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prepared by	Phillip Salisbury and Allison Gurung
co-authors/ contributors/ collaborators	Fu Tingdong, Wang Hanzhong, Wang Yanfei, Ma Chaozhi, Li Yunchang, Chen Yuehua, Lin Ping (China) Arvind Kumar, Surinder Banga, Dhiraj Singh, Abha Agnihotri, Jitendra Chauhan (India) Martin Barbetti, Wallace Cowling, Wayne Burton, Neil Wratten, Trent Potter, Sheng Chen, Mohammad Amjad (Australia)
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Contents

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AUSTRALIA

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South Australia Research and Development Institute

Department of Agriculture and Food WA

CHINA

Huazhong Agricultural University

Institute of Oil Crops Research, Chinese Academy of Agricultural Sciences

Institute of Economic Crops, Xinjiang Academy of Agricultural Sciences

INDIA

Indian Council of Agricultural Research

National Research Centre on Rapeseed-Mustard

Punjab Agricultural University

CCS Haryana Agricultural University

The Energy and Resources Institute

2 Executive summary

The overall aim of this trilateral ACIAR/GRDC project was to utilise germplasm from China, India and Australia to enhance productivity of canola quality *Brassica napus* and *B. juncea* in all three countries. The project was led by Associate Professor Phillip Salisbury from the University of Melbourne and involved 13 institutes across the three countries. The project was in line with ACIAR's objective of assisting developing countries in improving their own skills and resources, whilst also seeking to enhance *Brassica* oilseed production in Australia.

The key breeding priorities of each country were identified at the start of the project. In addition, the skills and resources of each institute were identified. From this information, the key traits for each institute to further develop as part of this project were selected. The key traits of interest for each country in the project included disease resistance, canola quality, drought tolerance, thermotolerance, shatter resistance and other agronomic traits. An additional key priority was to evaluate genetic diversity and heterosis in germplasm from the participating countries.

The project objectives were to:

- identify and/or develop effective screening/evaluation protocols for each key trait.
- identify appropriate variability for key traits through use of screening protocols.
- enhance germplasm in all countries for key traits through selection and breeding.
- identify genetic diversity and heterotic pools in the germplasm.
- develop/provide appropriate information on improved germplasm and diseases for incorporation into existing technology transfer protocols.
- increase the scientific skills of Chinese and Indian scientists through scientific exchanges, study tours and training.

To implement these objectives, germplasm exchange was undertaken among the three countries. In the first and third years of the project, each country contributed at least 30 lines of *B. napus* and/or *B. juncea* with variation for all key traits of interest. Field testing for each series of lines occurred for two years in each country.

The project outputs were as follows:

- appropriate and effective screening/evaluation protocols were identified for the key traits.
- appropriate variability was identified for the key traits.
- *Brassica* germplasm was enhanced in all countries though germplasm exchange, crossing and selection.
- genetic distance studies were undertaken and heterotic pools in the germplasm were identified.
- understanding of white rust and Sclerotinia diseases was improved and information packages on white rust and Sclerotinia were developed.
- scientific skills of Chinese and Indian scientists were enhanced through scientific exchanges and training.

The project provided clear benefits to all institutes, with the availability of enhanced germplasm expected to have major short term and long term impacts on oilseed *Brassica* productivity in participating countries.

3 Background

Oilseed *Brassicas* are the dominant oilseed crop in China and Australia and the second most important oilseed crop in India, in terms of area sown and production. The major oilseed *Brassica* species in Australia and China is *B. napus*, while in India *B. juncea* predominates, with only small areas of *B. napus* grown. The area of oilseed *Brassica* production has increased significantly in the last 15 years in each country. It has doubled in India and China and increased more than ten fold in Australia. Combined, the three countries represent more than half of the world's oilseed *Brassica* area (Table 1). However, despite these increases, oilseed production in China and India is insufficient to meet domestic consumption.

Table 1. Area and production in major oilseed *Brassicas* producing countries in the world in 2007 (FAOSTAT)

Country	Area 2007 ('000 ha)	Production 2007 ('000 tonnes)
China	7,050	10,375
India	6,790	7,438
Australia	1,061	1,065
Canada	6,277	9,528
Europe	8,114	20,400
Total	29,292	48,806

Despite the considerable progress in breeding programs in each country, a number of limitations to yield and quality still need to be addressed. The overall aim of this trilateral project was to utilise germplasm from China, India and Australia to enhance productivity of canola quality *B. napus* and *B. juncea* in all three countries. The project was in keeping with ACIAR's objective of assisting developing countries in improving their own skills and resources, whilst also working towards resolving Australia's own agricultural problems.

The top breeding priorities of each country were identified when the project was initiated. In addition, the skills and resources of each country were identified. From this information, the key traits for each country to further develop as part of this project were selected. Each country was responsible for exchanging Brassica germplasm expressing these key traits. The key traits of interest for each country in the project are summarised in Table 2 and the organisations involved in the project are listed in Table 3. It was expected that the *Brassica* germplasm would also be utilised as a source of other key agronomic traits, including high yield and oil content.

At the time of project initiation *B. napus* production was based predominantly on open-pollinated cultivars in Australia and China, with only a small area of hybrid canola grown. However, there was increasing interest in the utilisation of hybrids to enhance yield potential in each country. Thus, the project included the objective of examining genetic distance and heterosis among the exchanged germplasm.

Table 2. Key traits of interest

Trait	Species	Country
Canola quality ('00')	*B. juncea*	India, Australia, China
Drought tolerance and thermotolerance	*B. napus* *B. juncea*	Australia, India India, Australia
Sclerotinia	*B. napus* *B. juncea*	China, Australia India, Australia, China
White rust	*B. juncea*	Australia, (India)[a], (China)[a]
Shatter resistance	*B. napus*	Australia, India, (China)[a]

[a]*Indicates country not involved in trait development, but would still utilise the developed lines.*

Table 3. Names and acronyms of the organisations involved in the project

Country	Institute	Abbreviation	Key personnel
China	Huazhong Agricultural University	HZAU	Prof Fu Dr Ma Chaozhi
	Institute of Oil Crops Research, Chinese Academy of Agricultural Sciences	IOCR	Dr Wang Hanzhong Dr Li Yunchang
	Institute of Economic Crops, Xinjiang Academy of Agricultural Sciences	XAAS	Dr Wang Yanfei Dr Chen Yuehua Dr Lin Ping
India	National Research Centre on Rapeseed - Mustard	NRCRM	Dr Arvind Kumar Dr Jitendra Chauhan
	Punjab Agricultural University	PAU	Dr Surinder Banga Dr Shashi Banga
	CCS Haryana Agricultural University	HAU	Dr Dhiraj Singh
	The Energy and Resources Institute	TERI	Dr Abha Agnihotri
Australia	The University of Melbourne	UM	Dr Phillip Salisbury Dr Allison Gurung
	The University of Western Australia (Barbetti)	UWA-B	Dr Martin Barbetti
	The University of Western Australia (Cowling)	UWA-C	Dr Wallace Cowling Dr Sheng Chen
	Department of Primary Industries, Victoria	Vic DPI	Dr Wayne Burton
	Department of Industry and Investment, NSW	NSW DPI	Mr Neil Wratten (ret.) Mr John Sykes
	South Australia Research and Development Institute	SARDI	Mr Trent Potter
	Department of Agriculture and Food WA	Agric WA	Dr Mohammad Amjad Mr Graham Walton (ret.)

Canola quality B. juncea

Development of canola quality *B. juncea* is a high priority for Australia, India and China. The first canola quality *B. juncea* cultivars were developed in Canada and released in 2002. In Australia, canola quality *B. juncea* was developed to extend oilseed *Brassica* production into the lower rainfall areas. The first commercial *B. juncea* cultivar, cv. Dune, was released in Australia in 2006. In India at the time of project initiation, all commercial *B. juncea* cultivars were high in erucic acid and glucosinolates although lines with improved quality had been identified and low erucic acid *B. juncea* cultivars were in farmer field trials. In China, canola quality *B. juncea* hybrids were also a key target. Chinese *B. juncea* production is centred in Xinjiang province where the key *B. juncea* breeding program is based at the Institute of Industrial Crops, Urumqi.

Drought tolerance and thermotolerance

Production of *Brassica* oilseed crops in Australia and India is largely based on the rain-fed farming system, with a growing season corresponding to the rainfall patterns within a year. The growing season in Australia generally has a terminal drought onset during spring when pod-filling takes place rapidly. In India, *Brassica* oilseed crops are grown in the dry (post monsoon/winter) season, and are reliant on stored soil moisture for growth. Thus, identification of genotypes tolerant of drought and heat stresses at pod filling is a priority for Australia and India. Thermotolerance at the seedling stage is also a priority for India, as seed is sown in the post monsoon season, with average day time temperatures in the high twenties.

Utilisation of *B. carinata* (Ethiopian mustard) as a source of drought tolerance had already begun in India. Interspecific hybridisation had been utilised at Bharatpur to transfer drought tolerance and other key characters (e.g. disease resistance and shatter resistance) from *B. carinata* into *B. napus* and *B. juncea*. Thus, the aim was to incorporate drought tolerance identified in this Indian work into useful *B. napus* and *B. juncea* backgrounds for Australia, India and China. In addition, *B. juncea* and *B. napus* germplasm from India, China and Australia were screened in field and laboratory trials to identify sources of drought- and thermotolerance. Crossing and selection programs were used to combine the different sources of drought tolerance and thermotolerance into adapted Indian and Australian lines.

Sclerotinia resistance

Sclerotinia stem rot disease is a significant agricultural problem of many crops worldwide. Sclerotinia stem rot of oilseed rape (*B. napus*) and mustard (*B. juncea*) is a particularly serious threat to oilseed production in regions of Asia, Europe, and North America. At the time of project initiation it was also becoming an increasingly important disease in canola and mustard growing areas in Australia, especially in parts of New South Wales, Victoria and the northern coastal region in Western Australia. Yield loss as high as 24% had been recorded under Australian conditions. Location and deployment of varietal resistance remains the most cost-effective, economic and sustainable means of managing this disease. Thus, the germplasm was screened in each country to expose the lines to potential pathogen diversity in other countries to assess how they performed and to identify lines that were relatively more resistant.

At the time of project initiation, Australia did not have any recognised capacity to screen for resistance or tolerance to Sclerotinia. Several different methods were used in other countries to screen for the disease, however, there was often very poor correlation in the degree and nature of resistance expressed between different test methods, especially in relation to expected field performance. An objective of this project was to develop a Sclerotinia screening protocol suitable for implementation in Australia.

White rust

White rust (caused by *Albugo candida*) is a highly destructive disease of cruciferous vegetable and oilseed crops. Most commercial *B. juncea* varieties are highly susceptible to this pathogen. Combined infection of leaf and inflorescence causes yield losses up to 60% or more in India, where *B. juncea* is the predominant oilseed *Brassica* grown. As canola quality *B. juncea* is being developed to extend oilseed *Brassica* production into the lower rainfall areas in Australia it is essential to rapidly identify useful sources of white rust resistance. White rust resistance is also important for the Chinese *B. juncea* breeding program, based at the Institute of Industrial Crops, Urumqi.

Australia did not have any recognised capacity to screen for resistance or tolerance to white rust. Several different methods were used in other countries to screen for the disease (eg. glasshouse vs field tests), so development of a standard screening protocol for Australia was a priority. Physiological specialization is readily evident in *Albugo candida* and individual races are not confined to a single *Brassica* species but can cross-affect other cruciferous hosts. Assessment of pathogenic variability of white rust attacking isolates of *B. juncea* in Australia was therefore an additional aim.

Shatter resistance

Shattering of pods before harvest is a major problem of *B. napus* crops in Australia and India. While some improvement in resistance has occurred with selection in Australia, many *B. napus* crops are currently swathed to minimise shattering losses. In India (at TERI), *Raphanobrasscia* had been used as a bridging material to introduce shattering resistance into *B. napus* from *Raphanus* in crosses with *B. napus*. Following backcrossing to *B. napus* at TERI, fertile *B. napus* plants with enhanced resistance to shattering had been developed. This material was screened in the field at TERI along with the germplasm exchanged as part of the project to identify the best sources of shatter resistance. A crossing and selection program at TERI was undertaken to combine the different sources and develop breeding populations of *B. napus* with enhanced shatter resistance for use in India, China and Australia.

Shatter resistance was also sought from the *B. napus* x *B. carinata* crosses being developed by PAU. Incorporation of shatter resistance into commercial cultivars would reduce yield losses and remove the need for swathing, thus enhancing returns to growers.

Genetic Distance and heterosis

Increased utilisation of hybrids is considered to be an important way to increase yields. The project aimed to provide valuable data on economically important traits in crosses between Australian, Indian and Chinese *B. napus* lines, including information on F_1 hybrid vigour, additive genetic variance and molecular genetic distance between the parents.

The association between genetic distance and heterotic responses in *B. napus* was examined between parents originating from India, China and Australia. Key agronomic traits were evaluated including days to flowering, yield and seed weight. Genetic distance in *B. juncea* germplasm from the three countries was also assessed.

4 Objectives

1. **Identification and/or development of effective screening/evaluation protocols for each key trait.**
   - Milestone 1: Screening/evaluation techniques identified or developed for each key trait.

2. **Identification of appropriate variability for key traits through use of screening protocols.**
   - Milestone 2: Series I germplasm screened for all key traits.
   - Milestone 3: Series II germplasm screened for all key traits.

3. **Enhancement of germplasm in all countries for key traits through selection and breeding.**
   - Milestone 4: Backcross *B. napus* and *B. juncea* lines screened for key traits from *B. carinata*.
   - Milestone 5: Crossing programs for development of specific breeding populations initiated.
   - Milestone 6: Breeding populations for each specific character developed.

4. **Identification of genetic distance and heterotic pools.**
   - Milestone 7: First genetic distance data available.
   - Milestone 8: First F_1 hybrid vigour measurements available.
   - Milestone 9: Report relationship between genetic distance and heterosis.

5. **Development/provision of appropriate information on improved germplasm and diseases for incorporation into existing technology transfer protocols.**
   - Milestone 10: Information packages on disease resistance prepared for white rust and Sclerotinia in Australia.

6. **Increase the scientific skills of Chinese and Indian scientists through scientific exchanges, study tours and training.**
   - Milestone 11: Training programs for Chinese scientists completed.
   - Milestone 12: Training programs for Indian scientists completed.

5 Methodology

Germplasm exchange

Two hundred and ten lines of *B. napus* and *B. juncea* with variation for all key traits of interest were exchanged as part of the project. The first exchange (series I) was carried out in 2004/05 and the second exchange (series II) was carried out in 2006/07. A summary of the numbers of *B. napus* and *B. juncea* lines that were exchanged by the three countries is presented in Table 4 and the full list of lines including their characteristics are provided in Appendices 1 and 2.

Field screening of the series I and series II *B. napus* and *B. juncea* lines occurred for two years in each country. Table 5 outlines the timing of field trials in each country. Replicated plot trials were sown to measure yield, quality and agronomic characters, while specific disease nurseries and screening trials for drought tolerance, thermotolerance and shatter tolerance were also established. Further details of methods are provided in Section 7 (Key results and discussion).

Table 4. Series I and Series II germplasm exchange (further details in Appendices 1 and 2)

Country	Series I lines exchanged (2004/05)		Series II lines exchanged (2006/07)	
	B. napus	***B. juncea***	***B. napus***	***B. juncea***
China	20	10	25	20
India	3	22	2	23
Australia	25	12	31	17
TOTAL	48	44	58	60

Table 5. Project timeline

		2004		**2005**		**2006**		**2007**		**2008**		**2009**[a]	
Period:		Jan-Jun	Jul-Dec	Jan-Jun	Jul-Dec	Jan-Jun	Jul-Dec	Jan-Jun	Jul-Dec	Jan-Jun	Jul-Dec	Jan-Jun	Jul-Dec
Field trials:	Aus				Yr1		Yr 2		Yr3		Yr4		
	China					Yr1		Yr 2		Yr 3		Yr 4	
	India			Prelim		Yr1		Yr 2		Yr 3		Yr 4	
Meetings								China Prog Mtg		India Prog Mtg			Aus Final Mtg

[a]*Extended end date: 31 Dec 2009*

Yr 1, Yr 2 etc refer to the growing seasons in each country.

6 Achievements against activities and outputs/milestones

Objective 1: To identify and or develop effective screening/evaluation protocols for each key character

no.	activity	outputs/ milestones	completion date	comments
1.1	Determine if existing protocols are available and appropriate	Screening/ evaluation techniques identified for each key character	2006	Several existing screening protocols were utilised. These included protocols for quality, blackleg resistance, agronomic traits and drought tolerance.
1.2	Where required, development of new methods or comparisons of potential methods will be undertaken to ensure reliable new protocols for screening in required countries	Screening/ evaluation techniques developed for each key character	2007	Screening techniques for Sclerotinia and white rust resistance (UWA-B) and shatter resistance (TERI) were assessed.

Objective 2: To identify appropriate variability for key characters through use of screening protocols

no.	activity	outputs/ milestones	completion date	comments
2.1	Existing variability for key characters in series I *B. juncea* and *B. napus* lines identified	Series I lines screened for all key characters	2006	There was significant variation among lines for all of the traits in Australia, India and China.
2.2	Existing variability for key characters in series II *B. juncea* and *B. napus* lines identified	Series II lines screened for all key characters	2009	There was significant variation among lines for all of the traits in Australia, India and China.

Objective 3: To enhance germplasm in all countries for key characters through selection and breeding

no.	activity	outputs/ milestones	completion date	Comments
3.1	Interspecific hybridisation between *B. carinata* and *B. napus* and *B. juncea* initiated, backcrossing carried out and progeny checked for introgression of *B. carinata* genes	Backcross *B. napus* and *B. juncea* lines screened for key characters from *B. carinata*	2009	Crosses were carried out in India (PAU) between *B. napus* and *B. carinata*. *B. napus* type plants with high pollen fertility were probed for B-genome introgressions and those plants carrying B-genome introgressions were screened for white rust, shatter resistance, male sterility and other disease traits.
3.2	Initiate development of specific breeding populations by combining available sources of variation from country	Crossing programs for development of specific breeding populations initiated	2006, 2007, 2008	Crossing programs for the key traits were carried out using series I and series II lines.
3.3	Continue selection (and crossing) to enhance level of selected key characters in breeding (at least 1 generation/yr) population	Breeding populations for each specific character developed	2009 2010	Breeding populations (backcross, F_2 , F_3, F_4, & F_5) were developed for key traits in each country. The list of populations to be exchanged has been agreed upon and the exchange of material is currently underway.

Objective 4: To identify genetic distance and heterotic pools

no.	activity	outputs/ milestones	completion date	Comments
4.1	Seed of pure breeding lines produced and bulked	Seed of pure breeding lines sent to each country	2007	Seed of pure breeding lines of *B. napus* were developed by single seed descent or double haploidy at UWA-C.
4.2	Genetic distance studies of *B. napus*	First genetic distance data available	2007	Genetic distance data was generated at UWA-C, HZAU and PAU. Analysis of the data showed significant divergence among *B. napus* breeding lines.
4.3	Seed production of *B. napus* F1 populations	Preliminary diallel cross program Major diallel cross program	2006 2008	F_1 hybrids were generated from 13 lines and about 800 F_1 hybrid seeds per cross were produced and distributed in 2007 together with pure seed for the multi-location experiments in Australia, China and India. Approximately 1000 F_1 hybrid seeds per cross were produced at UWA for the hybrid trial in 2008. Seed was distributed in 2008 for the multi-location experiments.

