.IN.32	1CA1866.186-A1096	CO.FF.TH.32	TC1866.186
20	CO.BC.IN.31	1CA1866.227-A1096	CO.BC.TH.31	TC1866.227	20	CO.FF.IN.33	1CA1866.192-A1096	CO.FF.TH.33	TC1866.192
21	CO.BC.IN.32	1CA1866.228-A1096	CO.BC.TH.32	TC1866.228	21	CO.FF.IN.34	1CA1204.233-A1096	CO.FF.TH.34	TC1204.233
22	CO.BC.IN.33	1CA1866.216-A1096	CO.BC.TH.33	TC1866.216	22	CO.FF.IN.35	1CA1866.190-A1096	CO.FF.TH.35	TC1866.190
23	CO.BC.IN.34	1CA1866.221-A1096	CO.BC.TH.34	TC1866.221	23	CO.FF.IN.36	1CA1866.191-A1096	CO.FF.TH.36	TC1866.191
24	CO.BC.IN.36	1CA1866.220-A1096	CO.BC.TH.36	TC1866.220	24	CO.FF.IN.38	1CA1866.194-A1096	CO.FF.TH.38	TC1866.194
25	CO.BC.IN.37	1CA1866.230-A1096	CO.BC.TH.37	TC1866.230	25	CO.FF.IN.39	1CA1866.198-A1096	CO.FF.TH.39	TC1866.198
					26	CO.FF.IN.40	1CA1866.207-A1096	CO.FF.TH.40	TC1866.207

* Serial number.

Table A3.17: List of the inbred lines derived from the Rutabaga-BF × A07-26NR cross and their test hybrids (produced by crossing A07-26NR to the inbred lines).

Row	BC_1-derived inbred lines/test hybrids				Row	F_2-derived inbred lines/test hybrids			
	Inbred line		Test hybrid			Inbred line		Test hybrid	
	Code	S/N*	Code	S/N*		Code	S/N*	Code	S/N*
1	RR.BC.IN.01	1RA1869.217-A1096	RR.BC.TH.01	TC1869.217	1	RR.FF.IN.13	1RA1869.187-A1096	RR.FF.TH.13	TC1869.187
2	RR.BC.IN.03	1RA1869.210-A1096	RR.BC.TH.03	TC1869.210	2	RR.FF.IN.17	1RA1869.185-A1096	RR.FF.TH.17	TC1869.185
3	RR.BC.IN.05	1RA1869.223-A1096	RR.BC.TH.05	TC1869.223	3	RR.FF.IN.18	1RA1869.188-A1096	RR.FF.TH.18	TC1869.188
4	RR.BC.IN.14	1RA1869.209-A1096	RR.BC.TH.14	TC1869.209	4	RR.FF.IN.19	1RA1869.189-A1096	RR.FF.TH.19	TC1869.189
5	RR.BC.IN.15	1RA1869.214-A1096	RR.BC.TH.15	TC1869.214	5	RR.FF.IN.20	1RA1869.190-A1096	RR.FF.TH.20	TC1869.190
6	RR.BC.IN.17	1RA1869.219-A1096	RR.BC.TH.17	TC1869.219	6	RR.FF.IN.21	1RA1869.191-A1096	RR.FF.TH.21	TC1869.191
7	RR.BC.IN.18	1RA1869.221-A1096	RR.BC.TH.18	TC1869.221	7	RR.FF.IN.22	1RA1869.192-A1096	RR.FF.TH.22	TC1869.192
8	RR.BC.IN.19	1RA1198.105-A1096	RR.BC.TH.19	TC1198.105	8	RR.FF.IN.23	1RA1869.193-A1096	RR.FF.TH.23	TC1869.193
9	RR.BC.IN.21	1RA1869.220-A1096	RR.BC.TH.21	TC1869.220	9	RR.FF.IN.24	1RA1869.194-A1096	RR.FF.TH.24	TC1869.194
10	RR.BC.IN.22	1RA1869.224-A1096	RR.BC.TH.22	TC1869.224	10	RR.FF.IN.25	1RA1869.195-A1096	RR.FF.TH.25	TC1869.195
11	RR.BC.IN.23	1RA1869.225-A1096	RR.BC.TH.23	TC1869.225	11	RR.FF.IN.26	1RA1869.196-A1096	RR.FF.TH.26	TC1869.196
12	RR.BC.IN.24	1RA1869.227-A1096	RR.BC.TH.24	TC1869.227	12	RR.FF.IN.27	1RA1869.197-A1096	RR.FF.TH.27	TC1869.197
13	RR.BC.IN.25	1RA1869.228-A1096	RR.BC.TH.25	TC1869.228	13	RR.FF.IN.28	1RA1869.198-A1096	RR.FF.TH.28	TC1869.198
14	RR.BC.IN.26	1RA1869.230-A1096	RR.BC.TH.26	TC1869.230	14	RR.FF.IN.29	1RA1869.199-A1096	RR.FF.TH.29	TC1869.199
15	RR.BC.IN.27	1RA1198.103-A1096	RR.BC.TH.27	TC1198.103	15	RR.FF.IN.30	1RA1869.201-A1096	RR.FF.TH.30	TC1869.201
16	RR.BC.IN.28	1RA1869.211-A1096	RR.BC.TH.28	TC1869.211	16	RR.FF.IN.31	1RA1869.204-A1096	RR.FF.TH.31	TC1869.204
17	RR.BC.IN.29	1RA1869.212-A1096	RR.BC.TH.29	TC1869.212	17	RR.FF.IN.33	1RA1869.200-A1096	RR.FF.TH.33	TC1869.200
18	RR.BC.IN.30	1RA1869.229-A1096	RR.BC.TH.30	TC1869.229	18	RR.FF.IN.34	1RA1869.203-A1096	RR.FF.TH.34	TC1869.203
19	RR.BC.IN.31	1RA1198.104-A1096	RR.BC.TH.31	TC1198.104	19	RR.FF.IN.35	1RA1869.183-A1096	RR.FF.TH.35	TC1869.183
20	RR.BC.IN.32	1RA1869.222-A1096	RR.BC.TH.32	TC1869.222	20	RR.FF.IN.37	1RA1869.202-A1096	RR.FF.TH.37	TC1869.202
21	RR.BC.IN.33	1RA1869.226-A1096	RR.BC.TH.33	TC1869.226	21	RR.FF.IN.38	1RA1869.205-A1096	RR.FF.TH.38	TC1869.205

* Serial number.