4.4	F1 heterosis field study for *B. napus*	First F_1 hybrid vigour measurements available Second F_1 hybrid vigour measurements available Heterosis measured in F_1 hybrids and GCA/SCA measured in diallel crosses of pure lines	2008 2009 2008	Seven agronomic traits were measured (vegetative vigour, date of 50% flowering, height of first branch, height of first pod, mature height, seed yield and 1000-seed weight). Results were analysed across the 12 trials in 3 countries.
4.5	Analyse relationship between molecular genetic distance and heterosis	Report relationship between molecular genetic distance and heterosis	2009	F_1 heterosis data and molecular marker data were analysed and reported.
4.6	Heritability estimates for key characters	F_1 hybrid seed will be generated and sown at 2-3 sites in Australia, seed quality of individual plants will be measured and heritability will be estimated from the variance of parents, F_1 and F_2	2009	Broad heritability has been estimated.
4.7	Genetic distance studies of *B. juncea*	First genetic distance data available	2008	Genetic distance data for *B. juncea* lines was generated at UWA-C and PAU using the SSR technique. The results indicated that abundant genetic diversity exists in the *B. juncea* germplasm. Genetic distance data for an additional 32 lines has been completed by UWA-C and the analysis has been combined with the first study.

4.8	Additional Milestone: DArT genotyping of *B. napus* parent lines for hybrid trial and identifying heterosis-related alleles	Genotyping the 13 *B. napus* parent lines used for hybrid production in ACIAR project, using high throughput DArT markers. Allele-trait association analysis to identify the heterosis-related alleles	2009	800-1000 polymorphic DArT (diversity array technology) alleles have been generated. Additional research on identification of heterosis-related alleles is continuing beyond the project.

Objective 5: To develop/provide appropriate information on improved germplasm and diseases for incorporation into existing technology transfer protocols

no.	activity	outputs/ milestones	completion date	Comments
5.1	Information packages on disease resistance prepared and available for incorporation into integrated pest management of Sclerotinia and white rust	Information packages on disease resistance prepared for white rust and Sclerotinia in Australia	June 2009	The results obtained from the evaluation of assessment protocols and resistance screening, have been used to prepare information packages on white rust and Sclerotinia in Australia.

Objective 6: To increase the scientific skills of Chinese and Indian scientists through scientific exchanges, study tours and training

no.	activity	outputs/ milestones	completion date	Comments
6.1	Chinese scientists visit Australia for 3-6 months training in key areas (e.g. molecular biology, analytical chemistry)	Training programs for Chinese scientists completed	2007 2007 2009 (not completed)	Mr Wan Zhengijie, Huazhong Agricultural University (UWA-C, Oct 06- Mar 07) Dr Mei Desheng, Oil Crops Research Institute (CSIRO, Oct 06-Mar 07) Institute of Industrial Crops, XAAS The plan for a small group of XAAS breeders to participate in a study tour after the final meeting in Australia in Sep 2009 was not carried out due to political issues in Xinjiang province affecting their ability to get visas.

6.2	Indian scientists visit Australia for 3-6 weeks training in key areas (e.g. molecular biology, analytical chemistry)	Training programs for Indian scientists completed	2007 2005 2005 2004	Dr Chirantan Chattopadhyay, NRCRM, India (Sep 2007) Dr Maharaj Singh, NRCRM, India (DPI Vic, Sep 05-Dec 05) Short term training program (3 weeks): Dr Surinder Banga, PAU, India Dr Dhiraj Singh, HAU, India Dr Abha Agnihotri, TERI, India Short term training program (3 weeks): Dr NB Singh, ICAR, India Dr Arvind Kumar, Dr Chauhan, NRCRM, India Dr Surinder Banga, Dr Shashi Banga, PAU, India Dr Dhiraj Singh, HAU, India Dr Abha Agnihotri, TERI, India

7 Key results and discussion

7.1 Sclerotinia tolerance (Objectives 1, 2 & 3)

7.1.1 Aims

i. To develop/evaluate disease screening protocols for Sclerotinia in Australia.

ii. To screen B. napus and B. juncea germplasm in each country to identify new genotypes that have the required levels of host resistance essential for better management of this disease.

iii. To expose B. napus and B. juncea cultivars/breeding lines to potential pathogen diversity in other countries to assess how they perform.

7.1.2 Summary of results

Development of screening protocols (Objective 1)

Evaluation of screening techniques for Sclerotinia stem rot resistance was undertaken at UWA-B in controlled environment rooms using *B. napus* and *B. juncea* lines. The methods that were tested included stem agar inoculation, petiole inoculation, cotyledon inoculation, detached leaf inoculation and attached leaf inoculation. There was relatively little correlation between the results of these tests. However, experiments at UWA-B showed the usefulness of the stem agar inoculation method in differentiating responses to Sclerotinia under controlled environment conditions. It was found that using this method, stem lesion length 3 weeks after inoculation was significantly and positively correlated with the percentage of plant death at maturity ($r = 0.80$, $P < 0.001$, $n = 54$). Three weeks provided adequate time for symptom development on both susceptible and resistant genotypes, with stem lesion length in the experiment ranging from 3 to 21cm and plant death ranging from 23 to 97%. This method has previously been successfully used by Li et al. (2004) and Buchwaldt et al. (2005) for the effective identification of Sclerotinia resistance under field conditions.

A recent breakthrough for identifying resistance to Sclerotinia stem rot in Australia has been the development in Western Australia of a unique cotyledon assay that allows rapid differentiation of the reactions of *B. napus* genotypes against Sclerotinia stem rot (Garg et al. 2008). Experiments at UWA-B have also indicated a possible link between stem diameter and disease severity caused by Sclerotinia infection (Li et al. 2006).

In China, the national protocol for scoring Sclerotinia stem rot was used. Experiments were conducted in an artificial disease nursery with an overhead, water spraying system where inoculum levels were maintained by growing *B. napus* in consecutive years and placing two sclerotia in each row before sowing. Disease severity was scored on a 0 to 4 scale, based on % stem circumference with lesions (or % branches with lesions) and % pods affected. Disease pressure using this method in China was high enough to differentiate quantitative resistance among the lines tested. For example, plants with lesions in the Chinese resistant control Zhongyou 821 ranged from 35% to 68% between years, whereas, in the most susceptible lines 100% of plants had lesions.

Identification of variability (Objective 2)

Disease screening trials over the past 5 years by UWA-B, IOCR, NRCRM, HAU, UM, Agriculture and Agri-Food Canada (AAFC, Canada) and University of Georgia (USA) have indicated that there is substantial variability in terms of resistance/susceptibility of *B. napus* and *B. juncea* genotypes to Sclerotinia stem rot, although no sources of complete resistance within canola or mustard have yet been identified (Appendix 3).

Among the *B. napus* genotypes tested in field tests for stem resistance to Sclerotinia stem rot in Australia, ZY006 showed good resistance, with a mean stem lesion length of <0.5 cm in the 2007 screening. *B. napus* genotypes 06-6-3792, Fan168, ZY004 and Zhongyou-za No.8 from China and genotypes RT108, Oscar and RT057 from Australia also showed higher levels of resistance in Australia, with mean stem lesion lengths significantly less than many of the other *B. napus* lines.

In China, most of lines tested ranged from low to moderate susceptibility. None of the lines tested were significantly more resistant than the resistant control line, Zhongyou 821, based on % plants with lesions, however, some Chinese lines were as resistant as Zhongyou 821 (Appendix 3). Of the Australian *B. napus* lines, RT108 performed the best in the Chinese Sclerotinia nursery.

Table 6 summarises the best *B. napus* lines in varietal resistance rankings (top five) across experiments in the four countries, although not all lines were included in all screening experiments. Despite screening and assessment methods varying between countries, there were some lines that performed consistently well across countries and experiments. Differences in performance of lines between countries may indicate variation in the pathogenic variability of Sclerotinia isolates.

Table 6. Best performing *B. napus* lines for Sclerotinia resistance across experiments

Line	Source	Rating[a]	Number of Top 5 rankings
Zhongshu-ang No. 9 (30920)	China	Highly tolerant	4
RQ011	Aus		4
Fan168	China		3
Oscar	Aus		3
RR002	Aus		3
Zhongyou 821	China	Highly tolerant	2
P624	China	Highly tolerant	2
Zhongshu-ang No.4 (30872)	China	Tolerant	2
ZY002	China	Moderately tolerant	2
06-06-3792	China		2
Ag-Outback	Aus		2
Ag-Spectrum	Aus		2
Rivette	Aus		2
RR013	Aus		2
RT108	Aus		2
ZY006[b]	China	Highly tolerant	1

[a] *Rating assigned by IOCR pre-exchange of germplasm*

[b] *ZY006 was only included in one experiment because only a small amount of seed was available.*

B. juncea was generally more susceptible to Sclerotinia stem rot than *B. napus* in all experiments. In Australia, the most resistant *B. juncea* genotypes included Australian lines JM06018 and JM06006 and Chinese line B. juncea 2. The *B. juncea* lines with the

most tops five rankings across all experiments are shown in Table 7 (although not all lines were included in all screening experiments).

Table 7. Best performing *B. juncea* lines for Sclerotinia resistance across experiments

Line	Source	Rating	Number of Top 5 rankings
B. juncea 2	China		2
JM06006	Aus		2
JM06018	Aus		2
JM18	Aus		2
JN031	Aus		2
JN032	Aus		2
JN033	Aus		2
RH 13	China		2
Qianxianjiecai	China		2

Enhancement of germplasm through selection and breeding (Objective 3)

A crossing program was initiated at HZAU in 2005 with GMS (genic male sterile) lines, using pol cms restorers with strong tolerance to Sclerotinia. New restorers were added into the population in 2006. Continued selection and screening for Sclerotinia resistance was done in 2008 and 2009. Six breeding lines with Sclerotinia tolerance have been selected for exchange with project partners.

At DPI Vic 20-30 crosses between Chinese and Australian *B. napus* lines were made for Sclerotinia resistance. Five doubled haploid populations have been made from the *B. napus* crosses for Sclerotinia resistance and these are awaiting screening for resistance to Sclerotinia to study inheritance.

7.1.3 Key findings

- *B. napus* was generally more resistant to Sclerotinia stem rot than *B. juncea.*
- Genotypes ZY006, 06-6-3792, Fan168, ZY004 and Zhongyou-za No.8 from China and genotypes RT108, Oscar and RT057 from Australia showed good resistance to Sclerotinia stem rot in Australia relative to the other lines.
- *B. napus* lines were identified that were consistently more resistant across experiments, providing a positive sign for the screening technique and for confidence in selecting for resistance.
- Variation in pathogenic variability of Sclerotinia isolates between countries was suggested.

7.1.4 Recommendations

- Definition and monitoring of pathotypes of Sclerotinia will be essential for managing resistance in the future.
- Mechanisms of resistance(s) should be defined.
- Genetics of host resistance(s) in segregating populations should be characterised in the double haploid populations.

7.2 White rust (Objectives 1, 2 & 3)

7.2.1 Aims

i. To develop/evaluate disease screening protocols for white rust in Australia.

ii. To screen B. juncea germplasm in each country to identify new genotypes that have the required levels of host resistance essential for better management of this disease.

iii. To expose B. juncea cultivars/breeding lines to potential pathogen diversity in other countries to assess how they perform.

iv. To assess pathogenic variability of white rust attacking isolates of B. juncea from around Australia using a differential set to provide an understanding of the race structure.

7.2.2 Summary of results

Development of screening protocols (Objective 1)

Experiments were undertaken by UWA-B under controlled environmental and field conditions to evaluate the expression and relationships of resistance to white rust at the cotyledonary, seedling and flowering stages. Overall, disease severity on cotyledons and leaves at the different growth stages was significantly and positively correlated. However, there was no significant correlation between the number of stagheads (hypertrophied inflorescences filled with oospores) and any of the other disease parameters measured (Li et al. 2007). The study demonstrated that controlled environmental conditions are suitable for rapid identification of resistant genotypes and that genotypes with high levels of resistance can be reliably identified at either the cotyledonary, seedling, or flowering stages.

Physiological specialization is readily evident in *Albugo candida*, the casual agent of white rust. The situation regarding races of white rust affecting oilseed *Brassicas* is complicated by the fact that many individual races are not confined to a single *Brassica* or cruciferous species and can cross-infect other *Brassica* hosts. Race 2 (pathotype 2A and/or 2V) affects oilseed *B. juncea* in Canada. Pathotyping of *B. juncea*-infecting *A. candida* isolates collected from infected canola quality *B. juncea* from eight locations in eastern Australia was carried out using a set of host differentials. The results indicated that the most common isolate in Australia is pathotype 2A. However, recently the presence of pathotype 2V has been reported in Western Australia (Kaur et al. 2008). Thus, further regular screening of Australian isolates to confirm races and pathotypes is required.

Identification of variability (Objective 2)

B. juncea genotypes differed greatly in their levels of resistance/susceptibility to white rust. In India and China, most of the Australian lines were a very good source of white rust resistance relative to local lines which suggests the presence of variation in the pathogen between the countries (Table 8). In Australia, a few genotypes were also identified that showed very good resistance to the most commonly occurring isolates (Table 8). Further work is required to locate resistance to other pathotypes of the pathogen.

Table 8. White rust resistance in *B. juncea*

Country	Institute	Material	Most resistant lines
Australia	UWA-B	Australian, Chinese, Indian	Series I: CBJ-001, CBJ-002, CBJ-003, CBJ-004, JR049 Series II: JM06011, JM06010, JM06021, JM06004, JM06013
India	PAU	Australian, Indian	All Australian lines RH 819 (Indian line)
	TERI	Australian, Indian	All Australian lines
China	XAAS	Australian, Indian	Majority of Australian lines (only a few Australian lines developed light to moderate infection later in growing period)

Enhancement of germplasm through selection and breeding (Objective 3)

The techniques used in the project for white rust screening have subsequently been used to annually screen lines from the Australian canola quality *B. juncea* breeding program to assist with selection and breeding for white rust resistance.

In 2007, Australian lines with canola quality and white rust resistance were used in crosses with the Chinese and Indian lines and F_2 seed was produced in the glasshouse (123 crosses). In 2008, approximately 800 single plant selections were taken from 40 of the 123 crosses. Quality data from these selections indicates that the F_3's will need another round of selection and crossing to be directly usable.

7.2.3 Key findings

- Australian *B. juncea* lines are a very good source of white rust resistance for China and India.
- Breeding populations have been produced incorporating canola quality and white rust resistance into Chinese and Indian backgrounds, which will benefit the Chinese and Indian *B. juncea* breeders. These lines are being exchanged as part of the project.

7.2.4 Recommendations

- The opportunity exists to further define races and pathotypes of *A. candida* occurring in Australia. In particular, a national standard host differential set (for cotyledon and staghead infection) should be developed.
- White rust screening of the breeding populations developed during the project, particularly those incorporating genes from *B. carinata*, should be undertaken.
- The relationship between staghead formation and vegetative stage resistance requires further investigation.

7.3 Thermotolerance (Objectives 1, 2 & 3)

i. To identify variation for thermotolerance in germplasm.

ii. To enhance B. napus and B. juncea germplasm through crossing and selection for thermotolerance.

7.3.1 Summary of results

Development of screening protocols (Objective 1)

Seedling stage (HAU)

A series of experiments were carried out at HAU to test protocols for identifying thermotolerant cultivars. This included identification of the optimum temperature for seed germination and examination of the effect of high temperature on seedling mortality. The optimum temperature for shoot and root growth was 25ºC. Examination of the effect of high temperature on seedling mortality indicated that the threshold high temperature in mustard genotypes is 47.5ºC at which screening of genotypes can be done on a large scale. Thus the following screening protocol was used to screen for thermotolerance at HAU: seedlings were grown at optimum temperature (25ºC), five days after sowing the seedlings were exposed to the threshold high temperature (47.5ºC) in the seed germinator. Assessment was based on the time needed for a line to reach 50% mortality, with the longer the time, the more tolerant to high temperature. Based upon this result, the lines were grouped into thermotolerant, moderately thermotolerant, and thermosusceptible genotypes.

Seedling and terminal stage (PAU)

Seedling thermotolerance was assessed in the laboratory by exposing 21 days old seedlings to 45ºC for 5 hrs and measuring percentage survival. Seedling thermotolerance was also evaluated in the field based on seedling survival under early sowing. Terminal stage thermotolerance was measured in the field by the decrease in number of pods on the main shoot, pod length and seed size at normal sowing time vs late sowing. This was done to ensure that the reproductive phase in the late sowing experienced high terminal stage temperature stress.

Seedling and terminal stage (NRCRM)

Brassica juncea was evaluated for high temperature tolerance at seedling emergence and seed development stage by comparing the performance of key traits at early sowing time vs late sowing time (that exposed the germination/emergence and seed development stages to high temperature (~40ºC) in the field). Two hundred seeds/genotype were sown in 2 row plots of 5 m length. Two irrigations were given at 35-40 and 75-80 DAS. Measurements included % emergence, height, primary branches, siliqua length, seeds/siliqua, 1000-seed weight, seed oil and protein content.

Seedling thermotolerance was also assessed in the laboratory by measuring germination, mortality and amylase activity at 45±1ºC. The treatment involved keeping seeds at 45±1ºC for 4 hours each day then returning them to optimum temperature (25ºC) for 7 days. Percentage germination and mortality were recorded. The experiment was carried out in Petri dishes and also in soil.

Identification of variability (Objective 2)

Variation in thermotolerance was observed among the lines, although the method of assessing tolerance had a substantial influence on the lines identified as most tolerant (Appendices 4 and 5). In addition, seedling stage tolerance did not appear to correlate with terminal stage tolerance. *B. juncea* lines were generally more tolerant at the seedling stage than *B. napus*.

In the NRCRM trials, Indian *B. juncea* lines stood out as showing seedling thermotolerance using different screening methods. The best performing lines included Varuna, Prakash, CS52 and RH819 (Table 9; Appendix 4).

Indian *B. juncea* cultivar Kranti appeared to possess relatively better terminal heat stress tolerance (Table 10). In the PAU trial, Kranti showed a low reduction in pods on the main shoot as well as a low reduction in seed size (Appendix 4). A few of the Australian *B. juncea* lines also performed well for thermotolerance in the Indian trials (Table 10).

None of the genotypes showed tolerance to high temperature in terms of all the traits measured.

Table 9. Best performing lines for seedling stage thermotolerance

Line	Series	Source	Number of times line recorded as thermotolerant at seedling stage (relative to other lines in Indian trials)
Varuna	I	India	5
Prakash	I	India	5
CS52	II	India	3
RH819	I	India	3
Kranti	I	India	2
Loiret	I	India	2
PCR 7	I	India	2
Vaibhav	I	India	2
JN032	I	Australia	2
JR042	I	Australia	2
Aravali	II	India	2
Basanti	II	India	2
Jagannath	II	India	2
RGN 13	II	India	2
Urvashi	II	India	2

Table 10. Best performing lines for terminal stage thermotolerance

Line	Series	Source	Number of times line recorded as thermotolerant at terminal stage (relative to other lines in Indian trials)
Kranti	I	India	3
JM06006	II	Australia	3
Bio902	I	India	2
JM2	II	India	2
JM06015	II	Australia	2
JM06018	II	Australia	2

Enhancement of germplasm through selection and breeding (Objective 3)

The crossing program for development of drought tolerant and thermotolerant breeding populations of *B. juncea* was undertaken by HAU and NRCRM. In 2006, *B. juncea* populations for drought tolerance were produced from 10 crosses between series I drought tolerant lines and high yielding lines at NRCRM. In 2007, 27 F_1 and 47 F_2 populations from crosses between Chinese, Australian and Indian series I *B. juncea* lines were grown and selections were made in the F_2. In addition, 60 F_1 crosses were attempted between Australian and Indian series II *B. juncea* lines.

7.3.2 Key findings

- Substantial variation was evident for all components of seedling and terminal thermotolerance.
- Sources of high temperature tolerance identified are being utilized in the breeding programs.

7.3.3 Recommendations

- The different screening methodologies need to be compared to define better indicators for thermotolerance.
- The methodologies for assessing thermotolerance need to be standardised (e.g. the screening temperature and time exposed to extreme temperature for the seedling stage testing).
- The response of physiological and biochemical parameters need to be compared to yield response to determine the most efficient parameter for screening large populations effectively.

7.4 Drought tolerance (Objectives 1, 2 & 3)

7.4.1 Aims

i. To identify variation for drought tolerance in germplasm.

ii. To enhance B. napus and B. juncea germplasm through crossing and selection for drought tolerance.

7.4.2 Summary of results

Development of screening protocols (Objective 1)

HAU

The effect of drought conditions on root characteristics, plant water relations and yield was examined at HAU. The results indicated that *B. napus* and *B. juncea* adopted different mechanisms in response to water stress. In *B. juncea*, water deficit decreased leaf water potential (WP) and leaf relative water content (RWC) resulting into greater osmotic adjustment and higher root growth. This helped the plants to explore greater soil volume for water resulting in better yield attributes and ultimately seed yield. In *B. napus*, however, RWC followed osmotic potential, which indicated that *B. juncea* had greater osmotic adjustment than *B. napus*. The argument is further supported by the fact that decreases in WP, RWC and osmotic potential (OP) promoted root growth in *B. juncea* but not in *B. napus*.

Experiments at HAU indicated that measurements of plant water status at midday and root zone depth at siliqua formation could be used for screening relatively large numbers of germplasm lines for drought tolerance.

PAU

PAU conducted field trials to identify drought tolerant lines. The PAU experiment consisted of three treatments, no irrigation, one irrigation and a recommended control of two irrigations. Irrigation treatments and varieties were included as main and sub plots. Data were collected for traits including height, seed weight, yield, leaf characteristics, chlorophyll fluorescence, root characteristics and seeds/siliqua. Lines showing the least reduction in the expression of a trait following restricted irrigation were considered to be more drought tolerant.

NRCRM

Field trials were conducted under rain-fed conditions and measurements taken included specific leaf area, transpiration rate and water potential at 65-75 days after sowing.

Identification of variability (Objective 2)

Field evaluation for drought tolerance was carried out in India. The *B. juncea* and *B. napus* varieties displayed variation for all key traits (Appendices 6 and 7). Identification of the best performing lines was strongly influenced by assessment method and stage of plant development (Appendices 6 and 7). Generally, the Indian lines performed better for all the traits that were measured to test drought tolerance in India. *Brassica juncea* lines that regularly performed well across years, testing method and/or sites included PCR7, Varuna, Kranti Sej2, NDR8501, RH819 and JM018.

In addition to the series I and II *B. napus* lines, an extra set of 24 Australian *B. napus* lines were sent to India in 2007 to be assessed for tolerance to terminal drought stress at PAU (Appendix 8). In this trial, variation due to drought treatment (irrigation) as well as genotypes was significant and genotype x environment interactions were also significant. There was a greater difference between one irrigation and no irrigation than between one irrigation and two irrigations. Overall Australian *B. napus* line RT117 appeared to be the most drought tolerant of the lines as it showed the least reduction in leaf area and leaf ratio and it also performed well for seed size, seed yield and root area (Figure 1; Appendix 8). Based on chlorophyll fluorescence analysis, Monty, Tarcoola and Sapphire showed lower drought stress, while for seeds/siliquae, GSC6 and ATR-Summit showed the least reduction.

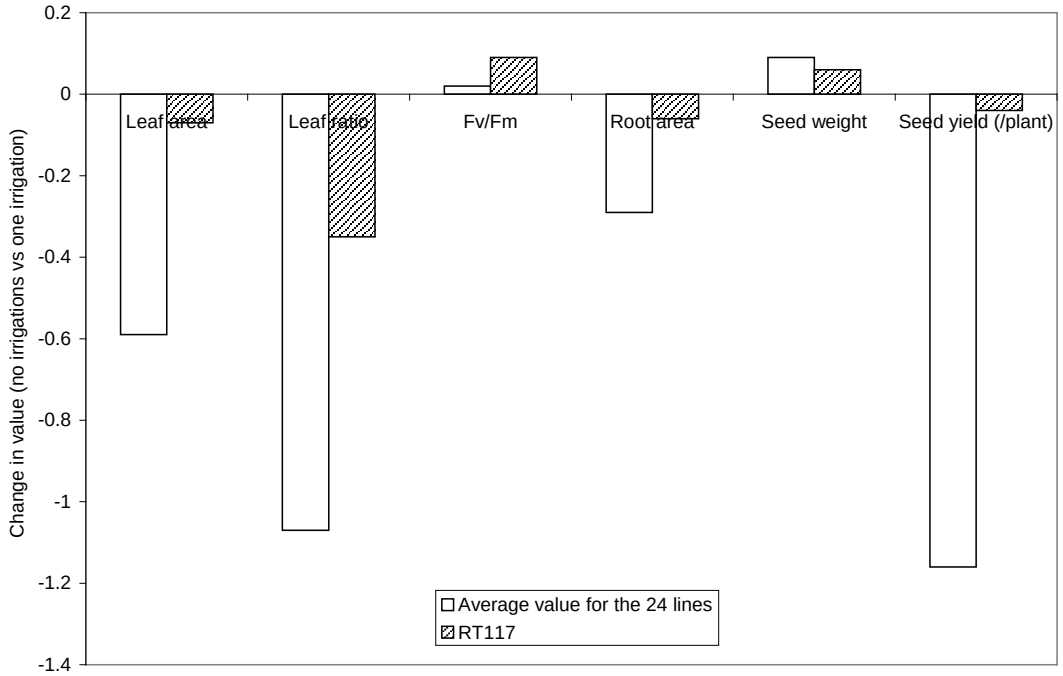

Fig. 1. Terminal drought tolerance screening of Australian *B. napus* lines in India (most tolerant line and average of all lines are presented)

Enhancement of germplasm through selection and breeding (Objective 3)

At HAU crosses were made between *B. juncea* from Australia, China and India for producing breeding populations for drought tolerance.

At NRCRM, the crossing program for development of drought- and thermotolerant breeding populations of canola quality *B. juncea* was undertaken. In 2006, *B. juncea* populations for drought tolerance were produced from 10 crosses between series I drought tolerant lines and high yielding lines. In 2007, 27 F_1 and 47 F_2 populations from crosses between Chinese, Australian and Indian series I *B. juncea* lines were grown and selections were made in the F_2. In addition, 60 F_1 crosses were carried out between Australian and Indian series II *B. juncea* lines.

7.4.3 Key findings

- *Brassica juncea* lines that regularly performed well in drought tolerance tests across years, testing methods and/or sites were generally the Indian lines. In particular, PCR7, Varuna, Kranti Sej2, NDR8501, RH819 (Indian) and JM018 (Australian) were relatively more drought tolerant.
- Of the extra Australian *B. napus* lines, RT117 appeared to be relatively more drought tolerant, as it showed the least reduction in leaf area and leaf ratio and it also performed well for seed size, seed yield and root area.

7.4.4 Recommendations

- The different screening methodologies need to be compared to define better indicators for drought tolerance.
- The methodologies for assessing drought tolerance need to be standardised.
- The response of physiological and biochemical parameters need to be compared to yield response to determine the most efficient parameter(s) for screening large populations effectively.

7.5 Canola quality *Brassica juncea* (Objectives 1, 2 & 3)

7.5.1 Aims

i. To identify new sources of variation for low erucic acid and low glucosinolate content in germplasm.

ii. To enhance B. juncea germplasm through crossing and selection for canola quality.

7.5.2 Summary of results

Development of screening protocols (Objective 1)

Existing standard wet chemistry and near infrared (NIR) protocols were used. In Australia, all samples were analysed at Wagga Wagga Agricultural Institute, NSW Department of Primary Industries by the AOCS approved Oil Testing Service. Seed quality results between and within institutes and countries did not always correlate well, so an additional goal was set up with the assistance of Dr Rod Mailer (NSW DPI) to assist with standardisation of oil quality testing at each institute. Seed from a single batch of seed of 6 *B. napus* and 6 *B. juncea* lines that covered the variation in oil quantity and glucosinolate content were analysed in replicate by Dr Mailer then these standard samples have been made available to Indian and Chinese collaborators for analysis and comparison of results.

Identification of variability (Objective 2)

The glucosinolate results revealed the presence of low glucosinolate lines in both the series I Chinese and Australian *B. juncea* germplasm (Table 11). The Indian series I and II varieties all had very high glucosinolate contents (Table 11). The series II Chinese lines also had very high glucosinolate content relative to the Australian lines.

Table 11. Glucosinolate content of *B. juncea* lines averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
Aus	I	23-101 (ave 51)	Xinyou8	19-71 (ave 41)	JR042	68-94 (ave 87)	-
China*	I	-	Xinyou9, CBJ002	-	JM16, JR042, JR049	-	-
India	I	26-94 (ave 58)	CBJ004, CBJ001, CBJ002	13-72 (ave 32)	JR042, JR049	75-146 (ave 111)	-
Aus	II	83-105 (ave 94)	-	29-46 (ave 35)	JM06004, JM06009, JM06002	89-103 (ave 96)	-
China*	II	-	-	-	JM06002, JM06004, JM06009	-	-
India	II	30-124 (ave 74)	-	4-42 (ave 17)	JM06011	-	-

**XAAS, Urumqi*

Enhancement of germplasm through selection and breeding (Objective 3)

In 2007, crosses were made between Australian x Chinese and Australian x Indian *B. juncea* series I lines (123 crosses) and F_2 seed was produced in the glasshouse at DPI Vic. The Australian lines that were used in the crossing had canola quality (as well as white rust resistance).

10-20 *B. juncea* F_4 lines from Indian background that appear to have reasonable quality were evaluated in 2008 trials and backcrossing to Indian material (BC_2 derived lines) was continued.

7.5.3 Key findings

- New canola quality *B. napus* and *B. juncea* lines from Australia will be widely used in Chinese and Indian breeding programs.

7.5.4 Recommendations

- Participation of institutes in the analysis of standard samples provided by Dr Rod Mailer (NSW DPI) is recommended to assist with standardisation of oil testing protocols in each laboratory.

7.6 Shatter resistance and *B. carinata* breeding (Objectives 1, 2 & 3)

7.6.1 Aims

i. To assess shatter resistance screening techniques.

ii. To identify new sources of variation for shatter resistance.

iii. To enhance B. napus and B. juncea germplasm for shatter resistance through incorporation of new sources of interspecific variation.

7.6.2 Summary of results

Development of screening protocols (Objective 1)

Experiments were undertaken at TERI to compare visual shatter observations with percentage shattering on the main stem. About 50% of plants of each line grown during 2006/2007 in India were left standing in the field 4 weeks post maturity. Visual shatter observations were taken using a 1 to 9 scale (1 – high shattering and 9 – low shattering) and the % shattering was also calculated [(no. of pods shattered/total no. of pods on main shoot) x 100]. Data from both methods were compared and found to have good correlation (*B. juncea* r^2=0.94; *B. napus* r^2=0.90), indicating that with an experienced assessor, visual observations are sufficient.

Identification of variability (Objective 2)

At TERI, the series I lines were evaluated for shatter resistance by two methods, visual observations and % shattering. Pod shattering was generally low in all lines during 2006/2007 screening at TERI due to environmental conditions. However, in 2007/2008 significant variability was observed between the lines as the environmental conditions were very suitable for screening, including heavy rains. At 7 to 8 weeks post maturity the % shattering among the lines showed significant variation. Of the 58 *B. juncea* and 82 *B. napus* (Series I & II) lines, only 11 and 8 respectively showed <20% shattering. The remaining accessions had high shattering ranging 21 to 60%. The accessions ZY005 and ZY014 had > 90% shattering. Even the series I accessions that showed good tolerance to shatter 4 to 5 weeks post maturity during the previous years (when conditions were less unfavourable), showed significantly high shattering 8 weeks post maturity following the heavy rains in 2007-08. The best performing lines are listed in Table 12.

Table 12. Range of shattering observed in Series I and II Australian and Chinese *B. napus* and *B. juncea* lines at TERI (2007/08)

Species	Range	Best lines
B. napus (Australian and Chinese)	16-95%	Ag-Outback, Tranby, RR009, RR001, Skipton, TQ00502W2, BLN3243 (<20% shattering)
B. juncea (Australian and Chinese)	6-65%	RK2, RL, Haoyou11, Tunliuhuangjie (<10% shattering) CBJ001, JMO6026, RH13, Ringot I, AmoraII, Datonghuangyoucai, Qianxianjiecai (<20% shattering)

Shatter resistance was also visually assessed at PAU in the field at harvest using the 1 to 9 scale (Table 13). In the PAU trial the conditions were very favourable for shattering and the *B. napus* lines ranged from scores of 1 to 4. The line TERI(00)R9903 with reported

resistance to shattering, performed relatively well with a score of 3. This line has been exchanged as part of the project.

Table 13. Range of shattering observed in Series II Australian, Indian and Chinese *B. napus* and *B. juncea* lines at PAU

Species	Range[a]	Best lines
B. juncea (Australian, Chinese, Indian)	2-8	JM06020, Haoyou11, RK 9501, Pusa Mahak
B. napus (Australian, Chinese, Indian)	1-4	RT057, 06-P71-1, ZY008, ZY010

[a] *1 = high shattering, 9 = low shattering*

In 2007 and 2008, experiments were carried out in Australia (by DPI Vic) to compare shattering data from India and Australia to confirm whether genotypes are performing similarly under different growing environments. However, the prevailing environmental conditions in Australia led to very limited shattering in 2007 and the trial failed in 2008 due to drought. This trial was successfully completed in 2009/10.

Enhancement of germplasm through selection and breeding (Objective 3)

TERI

At TERI crosses were made between six Australian *B. napus* genotypes (Surpass, Trilogy Trigold, Tranby, Monty and RR005) and three Indian low erucic/canola quality/shattering tolerant *B. napus* genotypes (TERI(00)R9903, TERI(00)GS17 and TERI(00)EM05). The progeny were grown in single rows during 2006/07, along with the parents. These were evaluated, selfed, and also backcrossed to the Australian parents for developing breeding populations for shatter tolerance. The plants were also evaluated for agronomic and biochemical traits. Most of the F_1/F_2 populations recorded shattering of 15 to 30%, with the exception of crosses with Monty that recorded shattering up to 72% (Appendix 9).

The F_2/F_3 and BC_1/BC_2 populations from these crosses were grown in single plant progeny rows during 2007/08, along with the parents, for evaluation, generation advance, and backcrossing to the Australian parents to develop breeding populations. There was significant variation in percentage shattering at 7 to 8 weeks post maturity. F_2, F_3, BC_1 and BC_2 populations showed between 21 and 67% shattering under conditions favourable for shattering (Appendix 10). The following season (2008/09) the BC_2/BC_3 seeds were sown in two replications for generation advance and evaluation (Appendix 11). Shattering ranged from 7 to 31% at four weeks post maturity (Appendix 11). Breeding populations that will be exchanged as part of the project are shown in Table 14.

Table 14. Shatter evaluation of the four *B. napus* BC_2 and BC_3 breeding populations developed at TERI for exchange as part of the project (2008/09)

Line[a]	Days to 50% flowering	Days to maturity	Shattering % 4 weeks post maturity
R9903 x RR005 (BC_2)	50	139	7
Surpass400 x R9903 (BC_3)	49	142	8
Tranby x R9903 (BC_3)	64	143	14
Trilogy x R9903 (BC_2)	77	142	24
Average of all 26 breeding populations in the 2008/09 trial	57	140	17

[a] *R9903 = TERI(00)R9903*

PAU (B. carinata)

Crosses were undertaken at PAU to introgress B genome traits from *B. carinata* into *B. napus* backgrounds. Key Australian germplasm lines were used as *B. napus* recipients. Following the first backcross of the original cross combinations, five generations of selfing and selection were carried out. Further backcrossing was not carried out to avoid loss of B genome genetic information. Selections were carried out in every generation for the desired traits (Table 15). At the end of five generations of selfing, DNA from 165 selected plants was harvested. The DNA was probed using a B genome specific molecular marker for the presence of B chromatin. The sequence information of the B genome specific primer (pBNBH35 R) is as below:

5'-GGC ATC TGA AGA GAG AGT CCC TTT G-3'

5'-ATC TTC TTC TTG CCA TGA GTG GCC-3'

Table 15. *B. carinata* trait incorporation at PAU

Combination	Selective traits
B. napus x *B. carinata*	White rust resistance, shatter resistance and other disease resistance traits
B. carinata x *B. napus*	Male sterility and other disease resistance traits

Seventy three of the individually selected plants were analysed cytologically. None of them showed a chromosome number that was euploid for *B. napus*. Plants with least deviation in the euploid chromosome number are listed in Table 16. The lines proposed for exchange under the project are those highlighted in bold type. The plants will now be subjected to multicolour genomic *in situ* hybridization (GISH) to find out their genomic constitution. It is expected that there will be a deficiency of B genome chromosomes as well as some substitutions involving C genome chromosomes.

Table 16. Australian lines with introgressed *B. carinata* DNA[a]

Type	Genotype(s)	Chromosome No.
B. napus/B. carinata hybrids	Surpass400 NCB4	35
	Rivette NCB5	35
	Rainbow NCD5	36
	RQ011 NCB6	37
	RR001 NCA3	39
	Charlton NCA4	39
	Rivette NCB5	**39**
	Rainbow NCA4	39
	RR009 NCA2	40
	RR009 NCA2	40
	RR009 NCA2	40
	Surpass400 NCB4	**40**
	RQ011 NCB5	40
	Rainbow NCD5	40
	Rivette NCB4	40
	RR009 NCA2	41
	Surpass400 NCB5	41
	Charlton NCA4	42
	Spectrum NCB5	42

B. juncea/B. carinata hybrids	**JC134**	**36**
	JC67	**36**

[a]*Lines exchanged in the project are those highlighted in bold type*

Selection for shatter resistance was carried out on the introgression lines and some lines with putative shatter resistance were identified. Histological studies indicated some promise in Surpass400 NCB4 (Figure 2). The chromosome number, however, is still higher than the expected euploid number.

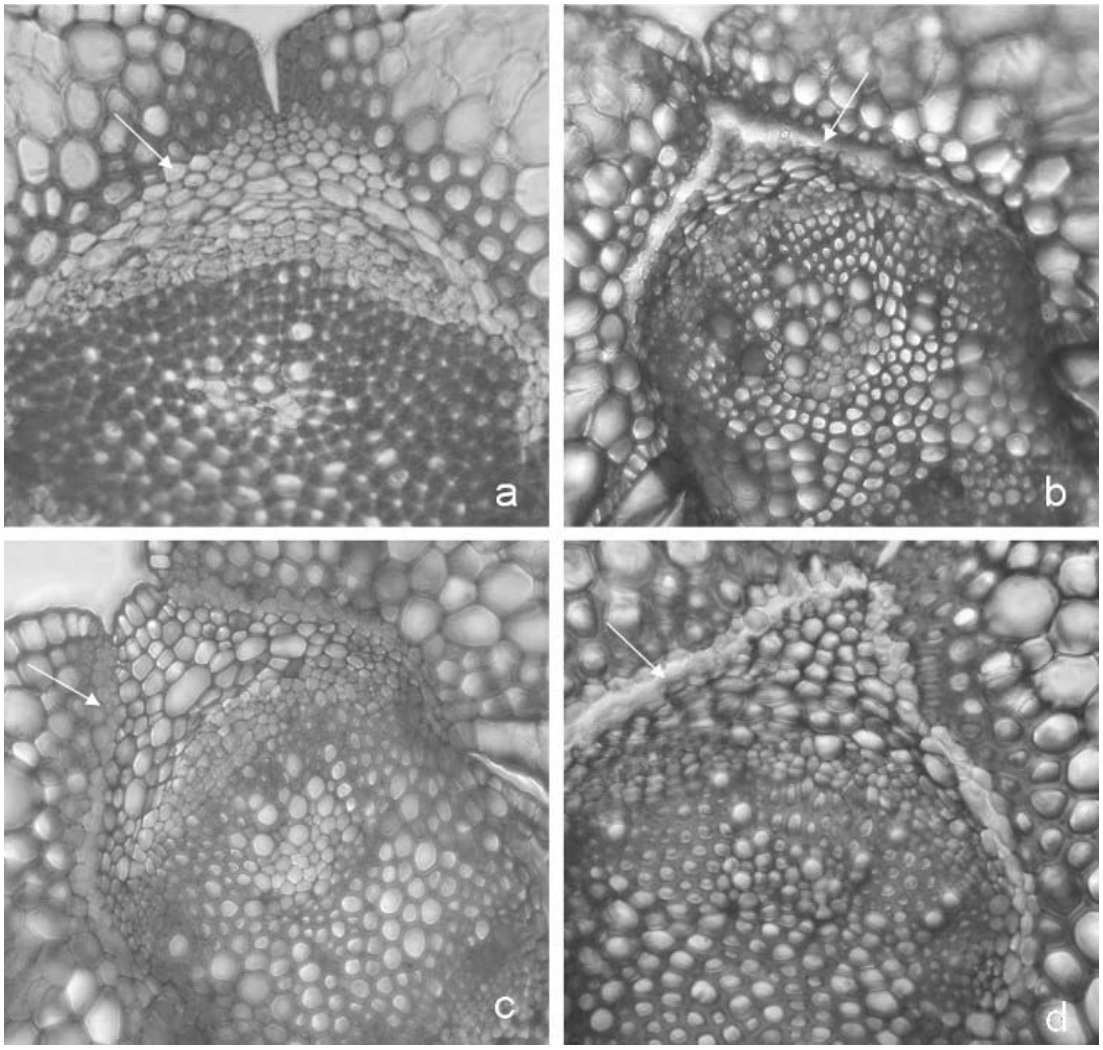

Fig. 2. Transverse section showing differences in the structure of shattering resistant and susceptible siliquae. a. *B. carinata* (note thick walled valve edge cells and absence of dehiscence zone) b. *B. napus* (note separation of cells in the dehiscence zone) c. Resistant hybrid (note intact thick walled cells in the dehiscence zone) d. Susceptible hybrid (note separation of cells in the dehiscence zone).

7.6.3 Key findings

- The new sources of shatter resistance have the potential to greatly benefit the Australian and Indian breeding programs.

7.6.4 Recommendations

- Breeding populations and *B. carinata* introgression lines need to be tested in all three countries to confirm shatter resistance and stability.
- *B. carinata* introgression lines should be tested for resistance/tolerance to other abiotic stresses and diseases.

7.7 Agronomic traits (Objectives 1, 2 & 3)

7.7.1 Aims

i. To identify new sources of variation for agronomic and quality traits.

ii. To enhance B. napus and B. juncea germplasm through crossing and selection for agronomic traits.

7.7.2 Summary of results

Development of screening protocols (Objective 1)

Existing standard protocols for assessing agronomic traits were used (Appendix 22).

Identification of variability (Objective 2)

B. napus

Overall, lines from the country of origin were generally the highest yielding in trials in that country, although some of the Chinese and Australian *B. napus* lines yielded better than the local cultivars in India (Appendix 12). In Australia, the yield of the Chinese and Indian lines was affected by their susceptibility to blackleg, even though seed dressing fungicides were used at some sites.

Cultivar and country differences in days to 50% flowering were evident (Table 17). In China (Wuhan), all Australian and Indian lines flowered much earlier than the Chinese accessions and the earliest lines suffered damage from freezing. Likewise, the Chinese lines were very late flowering in India, whereas the Australian lines had a similar range of flowering time as Indian lines. In Australia, the Chinese lines were on average later flowering and Indian lines on average earlier flowering than the Australian lines.

Table 17. Days to 50% flowering of *B. napus* lines averaged over two seasons in each country

Country	Series	Chinese lines	Australian lines	Indian lines
		Range and Average	Range and Average	Range and Average
Aus	I	80-98 (ave 89)	76-92 (ave 83)	79-83 (ave 80)
China	I	160-175 (ave 167)	111-155 (ave 137)	140-146 (ave 143)
India	I	104-127 (ave 115)	70-98 (ave 86)	87-9 (ave 90)
Aus	II	87-100 (ave 92)	80-95 (ave 87)	83-84 (ave 83)
China	II	153-176 (ave 162)	138-154 (ave 147)	112-113 (ave 113)
India	II	104-138 (ave 115)	69-121 (ave 89)	67-69 (ave 68)

The range of average oil content among series I lines was similar in Australia and India. However, in China, higher oil content was observed in the series I lines, most likely due to environmental effects (Appendix 13). There was similarity in the lines with highest oil

content across countries (Appendix 13). Likewise, the lines with highest protein content were similar.

The series I *B. napus* lines from Australia and China displayed similarly low glucosinolate content across sites, with the exception of two Chinese lines (Zhongyou 821 and Ding 110) that were high in glucosinolates and erucic acid (Appendix 14). The three Indian series I *B. napus* lines (GSL1, GSL2 and Neelam) were also not canola quality. The actual glucosinolate measurements varied between sites and countries, reflecting environmental differences and also differences in analytical techniques. Glucosinolate analysis of the series II lines showed that a few of the lines from China had levels as low as or lower than the best Australian lines (Appendix 14). Erucic acid composition of the series I Indian lines was approx 30%, whereas the series II Indian lines had lower erucic acid composition (average 3%; Appendix 15). The Chinese and Australian lines had very low erucic acid composition with the exception of two lines (ZY002 and ZY003).

Examination of the 1000 seed weight of series I lines across sites and years in India showed that Chinese and Indian lines were of a similarly large seed size, whereas, the Australian lines were of smaller seed size (Appendix 16). Comparison of seed weight of Chinese and Australian series II lines in Australia also showed that the Chinese lines were larger. Seed weight of the Chinese lines range from 3.2 to 5.3g compared to a range of 3.0 to 4.7g for the Australian lines.

Screening for blackleg resistance was undertaken at disease nurseries in each Australian state. Overall, most Australian genotypes had higher levels of resistance to blackleg than Chinese and Indian genotypes which had not previously been exposed to blackleg (Table 18). The best in varietal resistance in rankings across the Australian sites was RT108. Chinese lines 04-p34 and P617 (series I) and 06-6-3777 and 05-P71-11 (series II) were consistently the most resistant of the Chinese lines across sites.

Table 18. Blackleg survival percentages of series I and II *B. napus* lines in Australia averaged across years

Institute	Series	Chinese lines		Australian lines		Indian lines	
		Range (%)	Best lines	Range (%)	Best lines	Range (%)	Best lines
NSW	I	2-73	04-p34	23-62	Purler, RR009	21-88	Neelam
SA	I	0-32	P617	9-77	RQ011, RR001	5-31	-
Vic	I	0-53	04-p34	0-81	Surpass400	0-1	-
WA	I	0-8	P617	5-68	RR001	-	-
SA	II	1-45	05-P71-11	22-88	RT108	4-29	GSC5
Vic	II	0-27	06-6-3777	2-66	RT108	0	-
WA	II	2-51	05-P36-R	10-73	RT108	0-13	-

B. juncea

Australian *B. juncea* lines were the highest yielding on average across sites and years in Australia, however, the best yielding Indian lines were comparable to the best performing Australian lines in Australia (Appendix 17). In India, the series I lines from China and India out-yielded the Australian lines. However, for the series II lines in India, the Australian lines yielded as well as or better than the Indian lines.

Cultivar and country differences in days to 50% flowering (Appendix 18), oil content (Appendix 19) and seed weight (Appendix 20) were also evident. For example, the Chinese lines flowered much later in Australia and India and the Indian seeds were much larger than Australian *B. juncea* seeds.

Screening for blackleg resistance was undertaken at disease nurseries in each Australian state. There was a similar range of survival scores between lines from the three countries, with some lines experiencing very low survival (Appendix 21). Best performing lines varied between states.

Thirty of the series I *B. juncea* lines were also screened under alkaline and saline conditions in a controlled environment experiment by University of Melbourne PhD student Mr Muhammad Javid. Alkaline and saline soils are among several factors that limit crop production in the southern Mallee and Wimmera regions of Victoria. Two contrasting genotypes were selected for further experiments to compare the response to different combinations of alkalinity and salinity. The interactive effects of alkaline and salt stress on biomass and sodium ion uptake were significant in both genotypes ($p<0.05$), however, NDR8501 appeared to be far more tolerant to high salt under alkaline conditions than Xinyou 5 (Javid et al. 2009).

Enhancement of germplasm through selection and breeding (Objective 3)

Production of breeding populations of *B. juncea* for enhanced agronomic and quality traits was undertaken at TERI and DPI Vic.

Each institute has also been undertaking selection and breeding for agronomic traits using the germplasm from this project as part of their regular breeding program. For example, at IOCR, more than 200 crosses were carried out between the introduced *B. napus* lines with high oil content and IOCR's main breeding lines. Oil content of 53.5% was obtained in F_3 lines derived from a cross between a Chinese line (98V41) and Australian line (RR001).

7.7.3 Key findings

- Blackleg resistance in the *B. napus* line RT108 from Australia was consistently high across three Australian states. It could be used as a pre-emptive breeding line for blackleg resistance in India and China. Identification of Chinese *B. napus* lines with some tolerance to blackleg may also be useful for pre-emptive breeding for blackleg resistance in China.
- Chinese late flowering *B. napus* may have potential in Australia for use in developing varieties for spring sowing and the early vigour of the Chinese lines may also be useful for breeding grazing canola.
- New canola quality *B. juncea* lines from Australia will be widely used in Chinese and Indian breeding programs.
- Larger seed size from Indian *B. juncea* will benefit Australian and Chinese *B. juncea* breeding.
- The range of resistance to blackleg in *B. juncea* from each country will assist all breeding programs.
- Abiotic screening for tolerance to salinity and alkalinity identified the Indian line NDR8501 as relatively more tolerant than the other *B. juncea* lines.

7.7.4 Recommendations

- Double haploids and marker assisted selection will be essential longer term for incorporation of useful traits from Indian *B. juncea* material (e.g. yield, days to flowering, seed size) into Australian germplasm without a detrimental effect on quality.

7.8 Breeding populations summary (Objective 3)

7.8.1 Aims

i. To enhance germplasm through selection and breeding for each of the key traits

7.8.2 Summary of results

Each breeding organisation in the project was responsible for the development of one or more germplasm pools, targeting improvements in specific key characters (Table 19). After a generation of increase in Australia these breeding populations have been made freely available for use by breeders in the partner countries.

Table 19. Project breeding populations developed as part of the project and name of organisations responsible

Institute	Breeding populations being developed	Species	Name	Pedigree	Generation	Details
HZAU, China	Sclerotinia tolerance	*B. napus*	HZAU-1 HZAU-2 HZAU-3 HZAU-4 HZAU-5 HZAU-6	-	-	Sclerotinia tolerance " " " " " "
IOCR, China	Enhanced genetic diversity	*B. napus*	OCRI-1 OCRI-2 OCRI-3 OCRI-4 OCRI-5 OCRI-6	R1×Lantern R1×Neelam R6×Rainbow (R2×204-1)×Charlton 1055×RR001 [(1008×8908) ×5899]×Purler	F_5 F_5 F_5 F_4 F_4 F_4	Double low, early Double low, high oil content Double low Double low, high oil content Double low, high oil content Double low, high oil content

Institute	Breeding populations being developed	Species	Name	Pedigree	Generation	Details
NRCRM, India	Drought- and thermotolerance	*B. juncea*	BPR 1153 BPR 1165 BPR 1187 BPR 1191	JN033 x [(BJ1058 x PCR7) x (TERI OE M21 x PCR7)] JR042 x [(BJ 1058 x PCR7) x (TERI OE M21 x PCR7)] Xinyou 9 x (NUDhYJ-3 x Varuna) Xinyou-5 x (NUDhYJ-3 x PCR7)	F_2 F_2 F_2 F_2	Drought and high temperature tolerance (high seedling emergence in field, long pods, more seeds/siliqua, high 1000-seed weight and seed yield /plant)
PAU, India	*B. carinata* trait incorporation	*B. napus* / *B. carinata* hybrid	Surpass 400 NCB4 Rivette NCB5	Surpass 400 NCB4 (Chromosome no. = 40)[a] Rivette NCB5 (Chromosome no. = 39)[a]		B-genome presence, pod shape, hard stem B-genome presence
		B. juncea / *B. carinata* hybrid	JC 134 JC 67	JC 134 (Chromosome no. = 36)[a] JC 67 (Chromosome no. = 36)[a]		White rust resistance White rust resistance, hard stem
HAU, India	Drought tolerance, thermotolerance and sclerotinia tolerance	*B. juncea*	HAU-1 HAU-2 HAU-3	RH0270 x JM018 RH9902 x JN028 RH0202 x JN004	BC_2F_2 BC_2F_2 BC_2F_2	Sclerotinia tolerance Sclerotinia tolerance Drought tolerance
		B. napus	HAU-4 HAU-5 HAU-6 HAU-7	Fan 028 x Charlton Ding 110 x Oscar Lantern x GSC 3A Surpass 400 x HNS 0501	BC_2F_2 BC_2F_2 BC_2F_2 BC_2F_2	Sclerotinia tolerance Sclerotinia tolerance Thermotolerance Drought tolerance
TERI, India	Shatter resistance	*B. juncea*	TERI-1 TERI-2	Pusa Bold x JN010 PCR 7 x JR042	BC_3	Enhanced agronomic and quality traits
		B. napus	TERI-3 TERI-4	Tranby x R9903[b] Surpass x R9903[b]	BC_3 BC_3	Shatter resistance Shatter resistance

Institute	Breeding populations being developed	Species	Name	Pedigree	Generation	Details
Vic DPI, Aus	Canola quality and white rust resistance	*B. juncea*	Lines 1 to 4	-	Double haploid	Canola quality and white rust resistance
NSW DPI, Aus	Enhanced quality and blackleg resistance	*B. napus*	-	[c]	-	-

[a] *One backcross followed by five generations of selfing and selection.*

[b] *R9903 = TERI(00)R9903*

[c] *Lines lost due to successive droughts*

7.9 Genetic diversity and heterosis in *B. napus* (Objective 4)

7.9.1 Aims

i. To examine genetic diversity in Australian, Chinese and Indian B. napus.

ii. To measure heterosis in B. napus F_1 hybrids.

iii. To report the relationship between genetic distance and heterosis in B. napus.

iv. To estimate heritability for each trait.

7.9.2 Summary of results

Genetic diversity

UWA-C

At UWA-C, allelic diversity of 72 *B. napus* genotypes representing contemporary germplasm in Australia, China, India, Europe and Canada, was characterised by 55 polymorphic simple sequence repeat (SSR) markers spanning the entire *B. napus* genome. Data was analysed using both hierarchical clustering and two-dimensional multidimensional scaling (MDS). The results identified a Chinese group (China-1) that was separated from a "mixed" group of Australian, Chinese (China-2), European and Canadian lines. A small group from India was distinctly separated from all other *B. napus* genotypes. Chinese genotypes, especially in the China-1 group, have inherited unique alleles from interspecific crossing, primarily with *B. rapa*, and the China-2 group has many alleles in common with Australian genotypes (Figure 3).

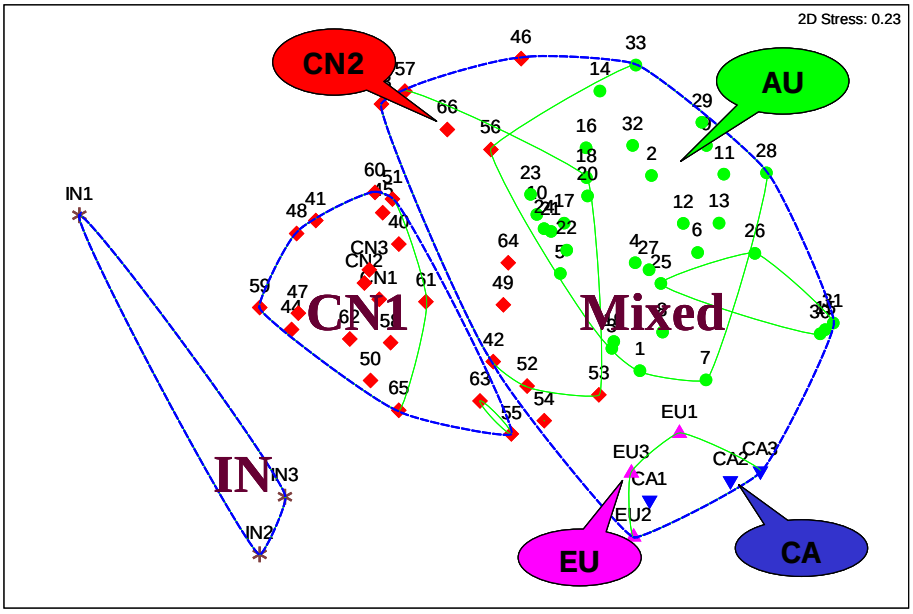

Fig. 3. 2-D MDS analysis of series I lines at UWA-C (IN=Indian lines, CN1=Chinese lines, CN2=Chinese lines, AU=Australian lines, EU=European lines, CA=Canadian lines) (Chen et al. 2008).

HZAU

At HZAU, genetic diversity of the series I germplasm was analysed using SSR and sequence-related amplified polymorphism (SRAP) techniques. Of the 114 SSR primer

combinations, 81 produced 195 polymorphic bands among 48 *B. napus* and 51 SRAP primer pairs produced 207 polymorphic bands. Molecular genetic distance detected using SSR markers was significantly correlated to that detected using SRAP markers (r=0.6135, P=0.002). However, the SSR markers detected genetic distance more accordant with the origins of the accessions. A dendrogram generated from the SSR data showed that the Chinese, Australian and Indian lines clustered based on their country of origin, with sub groups within each cluster that often reflected the national breeding program from which the lines originated (Figure 4).

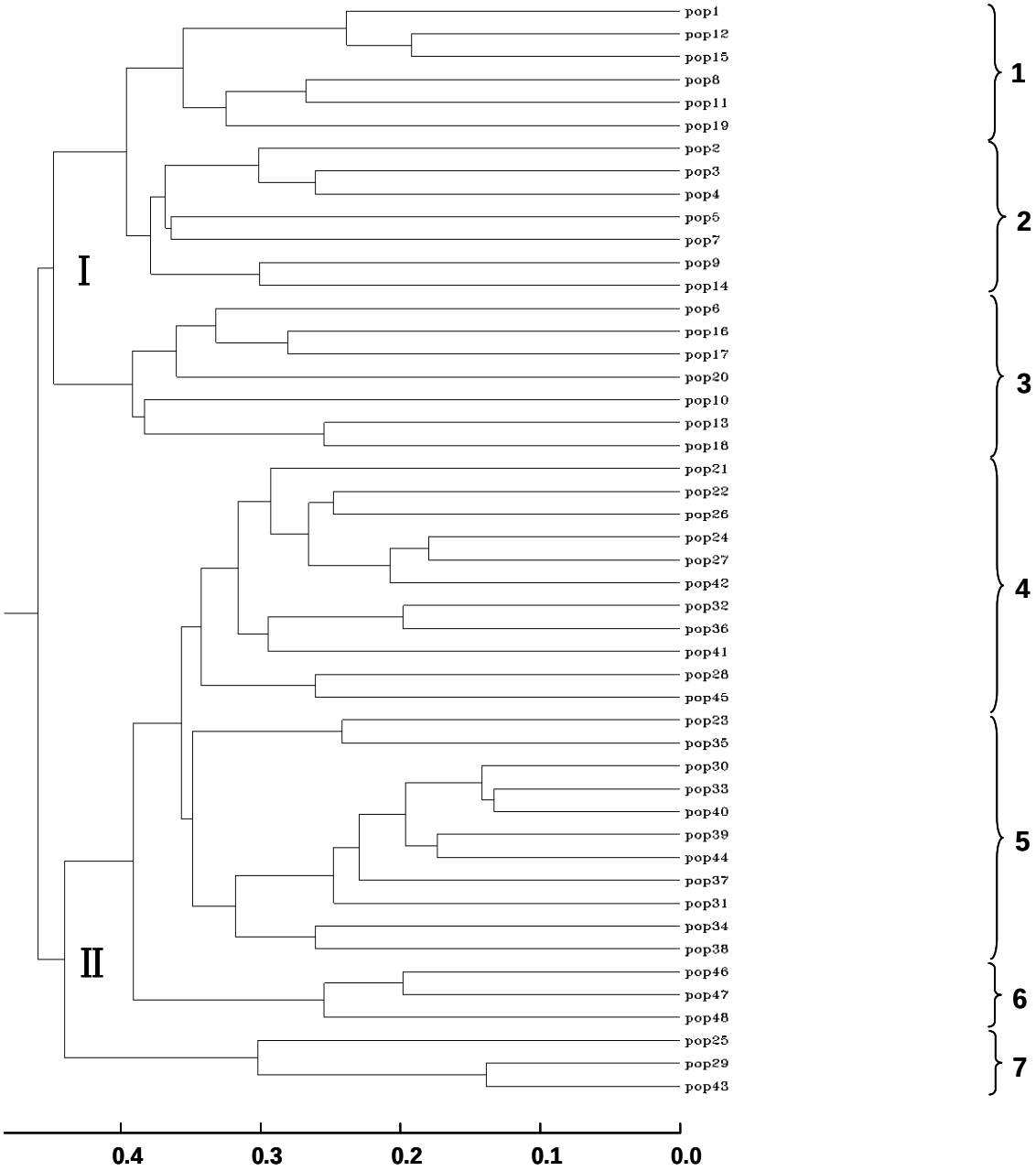

Fig. 4. Clustering analysis at HZAU of series I *B. napus* lines (note: OCRI = IOCR)

(Rainboa=Rainbow, Churlfoh=Charlton, AG-orfback=AG-Outback, Triloqy=Trilogy, Trlgold=Trigold)

PAU

At PAU, SSR analysis of series I *B. napus* lines found that many Indian lines clustered separately to the Australian and Chinese lines (Figure 5), which concurs with the results of

the analysis by Chen et al. (2008). In this study, additional Indian *B. napus* lines were included.

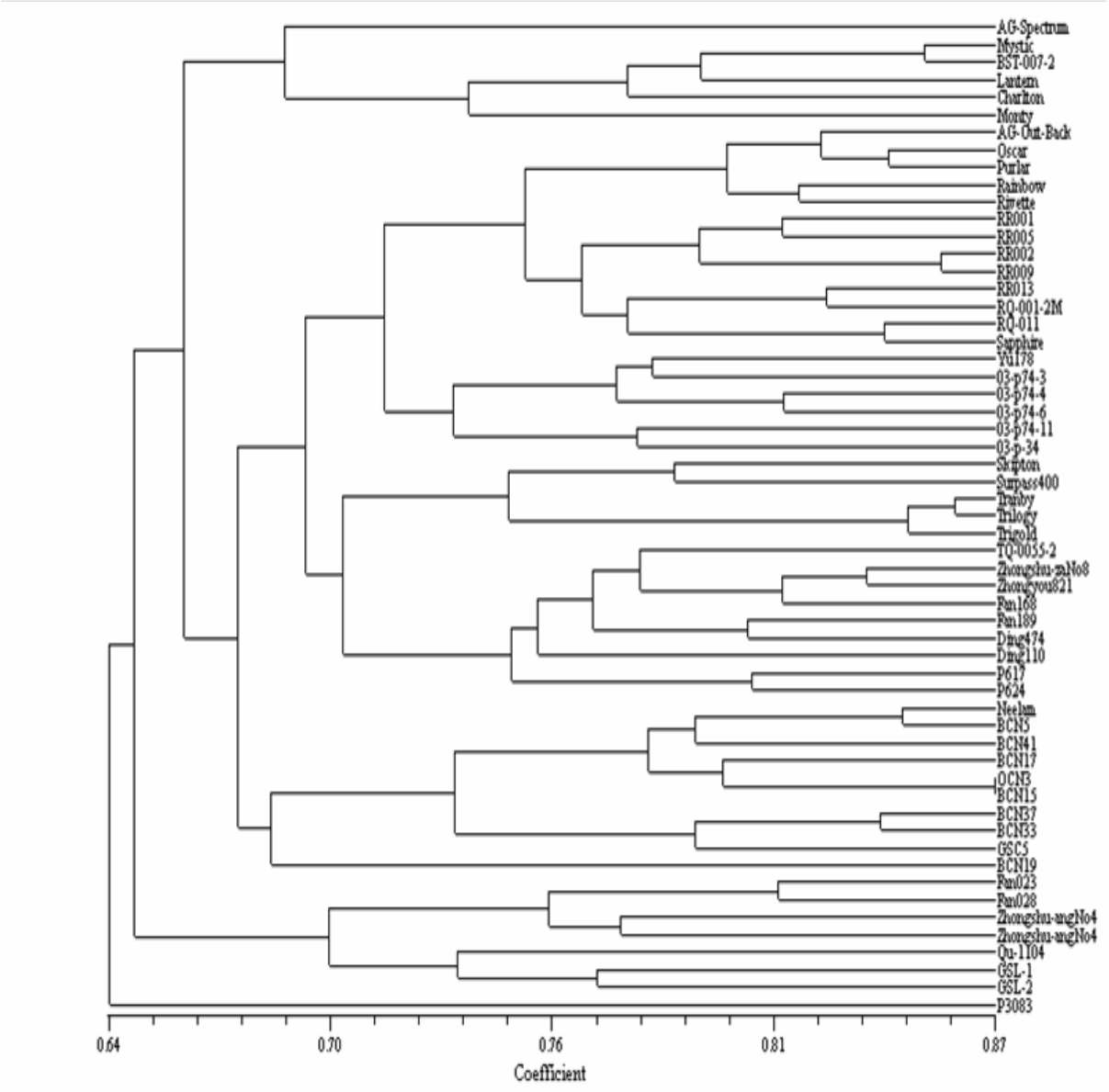

Fig. 5. Clustering analysis at PAU of series I *B. napus* lines based on SSR markers.

F_1 hybrid vigour analysis

UWA-C coordinated experiment in Australia, China and India

Based on the allelic diversity and the genetic distinctiveness of *B. napus* genotypes, 13 genotypes were selected as parents for the hybrid vigour experiment (4 Australian, 7 Chinese, 1 Indian and 1 European line; Table 20). Pure breeding lines of the parents were developed through selfing single plants. A half-diallel crossing population was generated between the 13 parent lines. In total, 78 crosses, plus 6 reciprocal crosses (84 F_1 hybrids) plus 13 parents (a total of 97 entries) were produced by hand crossing.

In the preliminary diallel cross program at UWA-C, F_1 hybrid seed was produced via hand-crossing in 2006 at UWA. About 800 F_1 hybrid seeds per cross on average were

produced and distributed in 2007 together with pure parental seed for sowing in Australia (WA, NSW, Vic), China (HZAU, IOCR) and India (PAU).

During 2007, the major cross program was undertaken for the Year 4 diallel evaluation study. About 1000 F_1 hybrid seeds per cross on average were produced via hand-crossing at UWA for the hybrid trial in 2008. Seed was distributed during 2008 to Australia (WA, NSW, Vic), China (IOCR) and India (PAU, HAU).

Table 20. *B. napus* lines used in a joint hybrid vigour experiment coordinated by UWA-C

Parent	Origin	Source	Notes
Rivette	Australia	NSW DPI	Cultivar released in 2000
Surpass400	Australia	DPI Vic	Cultivar released in 2000
RR001	Australia	DPI Vic	Breeding line
Karoo-057DH	Australia	CBWA	Double haploid (DH) line derived from cultivar Karoo
Campino	Europe	NPZ	Spring cultivar derived from Lambada/PF 50/95
Zhongshuang4	China	IOCR	Cultivar released in 1993; pedigree: Zhongyou821/Zhongshuang2
Zhongyou821	China	IOCR	Cultivar released in 1985; pedigree: Suzhao3///Baiyou1/Yunyou7// Ganyou3/Ganyou1
03-p74-3	China	Huazhong Agricultural University	Derived from Zhongyou821/ 81001//218/Regent
03-p74-6	China	Huazhong Agricultural University	Selected from Hui5148-2
CN01-104-2	China	Huazhong Agricultural University	*B. napus* x *B. rapa* descent
HAU02	China	Huazhong Agricultural University	Breeding line derived from interspecific hybridisation with Chinese *B. rapa*
HAU11	China	Huazhong Agricultural University	Breeding line derived from interspecific hybridisation, i.e. Chinese B. napus x (Ethiopian *B. carinata x* Chinese *B. rapa)*
GSL1	India	ACIAR Series I line from India	Cultivar released in 1986

The hybrid field trials were designed with a spatial randomisation program (DiGGer, developed by NSW Agriculture) for unbalanced designs. There were a variable number of 1 to 3 replicates per entry per site (some hybrids had only enough seed for 1 replicate per site, while others had 3 replicates per site). The seven agronomic traits that were measured at the sites in each country were vegetative vigour, date of 50% flowering, height of first branch, height of first pod, mature height, seed yield and 1000 seed weight. In addition, analysis of oil, protein and glucosinolate content of a 30g sample of seeds from each plot was undertaken.

The data across 12 experiments at 7 sites over 2 canola growing seasons (2007/08 and 2008/09) were analysed by mixed model techniques (Smith et al. 2001) using adjustments for spatial trends in the diagonal model (analogous to single site/year experiment analysis), before proceeding to multi-environment trial (MET) analysis with factor analytic (FA) models with all sites/years included in the model. Analysis was conducted using the software ASReml in the R environment (Butler et al. 2007).

Preliminary analysis of data by UWA-C is presented in Figures 6 and 7. Heterosis levels for yield exceeded 100% in some crosses. The highest yielding hybrids were Chinese x Chinese and Chinese x Indian. One Chinese parent in the best hybrids (HYB-072 and HYB-49) was derived from interspecific hybridisation with *B. rapa* and *B. carinata*.

In HYB-072, the hybrid with best yield performance over 12 trials, strongest mid-parent and hi-parent heterosis was observed in yield, followed by moderate mid-parent and hi-parent heterosis in leaf area, plant height and oil content. No obvious heterosis was observed in flowering time or in 1000 seed weight (Figure 7).

Examination of the three pairs of reciprocal hybrids showed no significant difference in Chinese cytoplasm or in Australian cytoplasm.

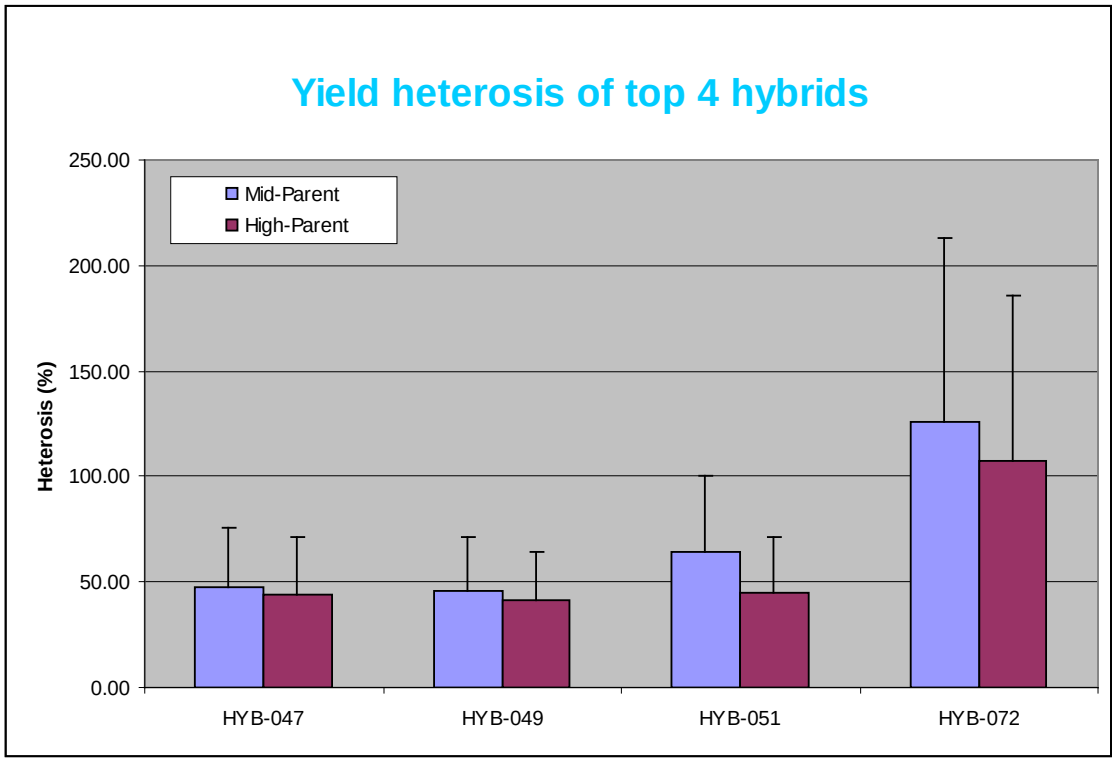

Fig. 6. Mid-parent and Hi-parent heterosis of yield in the top 4 hybrids (HYB-047= Zhongshuang4x03-p74-6; HYB-049= Zhongshuang4xHAU02; HYB-051= Zhongshuang4xGSL1; HYB-072=CN01-104-2xHAU11)

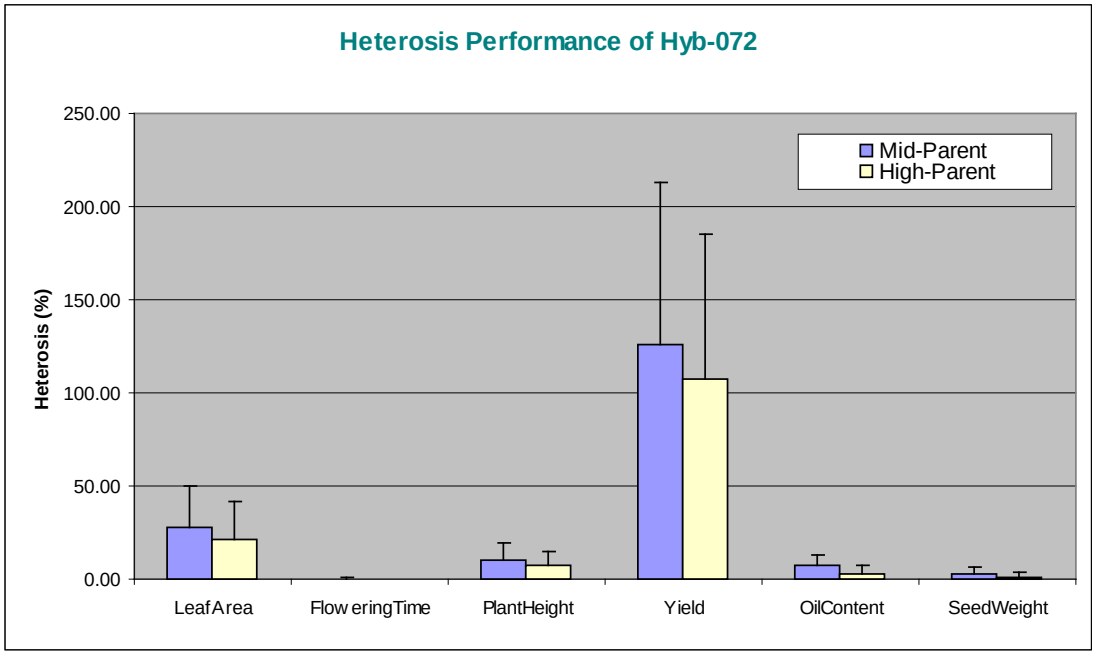

Fig. 7. Mid-parent and Hi-parent heterosis of 6 traits in Hyb-072, the hybrid with best yield performance over 12 trials.

A strong correlation between yield and leaf area was observed at each site in both years (Figure 8)

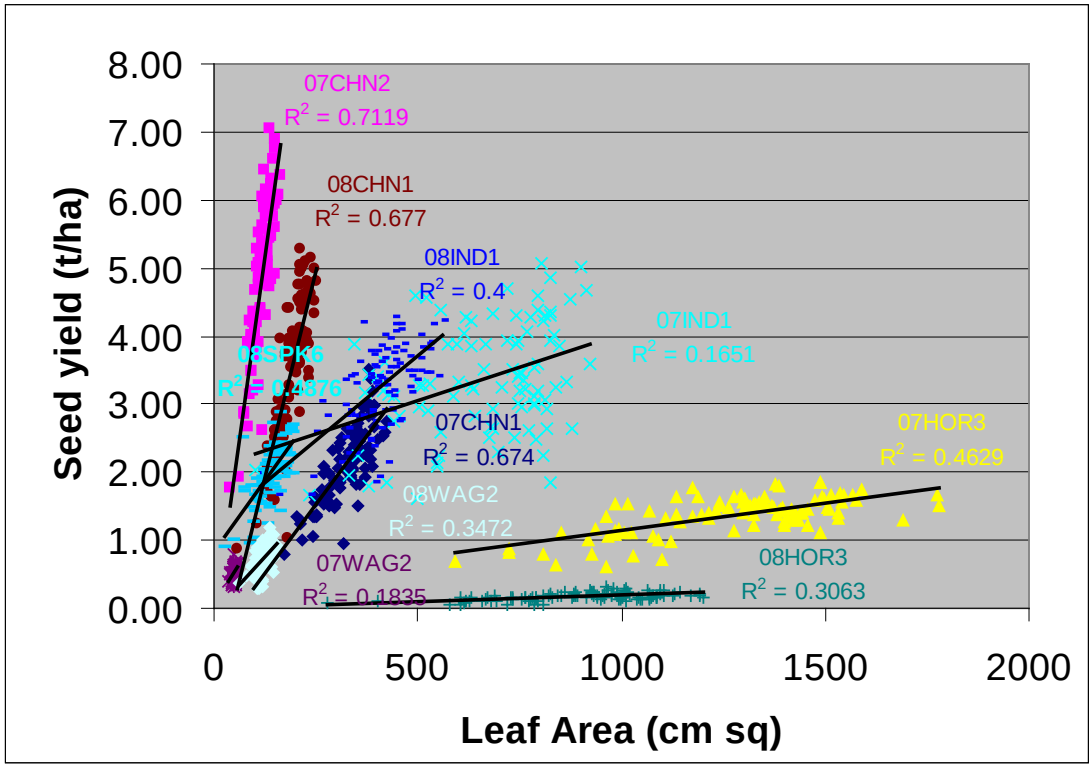

Fig. 8. Relationship between yield and leaf area at each site.

Heritability

An estimate of heritability of each trait after multi-environment trial (MET) analysis of all sites/years is presented in Table 21. In the preliminary analysis, pedigrees were not included, however further analysis will include pedigrees, which will allow estimation of additive, dominance and epistatic variance across sites. Additive genetic variance will be equivalent to General Combining Ability (GCA), and the deviation from this in individual crosses will be Specific Combining Ability (SCA).

Table 21. Heritability (h2) after MET/FA analysis. Heritability is defined as the percentage contribution of genetic to total variance.

Trial	Site	Year	Heritability				
			Leaf area	Height	Oil content	1000 seed weight	Yield
07CHN1	IOCR	1	0.89	0.85	0.81	0.94	0.89
07CHN2	HZAU	1	0.81	0.93	0.87	0.91	0.9
07HOR3	Horsham	1	0.77	0.91	0.96	0.84	0.87
07IND1	Punjab	1	0.81	0.87	NA	NA	0.94
07SPK6	Shenton Park	1	0.79	0.82	0.88	0.75	NA
07WAG2	Wagga Wagga	1	0.5	0.78	0.87	0.7	0.61
08CHN1	IOCR	2	0.89	0.92	0.79	0.89	0.89
08HOR3	Horsham	2	0.75	0.84	0.88	0.81	0.66
08IND1	Punjab	2	0.81	0.92	NA	0.9	0.92
08SPK6	Shenton Park	2	0.85	0.85	0.9	0.92	0.79
08WAG2	Wagga Wagga	2	0.92	0.86	0.95	0.78	0.79

Relationship between genetic distance and heterosis

UWA-C coordinated experiment in Australia, China and India

Examination of the relationship between genetic distance and heterosis based on the UWA-C coordinated experiments found no significant correlation between genetic diversity and heterosis, though a marginal positive correlation was observed for plant height and a marginal negative correlation for 1000 seed weight.

At UWA-C, 55 SSR markers were originally used for the genetic distance study in *B. napus*. These SSR markers revealed 220 polymorphic alleles among the 13 parent lines. However, this number was not enough to study the relationship between genetic distance and heterosis, particularly in such a multiple parent hybrid trial. Thus, in 2009 Diversity Array Technology (DArT) genotyping was applied to the 13 parent lines to produce more markers. 1918 polymorphic DArT alleles were identified. These DArT alleles greatly increased the resolution for trait-marker association.

These alleles, together with the 220 SSR polymorphic alleles, were then used in an attempt to identify genes and gene interactions responsible for hybrid vigour. Hybrid vigour is important to develop novel plant varieties. However, it has been difficult to discover genes for hybrid vigour in plants until the latest developments of association mapping. With the support of the GRDC In-Service Training award, Dr Sheng Chen visited Prof Carlos Bustamante's laboratory at Cornell University for 3 weeks in early 2009. The visit was very beneficial to integrate knowledge across multiple disciplines including molecular genetics, genomics, statistics and agronomy to discover novel alleles associated with agriculturally important traits. This knowledge has been applied in the data analysis of this project to reveal hybrid vigour related alleles and allelic interactions in canola. A program was developed for the association analysis over these phenotyping

and genotyping data based on (a) linear regression with only fixed effect, i.e. treat “Site” as fixed effect assuming the variance is the same across 7 sites; and (b) mixed model, i.e. treat “Site” as random effect, variance in each site is different. In this way, we are now able to detect the molecular marker alleles associated with yield and to exploit the genetic effects including additive, dominant and epistatic effects once a number of alleles associated with a trait are found. A new approach has also developed to identify unique alleles for hybrid vigour. Mid-parent heterosis (MPH) and high-parent heterosis (HPH) is being applied to all these traits for association analysis to reveal the allele(s) for hybrid vigour in canola hybrids.

HZAU experiment

At HZAU, 12 parents were used to produce 36 hybrids (6 Chinese x Chinese, 13 Chinese x Australian, 5 Chinese x Indian, 6 Australian x Australian, 5 Australian x Indian, 1 Indian x Indian), which were planted in three trials in China (Wuhan and Lanzhou) from 2006 to 2008 using a randomized complete block design with three replicates (Table 22). Eleven phenotypic traits were recorded on ten to fifteen plants per plot. An analysis of variance and correlation analysis were performed on phenotypic traits and mid-parent heterosis using the statistical package SAS.

Table 22. *B. napus* lines used in the HZAU hybrid vigour experiment

Parent	Origin
Ag-Spectrum	Australia
Skipton	Australia
Av-Sapphire	Australia
RR001	Australia
RQ011	Australia
Fan023	China
Zhongshuang9	China
Yu178	China
J474	China
03-p74-6	China
Neelam	India
GSL2	India

Plant height and the first branch height were observed to be significantly positively correlated in two environments. However, siliques per plant, seeds per silique, seed weight and seed yield were not correlated in two environments. Positive mid-parent heterosis and positive high parent heterosis of seed yield were observed at the two locations.

For the 36 F_1 hybrids, most of the correlations between genetic distance and mid-parent heterosis were positive, but few items were at a significant level. However, when intra- and inter-regional hybrids were analysed separately, correlations between genetic distance and mid-parent heterosis were higher among intra-regional hybrids for most traits, especially siliques per plant and seed yield per plant (Table 23).

Table 23. Correlations between genetic distance and mid-parent heterosis among intra- and inter-regional hybrids.

Trait		SSR		SRAP		SSR & SRAP		SSR + SRAP	
		Wuhan	Lanzhou	Wuhan	Lanzhou	Wuhan	Lanzhou	Wuhan	Lanzhou
Plant height	intra-regional	0.28	0.08	0.29	0.10	0.30	0.10	0.38	-0.15
	inter-regional	-0.33	0.49*	-0.12	0.36	-0.27	0.50*	0.07	0.33
Height of 1st branch	intra-regional	0.10	0.02	0.00	0.35	0.04	0.21	0.05	-0.03
	inter-regional	-0.37	0.46*	0.08	0.19	-0.17	0.36	0.00	0.36
Siliqua/ plant	intra-regional	0.63*	0.55	0.69**	0.40	0.71**	0.51	0.67*	0.59*
	inter-regional	0.04	0.00	-0.31	0.27	-0.17	0.16	0.02	-0.24
Seeds/ silique	intra-regional	-0.04	0.19	0.05	0.29	0.01	0.25	-0.24	0.10
	inter-regional	-0.12	-0.04	0.06	0.33	-0.03	0.18	0.11	0.09
1000 seed weight	intra-regional	-0.14	0.03	-0.32	-0.24	-0.25	-0.12	-0.11	0.13
	inter-regional	0.02	-0.13	-0.11	-0.01	-0.06	-0.08	-0.29	0.00
Yield/ plant	intra-regional	0.45	0.61*	0.61*	0.32	0.58*	0.49	0.33	0.41
	inter-regional	0.10	0.01	0.24	0.31	-0.21	0.20	0.07	0.07

7.9.3 Key findings

- Genetic distance analyses of *B. napus* germplasm using the SSR technique showed abundant genetic diversity among the lines of both species, and will assist breeders in all countries in their selection of the most diverse lines to widen their genepools.
- Hybrid vigour provides valuable yield improvements in canola. Highest levels of heterosis tended to be found in intra-country hybrids, rather than inter-country hybrids.
- The analysis of agronomic and molecular marker data (SSR and DArT) in canola hybrids and their parents will help improve the efficiency of breeding for hybrids in canola.

7.9.4 Recommendations

- More detailed analysis of the multi-environment data will provide further understanding and benefit to hybrid breeders.
- Further work is required to identify and understand the role of the key genes involved in heterosis.

7.10 Genetic diversity in *B. juncea* (Objective 4)

7.10.1 Aims

ii. To examine genetic diversity in Australian, Chinese and Indian B. juncea.

7.10.2 Summary of results

UWA-C

Genetic diversity was examined in 123 *B. juncea* accessions and five accessions representing other *Brassica* species in U's Brassica triangle. A total of 53 *B. juncea* accessions were from China, 46 from India, 12 from Australia, 9 from France and 3 from Ukraine. Ninety nine SSR primer pairs containing 51 from the A-genome and 48 from the B-genome were used for *B. juncea* genetic distance analysis (sequences for primers were provided by Dr Andrew Sharpe, Agriculture and AgriFood Canada). SSR alleles were evenly distributed on the A and B genome (Table 24). Chinese *B. juncea* (CN) had 1.70 (56/53) and 2.92 (70/24) times more distinct alleles than Indian *B. juncea* (IN) in the A-genome and B-genome, respectively (Table 25).

Data were analysed using both hierarchical clustering and two-dimensional multidimensional scaling. The results showed that some Indian and Chinese *B. juncea* formed one group which is apparently distinguished from another mixed group which consists of all the other Indian and Chinese lines as well as the *B. juncea* from Australia, France and Ukraine (Figure 9). Even in the mixed group, some Indian and Chinese lines formed a subgroup respectively to distinguish from the other lines. These results indicate that abundant genetic diversity exists in both China and India, and that the *B. juncea* germplasm from India and China will be invaluable in broadening the Australian *B. juncea* genetic diversity. However, since not many ancestral lines were included in this study, and the origin of many accessions is unknown, it was hard to identify evolutionary trends in terms of the private alleles (alleles found only in one population). Thus, 32 extra *B. juncea* landrace lines were added to this study. Most of these lines were from the ATFCC Brassica Collection in Horsham, Victoria and their origin was relatively clear. With these accessions the evolutionary relationships of the private alleles in Indian and Chinese *B. juncea* lines may be able to be tracked.

Table 24. Distribution of SSR alleles in *B. juncea* were evenly distributed on the A and B genome in *B. juncea*

Genome	Chromosome number	SSR number	Allele number	Alleles/chromosome	Alleles/SSR
A	10	51	337	33.7	6.6
B	8	48	297	37.1	6.2
Total	18	99	634	35.2	6.4

Table 25. Distribution of distinct alleles in Chinese (CN) and Indian (IN) *B. juncea*

Category	A-genome		B-genome		A+B	
	No. alleles	percentage	No. alleles	percentage	No. alleles	percentage
CN only	56	23.8%	70	29.9%	126	26.9%
IN only	33	14.0%	24	10.3%	57	12.2%
CN & IN shared	146	62.1%	140	59.8%	286	61.0%
Total	235	100%	234	100%	469	100%

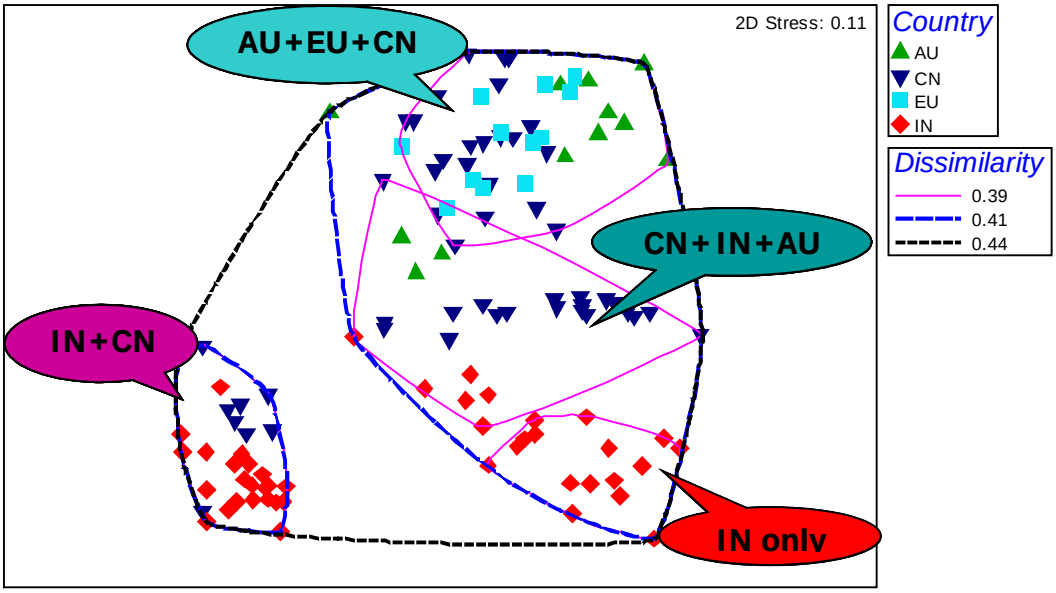

Fig. 9. Bi-plot of the first two MDS scales showing relationships among 109 *B. juncea* genotypes revealed by 2-D MDS analysis, based on 647 alleles from 99 SSR markers.

PAU

At PAU, SSR analysis of series I *B. juncea* lines found that there were distinct groupings of most of the Indian, Australian and Chinese lines (Figure 10).

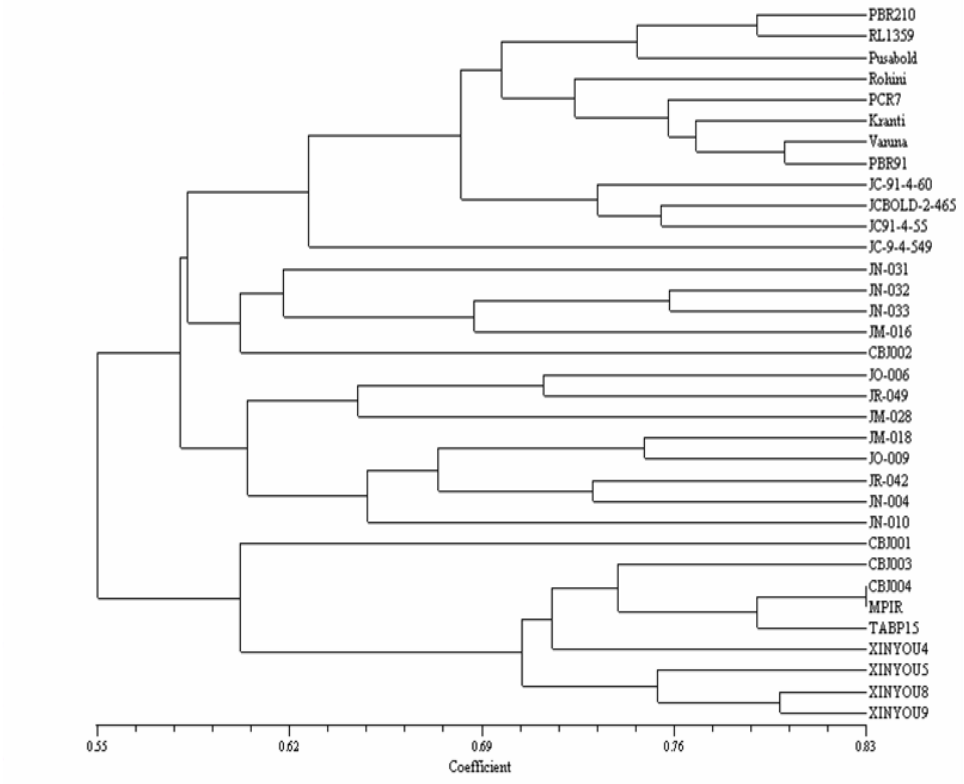

Fig. 10. Clustering analysis at PAU of series I *B. juncea* lines based on SSR markers.

7.10.3 Key findings

- Genetic distance analyses of *B. juncea* germplasm using SSR analysis showed abundant genetic diversity among the Chinese and Indian lines. This diversity will assist breeders in all countries in their selection of the most diverse lines to widen their genepools.

7.10.4 Recommendation

- It would be valuable to replicate for *B. juncea* the multi-environment heterosis study that was done with *B. napus*.

7.11

Information packages (Objective 5)

The two required information packages have been produced for Australia and provided to the project leader:

1. Barbetti MJ, Li C-X, Hua Li, Sivasithamparam K, Garg H (2009) Management in Australia of Sclerotinia Stem Rot (*Sclerotinia sclerotiorum*) in canola (*Brassica napus*) and mustard (*Brassica juncea*), 4 pp. (Appendix 23)

2. Barbetti MJ, Li C-X, Kaur P, Hua Li, Sivasithamparam K, Gurung A (2009) Management in Australia of White Rust (*Albugo candida*) in oilseed mustard (*Brassica juncea*), 4 pp. (Appendix 24)

7.12

Scientific skills enhancement (Objective 6)

- Significant progress was made towards increasing the scientific skills of collaborating scientists through scientific exchanges. These exchanges have also enhanced long term collaboration between Australian, Chinese and Indian scientists.
  - Mr Wan Zhengjie, HZAU, China, completed 5 months of molecular biology training at UWA. Mr Wan's project was "Genetic distance studies on *B. juncea*".
  - Dr Mei Desheng, IOCR, China, completed 6 months of molecular biology training at CSIRO Plant Industry. Dr Mei's project was "Cloning fatty acid biosynthesis genes from the *Crambe abyssinica* oilseed species".
  - Dr Chirantan Chattopadhyay, NRCRM, India, undertook a 3 week training program in Australia in 2007. The visit involved visiting state disease nurseries and attending Australian Research Assembly on Brassicas (ARAB) and Plant Pathology conferences.
  - Dr Maharaj Singh, NRCRM, India, participated in 3 months of training at DPI Vic, Horsham with Dr Rob Norton in 2005. His project was "Use of thermal imaging in screening of *B. napus* and *B. juncea* varieties for drought tolerance".
  - Dr NB Singh, ICAR, India, Dr Arvind Kumar and Dr Chauhan, NRCRM, India, Dr Surinder Banga and Dr Shashi Banga, PAU, India, Dr Dhiraj Singh, HAU, India and Dr Abha Agnihotri, TERI, India participated in a 3 week scientific interaction and planning trip in Australia in 2004. The trip included meetings with project leaders at UWA, tours of field sites in WA and participation in the 4th International Crop Science Congress in Brisbane.
  - Dr Surinder Banga, PAU, India, Dr Dhiraj Singh, HAU, India and Dr Abha Agnihotri, TERI, India also participated in a 3 week scientific interaction/study program in Australia in 2005. The program included 3 days of NIR (oil quality analysis) training at Wagga Wagga Research Institute NSW, visits to Brassica trials in NSW, Victoria and SA, meetings with scientists at CSIRO, Canberra, a meeting with John Cullen (ACIAR) and participation in the biennial conference of the Australian Research Assembly on Brassicas in Port Lincoln, SA.
  - Dr Caixia Li and Dr Martin Barbetti (UWA) attended the International Sclerotinia Workshop held in California in June 2005. Following this workshop, Dr Li visited Huazhong Agricultural University and Wuhan Oil Crops Research Institute in Wuhan, China, where she met with oilseed Brassica scientists working on this ACIAR project, and, in particular, had the opportunity to assess and develop common approaches on study of this disease and of the methods for evaluating host resistance to it.
  - Project collaborators attended progress meetings in China in April 2007 and India in January 2008 and the final project meeting in Australia in 2009. These meetings were an opportunity to discuss progress and also to visit field trials and laboratories in each country to enhance discussion and build linkages. Many new collaborations, including joint supervision of postgraduate students and the initiation of new projects have arisen from these meetings.

Fig. 11. Project participants discussing a field trial at Shenton Park, WA, Australia in 2004

Fig. 12. Project participants meeting with Indian farmers in Rajasthan late in the day as part of the progress meeting field tour in India in 2008

Fig. 13. Project collaborators participating in a progress meeting at Huazhong Agricultural University, Wuhan, China in April 2007

8 Impacts

8.1 Scientific impacts – now and in 5 years

The identification of new sources of disease resistance, agronomic traits, quality traits and drought tolerance traits has positively impacted breeding programs in all three countries. The exchanged germplasm is being widely utilised in the breeding of new cultivars that will lead to increased *Brassica* oilseed production in the partner countries. A few specific examples are described in the following paragraphs.

A small number of Chinese and Australian *B. napus* and *B. juncea* genotypes with relatively good resistance to Sclerotinia infection under Australian field conditions were identified. Lines from China that consistently displayed higher resistance have been used as a new source of Sclerotinia resistance in the Victorian *B. napus* breeding program.

The *B. napus* line RT108 from Australia displayed a consistently high level of blackleg resistance across trials in Australia. It should be used as a pre-emptive breeding line for blackleg resistance in India and China.

Australian *B. juncea* lines were a very good source of white rust resistance relative to local lines in China and India. Thus, this material is being used in Indian and Chinese *B. juncea* breeding programs. Canola quality *B. napus* and *B. juncea* lines from Australia are also being used in Chinese and Indian breeding programs.

Breeding populations have been developed for all key target traits and will be shared with the collaborating institutions. These populations will provide further enhancements for the key traits, positively impacting breeding programs and farmer yields in the future.

The genetic distance analyses of *B. napus* and *B. juncea* lines have had a positive impact on all breeding programs, in assisting breeders to identify which lines are the most divergent, in order to introduce new genetic variability into their breeding populations. The new information on hybrid vigour and genetic distance will lead to more productive hybrid breeding programs in each country.

The improvements in *B. napus* and *B. juncea* production will positively impact all three countries as described in the following paragraphs.

China: The improved canola quality cultivars of *B. napus* (hybrid and open-pollinated) and *B. juncea* will increase the production of canola quality *Brassica* oilseeds in China, both in the middle and lower reaches of the Yangtze River (winter-growing region) and in Xinjiang (summer dryland cropping region) of NW China. The main beneficiaries are the agricultural and food industries in China. Chinese canola researchers (from IOCR and HZAU) have also improved their expertise in Brassica molecular genetics through training in Australia (at CSIRO and UWA).

Australia: Incorporation of the germplasm from China and India into the Australian breeding programs will increase the genetic diversity of the gene pool in Australia. In addition, the Chinese *B. napus* lines with good resistance to Sclerotinia have been used in crosses with Australian lines to incorporate this resistance into the Australian material. Use of material from India with *B. carinata* introgressions will potentially provide the Australian breeding programs with greater drought tolerance and resistance to diseases which will improve the quality and stability of Brassica oilseed yields in Australia.

India: Traits incorporated into *B. juncea* cultivars will benefit the dryland agricultural systems in India while providing healthier canola quality oil and meal. Canola quality *B. napus* and *B. juncea* genotypes will benefit human nutrition in India through a simple substitution of traditional mustards with canola quality cultivars. Indian canola researchers have also improved their expertise in *Brassica* molecular genetics, plant physiology

(drought assessment methods), biochemistry (oil quality analysis) and disease epidemiology through training opportunities in Australia.

A Benefit/Cost analysis of this project was undertaken when the project was being developed. The base level of no ACIAR research funding assumed an increase in yield of 0.5% per annum from existing breeding programs in the three collaborating countries. It also assumed a 1% per annum increase in area sown. However, as a result of the ACIAR project, it is estimated that there will be a further increase of >1% yield increase per annum in each country from the end of the project onwards. Even assuming low adoption rates (a maximum of only 10%) and that there will be no additional area expansion from this project, the analysis at the beginning of the project showed that there will be significant returns from investment in this project with an estimated Net Present Value of $685 million and a Benefit Cost Ratio of 318 over a 25 year period commencing from the beginning of this project.

8.2 Capacity impacts – now and in 5 years

Capacity has been positively impacted by enhancing the skills of scientists through training. The facilities and technical expertise available at the Australian partner institutions has been utilised to provide training and skills development for Indian and Chinese scientists that will enhance the research capacities in both countries.

Dr Abha Agnihotri, Dr Dhiraj Singh, Dr Surinder Banga visited Australia from 20th Sep to 8th Oct 2005 for a scientific interaction/study program. The program included 3 days of NIR training at Wagga Wagga Research Institute NSW, visits to Brassica trials in NSW, Victoria and SA, meetings with scientists at CSIRO, Canberra, a meeting with John Cullen (ACIAR) and attendance at the biennial conference of the Australian Research Assembly on Brassicas in Port Lincoln, SA.

Dr Maharaj Singh (NRCRM, Bharatpur) visited Australia from 5th Sep to 4th Dec 2005 for training. Dr Singh conducted a drought tolerance project supervised by Dr Rob Norton (University of Melbourne) at DPI Horsham using Indian and Australian *Brassica* germplasm. Dr Singh learned to use the Licor 6400, the FLIR thermal camera, the leaf water potential pressure bomb and a range of other relevant equipment. He also visited scientists at CSIRO and ANU Canberra to discuss drought screening and attended the Australian Research Assembly on Brassicas in South Australia. Funding for conference registration and living expenses was supplied by the training funds provided to the commissioned organisation for Year 2 and travel and accommodation were funded from the project budget for NRCRM.

Dr Chirantan Chattopadhyay (Plant Pathologist, NRCRM, India) participated in a 2.5 week training visit to Australia in 2007. Dr Chattopadhyay presented a paper at the Australian Research Assembly on Brassicas, met with Australian Plant Pathologists and Breeders to discuss screening techniques, visited disease nurseries and attended a scientific writing workshop at the Australasian Plant Pathology conference.

In 2006, Mr Wan Zhengjie (PhD student, Huazhong Agricultural University, China) completed 5 months of molecular biology training at UWA (genetic distance studies on B. juncea) and Dr Mei Desheng (Oil Crops Research Institute, China) completed 6 months of molecular biology training at CSIRO Plant Industry (cloning fatty acid biosynthesis genes from the *Crambe abyssinica* oilseed species).

Further capacity building has occurred through exchange of information, materials, visits, interaction, and joint workshops. This has included the progress meetings in China in March 2007 and India in February 2008. It also included the visit by Dr Rod Mailer to the Indian institutes in Feb 2009 to discuss oil quality analysis.

The enhanced capacity of partners will ultimately lead to improved oilseed *Brassica* production in the respective countries.

8.3 Community impacts – now and in 5 years

8.3.1 Economic impacts

The main beneficiaries of this project will be the small farmers in India and China, and Australian farmers. The farmers in India and China currently set aside a small part of their land for oilseed *Brassica* production, often for their own consumption and sell the balance for cash income. This project aims to provide sustainable and increased yields, which will leave the farmers with more cash income from higher surplus production.

The outputs of the project will have direct positive effects on the yields and quality of produce resulting in a higher economic return.

8.3.2 Social impacts

A successful oilseed *Brassica* crop should improve the overall profitability of the farmers in the farming community. It has a role in improving the nutritional status of diet of the resource poor farmers and their families.

With the sustainability in production of oilseed *Brassica* crops, through adoption of higher yielding, better quality, and more disease resistant cultivars, farmers will be encouraged to grow greater areas of oilseed *Brassica*s which will provide employment, particularly to women who are responsible for weeding, harvesting and general management of oilseed *Brassica* crops.

The technology is scale neutral and this research project will not have any negative impact on gender, religious, political, ethnic or demographic groups.

The uptake of canola quality *B. juncea* (India and China) and *B. napus* (China) will also have a beneficial impact related to consumption of healthier oils.

8.3.3 Environmental impacts

Oilseed Brassicas are an integral and important component of the cereal based cropping systems in Australia and elsewhere due to benefits attributable to breaking the monotony of monoculture of cereals and they provide a disease, insect and weed break for cereals crops (eg. rice), reduce the use of pesticides, and thus protect the environment.

The use of improved resistant cultivars and cultural practices will help in sustaining oilseed Brassica production and will ultimately improve soil health with minimal use of chemical pesticides.

8.4 Communication and dissemination activities

8.4.1 Workshops/Meetings:

Feb 2007: Australian National Canola Pathology meeting, Perth WA (project collaborators contributed 6 presentations)

March 2007: 12th International Rapeseed congress (project collaborators contributed 12 presentations)

April 2007: ACIAR project Progress Meeting, 2nd-4th April, Wuhan China

May 2007: White Blister Meeting, Knoxfield VIC

Aug 2007: 14th Annual All India Rapeseed-Mustard Workers Group Meeting, SKUAS&T-Jammu (J&K), Aug 2-4 (review of progress report of the Brassica improvement project in China, India and Australia)

Sep 2007: 15th Australian Research Assembly on Brassicas, Geraldton WA

Oct 2007: Review of Foreign Aided Projects, NCIPM (IARI), New Delhi, October 29

Jan 2008: ACIAR project Progress Meeting, 28 Jan to 3 Feb, India

Feb 2008: Review of Foreign Aided Projects, NASC, New Delhi, February 15

Feb 2008: Australian National Canola Pathology meeting, Melbourne

Sep 2008: Review Meeting of externally aided projects (Oilseed Brassica Improvement In China, India and Australia), Deputy Director General (CS), ICAR, New Delhi.

May 2009:.Review Meeting of Oilseed Brassica Improvement In China, India and Australia, Asstt. Director General (OP), ICAR, New Delhi.

Sep 2009: ACIAR project Final Meeting, 9 -11 Sep, Melbourne, Australia

Sep 2009: 16th Australian Research Assembly on Brassicas, Ballarat Vic

9 Conclusions and recommendations

9.1 Conclusions

The project was highly successful in achieving the overall aim of exchanging germplasm among the three countries in order to enhance productivity of canola quality *B. napus* and *B. juncea*. The long term impact of the exchange will be seen through the new sources of disease resistance, agronomic traits, quality traits and drought tolerance traits the project has produced. They are being utilised in the breeding of new cultivars that will lead to increased *Brassica* oilseed production in the partner countries. The project has also contributed significantly to increasing the scientific skills of collaborating scientists and enhancing long term collaboration between Australian, Chinese and Indian scientists.

Specific conclusions of the project are outlined in the following paragraphs:

Sclerotinia:

- *B. napus* was generally more resistant to Sclerotinia stem rot than *B. juncea.*
- Genotypes ZY006, 06-6-3792, Fan168, ZY004 and Zhongyou-za No.8 from China and genotypes RT108, Oscar and RT057 from Australia showed good resistance to Sclerotinia stem rot in Australia relative to the other lines.
- *B. napus* lines were identified that were consistently more resistant across experiments, providing a positive sign for the screening technique and for confidence in selecting for resistance.
- Variation in pathogenic variability of Sclerotinia isolates between countries was suggested.

White rust:

- Australian *B. juncea* lines are a very good source of white rust resistance compared to local lines in China and India.
- Breeding populations have been produced incorporating canola quality and white rust resistance into Chinese and Indian backgrounds, which will benefit the Chinese and Indian *B. juncea* breeders.

Thermotolerance:

- Substantial variation was evident for all components of seedling and terminal thermotolerance.
- Sources of high temperature tolerance identified are being utilized in the breeding programs.

Drought tolerance:

- *Brassica juncea* lines that regularly performed well in drought tolerance tests across years, testing methods and/or sites were generally the Indian lines. In particular, PCR7, Varuna, Kranti Sej2, NDR8501, RH819 (Indian) and JM018 (Australian) were relatively more drought tolerant.

- Of the extra Australian *B. napus* lines, RT117 appeared to be relatively more drought tolerant, as it showed the least reduction in leaf area and leaf ratio and it also performed well for seed size, seed yield and root area.

Canola quality B. juncea

- New canola quality *B. napus* and *B. juncea* lines from Australia will be widely used in Chinese and Indian breeding programs.

Shatter resistance and B. carinata breeding

- The new sources of shatter resistance have the potential to greatly benefit the Australian and Indian breeding programs.

Agronomic traits

- Blackleg resistance in the *B. napus* line RT108 from Australia was consistently high across three Australian states. It could be used as a pre-emptive breeding line for blackleg resistance in India and China. Identification of Chinese *B. napus* lines with some tolerance to blackleg may also be useful for pre-emptive breeding for blackleg resistance in China.
- Chinese late flowering *B. napus* may have potential in Australia for use in developing varieties for spring sowing and the early vigour of the Chinese lines may also be useful for breeding grazing canola.
- New canola quality *B. juncea* lines from Australia will be widely used in Chinese and Indian breeding programs.
- Larger seed size from Indian *B. juncea* will benefit Australian and Chinese *B. juncea* breeding.
- The range of resistance to blackleg in *B. juncea* from each country will assist all breeding programs.
- Abiotic screening for tolerance to salinity and alkalinity identified the Indian line NDR8501 as relatively more tolerant than the other *B. juncea* lines.

Genetic diversity and heterosis in B. napus

- Genetic distance analyses of *B. napus* germplasm using the SSR technique showed abundant genetic diversity among the lines of both species, and will assist breeders in all countries in their selection of the most diverse lines to widen their genepools.
- Hybrid vigour provides valuable yield improvements in canola. Highest levels of heterosis tended to be found in intra-country hybrids, rather than inter-country hybrids.
- The analysis of agronomic and molecular marker data (SSR and DArT) in canola hybrids and their parents will help improve the efficiency of breeding for hybrids in canola.

Genetic diversity in B. juncea

- Genetic distance analyses of *B. juncea* germplasm using SSR analysis showed abundant genetic diversity among the Chinese and Indian lines. This diversity will assist breeders in all countries in their selection of the most diverse lines to widen their genepools.

9.2 Recommendations

Specific recommendations of the project are outlined in the following paragraphs:

Sclerotinia:

- Definition and monitoring of pathotypes of Sclerotinia will be essential for managing resistance in the future.
- Mechanisms of resistance(s) should be defined.
- Genetics of host resistance(s) in segregating populations should be characterised.

White rust:

- The opportunity exists to further define races and pathotypes of *A. candida* occurring in Australia. In particular, a national standard host differential set (for cotyledon and staghead infection) should be developed.
- White rust screening of the breeding populations developed during the project, particularly those incorporating genes from *B. carinata*, should be undertaken.
- The relationship between staghead formation and vegetative stage resistance requires further investigation.

Thermotolerance:

- The different screening methodologies need to be compared to define better indicators for thermotolerance.
- The methodologies for assessing thermotolerance need to be standardised (e.g. the screening temperature and time exposed to extreme temperature for the seedling stage testing).
- The response of physiological and biochemical parameters need to be compared to yield response to determine the most efficient parameter for screening large populations effectively.

Drought tolerance:

- The different screening methodologies need to be compared to define better indicators for drought tolerance.
- The methodologies for assessing drought tolerance need to be standardised.
- The response of physiological and biochemical parameters need to be compared to yield response to determine the most efficient parameter(s) for screening large populations effectively.

Canola quality B. juncea

- Participation of institutes in the analysis of standard samples provided by Dr Rod Mailer (NSW DPI) is recommended to assist with standardisation of oil testing protocols in each laboratory.

Shatter resistance and B. carinata breeding

- Breeding populations and *B. carinata* introgression lines need to be tested in all three countries to confirm shatter resistance and stability.
- *B. carinata* introgression lines should be tested for resistance/tolerance to other abiotic stresses and diseases.

Agronomic traits

- Double haploids and marker assisted selection will be essential longer term for incorporation of useful traits from Indian *B. juncea* material (e.g. yield, days to flowering, seed size) into Australian germplasm without a detrimental effect on quality.

Genetic diversity and heterosis in B. napus

- More detailed analysis of the multi-environment data will provide further understanding and benefit to hybrid breeders.
- Further work is required to identify and understand the role of the key genes involved in heterosis.

Genetic diversity in B. juncea

- It would be valuable to replicate for *B. juncea* the multi-environment heterosis study that was done with *B. napus*.

10 References

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10.1.1 Journals:

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11 Appendixes

11.1 Appendix 1: Name and characteristic of Series I germplasm

B. napus			
Line	**Number code**	**Source**	**Characteristics**
Rivette		Australia	conventional, early maturity
Oscar		Australia	conventional, mid maturity, wide adaptation
Purler		Australia	conventional, mid maturity, high oil and protein
Lantern		Australia	conventional, mid maturity, high oil and protein
Skipton		Australia	conventional, mid maturity, high oil and protein
Monty		Australia	conventional, very early maturity (older seed used)
Tranby		Australia	early maturity, short height and TT traits
BST7-02M2		Australia	early maturing TT line, adapted to low rainfall Western Australian environments
TQ055-02W2		Australia	early maturing TT line, adapted to low rainfall Western Australian environments
RQ001-02M2		Australia	conventional line with early maturity, adapted to low rainfall Western Australian environments
Trilogy		Australia	Large grain size, very early maturing, triazine tolerant, moderate-high blackleg resistance, widely adapted to low rainfall environments
Trigold		Australia	Early maturing, triazine tolerant, high yielding, high oil content, adapted to medium to low rainfall, moderate resistance to shattering
Ag-Outback		Australia	Early-mid maturity, moderate blackleg resistance, moderate oil content, moderate-high yield
Mystic		Australia	Early-mid maturity, moderate blackleg resistance, moderate oil content, moderate yield
Rainbow		Australia	Mid maturity, moderate blackleg resistance, moderate oil content, moderate-high yield
Surpass400		Australia	Early-mid maturity, high blackleg resistance, high oil content, moderate yield
RR001		Australia	Early maturity, high oil content, high yield
RR002		Australia	Early maturity, high oil content, high yield
RR005		Australia	Early maturity, high oil content, high yield
Charlton		Australia	Mid-late maturity, moderate blackleg resistance, high oil content, low yield (due to blackleg infection)
Ag-Spectrum		Australia	Mid maturity, blackleg resistance, high oil content, high yield
Av-Sapphire		Australia	Mid maturity, blackleg resistance, high oil content, high yield
RQ011		Australia	Mid-late maturity, blackleg resistance, high oil content, high yield
RR009		Australia	Mid maturity, high oil content, high yield
RR013		Australia	Mid maturity, high oil content, high yield
Neelam		India	Suitable for cooler environment
GSL1		India	Wide adaptation

GSL2		India	Atrazine resistant
P3083	30830	China	Double low quality, high oil content, high plant
Fan023	30851	China	Double low quality, big leaves, strong growth vigor
Fan028	30852	China	Double low quality, low branch location and many branches
Zhongshu-ang No.4	30872	China	Double low quality, tolerance to Sclerotinia sclerotiorum
Zhongshu-ang No.9 (originally incorrectly labelled as Zhongshu-ang No.4)	30920	China	Double low quality, high tolerance to Sclerotinia sclerotiorum and virus, lodging resistance
Qu1104	30930	China	Double low quality, long main stem, big seed
Zhongyou-za No.8	30940	China	Pol CMS Hybrid, high oil content
Zhongyou 821	30950	China	Double high quality, high tolerance to Sclerotinia sclerotiorum
Fan168	30960	China	Shattering resistance
Fan189	30970	China	Double high quality, low oil content
Ding110	20041	China	Double high quality, high tolerance to Sclerotinia sclerotiorum, middle length pod
Ding474	20042	China	Double low quality, high tolerance to Sclerotinia sclerotiorum and virus, lodging resistance
P617	20043	China	Pol CMS restorer, double low quality, long pod
P624	20044	China	Double low quality, high tolerance to Sclerotinia sclerotiorum, susceptible to white rust and resistance to lodging
Yu 178	20045	China	Double low quality, long pod and big seed
03-p74-3	20046	China	Double low quality, earliness
03-p74-4	20047	China	Double low quality, strong growth vigor
03-p74-6	20048	China	Double low quality, very early
03-p74-11	20049	China	Double low quality, lodging resistance and tolerance to Sclerotinia sclerotiorum
04-p34	20050	China	Double low quality, lodging resistance
B. juncea			
Line	Number code	Source	Characteristics
JM16		Australia	Mid maturity, high blackleg resistance, moderate-high oil content, <30 umol/g glucosinolates, <1% erucic acid, moderate yield
JM18		Australia	Mid maturity, high blackleg resistance, moderate-high oil content, <30 umol/g glucosinolates, <1% erucic acid, moderate yield
JN004		Australia	Early-mid maturity, high blackleg resistance, high oil content, <30 umol/g glucosinolates, 0% erucic acid, high yield
JN010		Australia	Early-mid maturity, high blackleg resistance, moderate-high oil content, <30 umol/g glucosinolates, 0% erucic acid, high yield
JN028		Australia	Early-mid maturity, high blackleg resistance, moderate-high oil content, <70 umol/g glucosinolates, <1% erucic acid, high yield
JN031		Australia	Mid maturity, high blackleg resistance, moderate-high oil content, <30 umol/g glucosinolates, 0% erucic acid, moderate yield

JN032		Australia	Mid maturity, high blackleg resistance, moderate-high oil content, <30 umol/g glucosinolates, 0% erucic acid, moderate yield
JN033		Australia	Mid maturity, high blackleg resistance, moderate-high oil content, <30 umol/g glucosinolates, 0% erucic acid, moderate yield
JO006		Australia	Early maturity, high blackleg resistance, moderate-high oil content, <15 umol/g glucosinolates, 0% erucic acid, moderate-high yield
JO009		Australia	Early maturity, high blackleg resistance, moderate-high oil content, <15 umol/g glucosinolates, 0% erucic acid, moderate-high yield
JR042		Australia	Mid maturity, high blackleg resistance, high oil content, <15 umol/g glucosinolates, 0% erucic acid, moderate-high yield
JR049		Australia	Early maturity, high blackleg resistance, high oil content, <15 umol/g glucosinolates, 0% erucic acid, very high yield
Varuna	1	India	Wide adaptation, bold seeded
RH819	2	India	Drought tolerant
RH781	3	India	Frost tolerant
Seeta	4	India	Dwarf plant type
Sej2	5	India	Thermotolerant
RL1359	6	India	Bold seeded
Kranti	7	India	High yielding, medium seed
PCR7	8	India	Drought tolerant
Sanjucta Asech	9	India	Early maturing
RH30	10	India	Non-shattering, bold seeded
Rohini	11	India	Suitable for irrigated conditions
PBR97	12	India	Drought tolerant
RH8812	13	India	Bold seeded, suitable for irrigated conditions
RH8113	14	India	Tolerant to major foliar diseases
PBR91	15	India	White rust tolerant
NDR8501	16	India	Salt tolerant
Vardan	17	India	Suitable for late sowing
Vaibhav	18	India	Suitable for rainfed conditions
RLM619	19	India	Responsive to fertilizer and irrigation
GM1	20	India	Early maturing
RRN-593	21	India	Drought tolerant
Prakash	22	India	High yield, small seeded, tall plant type
CBJ-001		China	Early-mid maturity, high blackleg resistance, moderate-high oil content, <30umol/g glucosinolates, <1% erucic acid, moderate yield
CBJ-002		China	Early-mid maturity, high blackleg resistance, high oil content, <30umol/g glucosinolates, <1% erucic acid, moderate yield
CBJ-003		China	Early-mid maturity, high blackleg resistance, high oil content, <30umol/g glucosinolates, <1% erucic acid, moderate yield
CBJ-004		China	Early-mid maturity, high blackleg resistance, high oil content, <30umol/g glucosinolates, <1% erucic acid, moderate yield

TABP-15		China	Mid maturity, moderate blackleg resistance, high oil content, <30umol/g glucosinolates, <1% erucic acid, moderate yield
MPIR		China	Mid maturity, moderate blackleg resistance, high oil content, <60umol/g glucosinolates, <1% erucic acid, moderate yield
XINYOU4		China	Mid maturity, high blackleg resistance, moderate-high oil content, <1% erucic acid, moderate yield
XINYOU5		China	Early-mid maturity, high blackleg resistance, moderate-high oil content, <1% erucic acid, moderate yield
XINYOU8		China	Mid maturity, high blackleg resistance, moderate-high oil content, <30umol/g glucosinolates, <1% erucic acid, moderate yield
XINYOU9		China	Mid maturity, high blackleg resistance, high oil content, <30umol/g glucosinolates, <1% erucic acid, moderate yield

11.2 Appendix 2: Name and characteristic of Series II germplasm

B. napus		
Line	**Source**	**Characteristics**
06-6-3725	China	Double low quality, strong growth vigor, thick pod shell, shattering resistance
06-6-3737	China	Double low quality, chill resistance
06-6-3771	China	Double low quality, early maturity, thick pod shell, shattering resistance
06-6-3777	China	Double low quality, lodging resistance
06-6-3792	China	Double low quality, moderate lodging resistance
06-P71-1	China	Double low quality, middle/long length pod, lodging resistance
06-P71-2	China	Double low quality, middle/long length pod, lodging resistance
05-P71-11	China	Double low quality
05-P36 R	China	Double low quality, middle/long length pod, lodging resistance
ZY001	China	Low resistance to Sclerotinia sclerotiorum
ZY002	China	Moderate resistance to Sclerotinia sclerotiorum
ZY003	China	Moderate resistance to Sclerotinia sclerotiorum
ZY004	China	Moderate resistance to Sclerotinia sclerotiorum
ZY005	China	Moderate resistance to Sclerotinia sclerotiorum
ZY006	China	High resistance to Sclerotinia sclerotiorum
ZY007	China	High resistance to Sclerotinia sclerotiorum, high lodging resistance
ZY008	China	Resistance/tolerance to Sclerotinia sclerotiorum
ZY009	China	Early maturity; big 1000-grain-weight
ZY010	China	Resistance/tolerance to Sclerotinia sclerotiorum
ZY011	China	Very early maturity; big 1000-grain-weight
ZY012	China	Thick pod shell, shattering resistance
ZY013	China	Dark green leaf
ZY014	China	High oil content

ZY015	China	High oil content
ZY016	China	High oil content
CB Boomer	Australia	TT, large seeds, early vigour, early to mid maturity
CB Tanami	Australia	TT, early vigour, drought tolerance
BLN3224	Australia	Early maturity, high oil, moderate protein, low glucs, glucs, moderately resistant to blackleg
BLN3245	Australia	Early maturity, moderate oil, high protein, low glucs, highly resistant to blackleg
BLN3342	Australia	Early maturity, moderate oil, moderate protein, low glucs, moderately resistant to blackleg
BLN3343	Australia	Early maturity, high oil, high protein, low glucs, moderately resistant to blackleg
BLN3344	Australia	Early maturity, high oil, high protein, low glucs, moderately resistant to blackleg
BLN3345	Australia	Early maturity, very high oil, high protein, low glucs, resistant to blackleg
BLN3346	Australia	Early maturity, very high oil, high protein, low glucs, resistant to blackleg
BLN3575	Australia	Early maturity, very high oil, very high protein, low glucs, resistant to blackleg
BLN3613	Australia	Early maturity, very high oil, very high protein, low glucs, moderately resistant to blackleg
BLN3616	Australia	Early maturity, very high oil, very high protein, low glucs, resistant to blackleg
BLN3189	Australia	Mid maturity, very high oil, very high protein, low glucs, resistant to blackleg
BLN3348	Australia	Mid maturity, high oil, very high protein, low glucs, resistant to blackleg
BLN3350	Australia	Mid maturity, very high oil, very high protein, low glucs, highly resistant to blackleg
BLN3351	Australia	Mid maturity, very high oil, very high protein, low glucs, resistant to blackleg
BLN3352	Australia	Mid maturity, high oil, very high protein, low glucs, resistant to blackleg
BLN3353	Australia	Mid maturity, very high oil, very high protein, low glucs, moderately resistant to blackleg
BLN3354	Australia	Mid maturity, very high oil, very high protein, low glucs, moderately resistant to blackleg
BLN3562	Australia	Mid maturity, very high oil, very high protein, low glucs, highly resistant to blackleg
BLN3579	Australia	Mid maturity, high oil, very high protein, low glucs, resistant to blackleg
BLN3630	Australia	Mid maturity, very high oil, very high protein, low glucs, resistant to blackleg
RT006	Australia	Early maturing, high oil, high blackleg resistance, high yielding line
RT057	Australia	Early maturing, high oil, high blackleg resistance, high yielding line
RT076	Australia	Early maturing, high oil, high blackleg resistance, high yielding line
RT058	Australia	Early maturing, high oil, high blackleg resistance, high yielding line
RT117	Australia	Mid maturing, high oil, high blackleg resistance, high yielding line

RT123	Australia	Mid maturing, high oil, high blackleg resistance, high yielding line
RT096	Australia	Mid maturing, high oil, high blackleg resistance, high yielding line
RT108	Australia	Mid maturing, high oil, high blackleg resistance, high yielding line
RT125	Australia	Mid maturing, high oil, high blackleg resistance, high yielding line
GSC5	India	Double low
TERI(00)R9903	India	Double low
B. juncea		
Line	**Source**	**Characteristics**
Loiret	China	
Ekla	China	
Montara	China	
Berry	China	
RH13	China	
Ringot I	China	
Rk2	China	
AmoraIII	China	
RL	China	
Haoyou11	China	
Tunliuhuangjie	China	
Datonghuangyoucai	China	
Qianxianjiecai	China	
Yilihuang	China	
Hetianyoucai	China	
Jinshahuang	China	
Manasihuang	China	
Brassica juncea 1	China	
Brassica juncea 2	China	
Brassica juncea 3	China	
JM06001	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06002	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06003	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06004	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06006	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06009	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06010	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06011	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06012	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06013	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06014	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06015	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06018	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06019	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06020	Australia	Low gluc, good yield and agronomics, mod - high oil
JM06021	Australia	Low gluc, good yield and agronomics, mod - high oil

JM06026	Australia	Low gluc, good yield and agronomics, mod - high oil
Ashirwad	India	High erucic acid and high glucosinolate content, for late sown conditions
Aravali	India	For rainfed conditions
Basanti	India	Yellow seeded
CS-52	India	For salt affected soils
CS-54	India	For salt affected soils
GM-2	India	Bold seeded
Geeta	India	Bold seeded, high seeds/siliqua and tetralocular pods, for rainfed conditions
GM-3	India	Medium seeded
Jagannath	India	For irrigated areas
JM-1	India	White rust resistant
JM-2	India	White rust resistant
JM-3	India	White rust resistant
Laxmi	India	Bold seeded
Maya	India	For irrigated areas
Narendra Ageti Rai–4	India	Early maturity
Narendra Swarna Rai-8 (NDYR-8)	India	Yellow seeded, high oil content
Pusa Mahak	India	Early maturity
RGN-13	India	High temperature tolerance
Swarna Jyoti	India	For late sown conditions
Vasundhra	India	For irrigated areas
Kanti	India	Early maturity
Urvashi	India	High temperature tolerance
Bio-902	India	Bold seeded

11.3 Appendix 3: Sclerotinia screening results for *B. napus*

Summary of Sclerotinia screening results for *B. napus* in each country[a]

Institute	Type of Sclerotinia test	Assessment method	Most resistant lines in each screening experiment*
IOCR	Disease nursery	% plants with lesions	03-p74-3, Zhongshu-ang No. 9 (30920), ZY002, 04-p34, Fan168 Zhongyou 821, Zhongshu-ang No. 9 (30920), RT108, Fan023, BLN3613 Zhongshu-ang No. 9 (30920), P624, 03-p74-3, 03-p74-4, Yu 178
UWA-B	Stem inoculation at flowering stage	Stem lesion length (& % plants dead)	Fan168, RR002, Ag-Spectrum, Oscar, Lantern ZY006, 06-6-3792, RT108, ZY004, RT057 Oscar, Zhongyou-za No.8, Fan168, 06-6-3792, Ding110
School of Botany, Univ Melb	Detached leaf test with agar plug Stem inoculation with infested barley grain	Leaf lesion size Stem lesion length	Limited variation seen between lines Ag-Outback, Rivette, RR002, RQ011, RT004
AAFC, Canada	Stem inoculation with agar plugs	Stem lesion length	Rivette, Ag-Outback, RQ011, Zhongshu-ang No. 9 (30920), P624
PAU	Stem inoculation	Stem lesion length	BLN3345, 06-6-3777, ZY002, ZY014, ZY011
NRCRM	Artificial inoculation of plots.	Disease incidence before harvest	Monty, RQ011, RR002 Fan028, Ding474
HAU	Soil inoculation at seedling stage. Stem inoculation at 50% flowering	Disease incidence Stem lesion length	Trigold, RQ11, Charlton, Ag-Spectrum, RR005 Ag-Outback, Rainbow, RQ011, RQ001-02M2, Neelam

[a]*Results from each screening experiments at each institute are separated by a blank line.*

11.4 Appendix 4: Thermotolerance of *B. juncea*

Best performing *B. juncea* lines for thermotolerance in India

Institute	Series	Thermotolerance screening stage	Thermotolerance screening method	Best lines
NRCRM	I (Australian, Indian) 2006/07	Seedling stage	Germination	Prakash, Kranti, Durgamani, RH8812, NDR8501
			Mortality	RH819, Prakash, Sej2, PCR7, Varuna
NRCRM	I (Australian, Indian) 2007/08	Seedling stage	Rate of imbibitions, amylase activity, vigour	Urvashi, CS52, Prakash, Basanti, Varuna
			Mortality	Urvashi, CS52, Prakash, Basanti, Varuna, RH819
			Germination	RH781, Varuna, Laxmi, Kranti, Vaibhav, Prakash, Vardan
NRCRM	I, II (Australian, Indian, Chinese[a]) 2008/09	Seedling stage	Mortality at 45ºC (lab)	RGN 13, JN031 JM016, JN032, JR042,
			Emergence (field)	JM3, JN032, JN004, Aravali, JR042
			Mortality (field)	RGN 13, Rohini, Aravali, CS52, NDYR 8, Varuna
		Terminal stage	Siliqua length	JM06018, JM2, GM3, JM06002, JM1
			Seeds/siliqua	PCR 7, JM06015, JN004, Bio902, JM06010
			Seed weight	JM06018, Bio 902, JR049, JM2, Maya
			Oil content	JM06015, JM06018, NDYR8, JM06006, CS54, NDR8501
HAU	I (Australian, Chinese, Indian) II (Australian, Chinese)	Seedling stage	Mortality at 47.5ºC	RH819, PCR7, RH30, JR049, PBR97, JN010, Vaibhav, PBR91
			Mortality at 47.5ºC	Tunliuhuangjie, Yilihuang, Hetianyoucai
PAU	II (Australian, Chinese, Indian) 2007/08 and 2008/09	Seedling stage	Mortality (lab)	Loiret, Jagannath, ZY005
		Seedling stage	Mortality (field)	JM06012, Loiret, GM-2, RH-9304, Jagannath
		Terminal stage	Number of pods	B. juncea 1, Qianxianjiecai JM06013, Kranti , JM06006, JM06009
			Pod length	RK2, AmoraIII, Haoyou11, Kranti
			Seed size	JM06003, JM06006, JM06011, JM0621, Laxmi, Kranti

[a]The Chinese lines were extremely late and seed set was very poor hence data pertaining to their response to high temperature during seed development could not be obtained.

11.5 Appendix 5: Thermotolerance of *B. napus*

Best performing *B. napus* lines for thermotolerance in India

Institute	Series	Thermotolerance screening stage	Thermotolerance screening method	Best lines
HAU	I (Australian)	Seedling stage	Mortality at 47.5ºC	RR001, Mystic, Lantern, Rainbow
HAU	I (Australian) & II (Australian, Chinese)	Seedling stage	Mortality at 47.5ºC	Fan189 (All *B. napus* were less tolerant than *B. juncea)*
PAU	II (Australian, Chinese, Indian) 2007/08 and 2008/09	Seedling stage	Mortality at 45ºC	BLN3348,Tanami, BLN3579
		Seedling stage	Mortality (field)	GSC5, Tanami, BLN3348, ZY006, 06-06-3777
		Terminal stage	Number of pods	CBBoomer, BLN3224 05-P-71-11, Tanami, BLN3579, BLN3348, BLN3354, ZY008, BLN3350
			Pod length	SARDI607, 06-06-3737, 05-P-71-11, ZY008, ZY003
			Seed size	RT117, BLN 3245, BLN 3343, GSC5, BLN 3344, RT006

11.6 Appendix 6: Drought tolerance of *B. juncea*

Best performing *B. juncea* lines for different characters under drought conditions in India

Institute	Series	Characters	Best lines
NRCRM	I (Australian, Indian) 2005/06	Specific leaf area (cm^2/g)	PCR7, Kranti, Seeta, JM018, NDR8501
		Transpiration ($mmoles/m^2/s$)	RH819, PCR7, Varuna, JO009, JM018
		Leaf Ψw (bar)	RH819, Varuna, Kranti, RH-781, PCR7
NRCRM	I (Australian, Indian) 2006/07	Specific leaf area (cm^2/g)	PCR7, NDR8501, RH30, Varuna, Vaibhav
		Transpiration ($mmoles/m^2/s$)	JR042, JO006, JM018, JO009
NRCRM	I and II (Indian, Chinese) 2007/08	Water use efficiency at 50% flowering	Rohini, Kranti, JM3, Maya, PCR7, Swarna Jyoti, Bio902, NDYR8, JM2, Geeta, Aravali
		Water use efficiency at full flowering	Ekla, Datonghuangyoucai, Qianxianjiecai, NDYR8, JM3
PAU	I (Australian, Indian)	Seedling stage	RH8113, JN028, JN010, Sanjucta Asech, Sej2
		Terminal stage	Sanjucta Asech, Sej2, Varuna, JR049
HAU	I (Australian)	Terminal stage	JN004, JR042, JR049

11.7 Appendix 7: Drought tolerance of *B. napus*

Best performing *B. napus* lines for drought tolerance

Institute	Series	Drought tolerance screening method	Best lines
PAU	I (Australian, Indian)	Seedling stage	RQ001-02M, Charlton, BST7-02M2
PAU	I Australian, Indian)	Terminal stage	Skipton, Monty, GSL-1, Tranby
HAU	I Australian, Indian)	Terminal stage	Ag-Outback, Monty, Charlton and Ag-Spectrum

11.8 Appendix 8: Drought tolerance of *B. napus* (additional Australian lines)

Additional Australian *B. napus* lines provided for terminal drought screening in India

Genotypes	Deviation between no irrigation (I0) vs one irrigation (I1) or two irrigations (I2)											
	Leaf Area (cm^2)		Leaf Ratio		Fv/Fm		Root Area (mm^2)		Seed wt. (g)		Seed Yield (g/plant)	
	I1	I2	I1	I2	I1	I2	I1	I2	I1	I2	I1	I2
RT117	-0.07	-0.20	-0.35	-0.52	0.09	0.13	-0.06	-0.14	0.06	0.00	-0.40	-1.71
AG-MUSTER	-0.66	-1.29	-0.95	-1.43	0.04	0.08	-0.36	-1.66	0.06	-0.37	-1.17	-1.21
ATR-BARRA	-1.07	-1.48	-1.13	-1.22	0.08	0.10	-0.21	-0.68	0.13	0.49	-0.51	-3.53
ATR-SUMMIT	-1.02	-1.75	0.57	0.40	0.10	0.12	-0.34	-0.54	0.07	0.27	-1.40	-2.82
AV-GARNET	-0.13	-0.40	-0.54	-0.56	0.04	0.11	-0.34	-0.89	0.17	0.44	-0.80	-2.36
AV-OPAL	-0.82	-1.20	-0.46	-0.67	0.04	0.07	-0.13	-0.18	0.18	0.04	-0.23	-2.86
AV-SAPPHIRE	-0.90	-0.95	-1.35	-1.58	0.01	0.02	-0.30	-0.51	0.04	0.24	-1.66	-2.58
BANJOTT	-1.06	-1.41	-1.78	-2.04	-0.01	0.18	-0.51	-0.51	0.03	-0.50	-0.33	-1.27
CC05032	-0.31	-0.93	-0.68	-0.81	0.03	0.11	-0.49	-1.70	0.03	0.67	-2.25	-7.24
CHARLTON	-1.27	-1.72	-2.69	-2.86	0.06	0.12	-0.07	-0.28	0.04	-0.66	-0.56	-1.43
EC609308	-0.26	-0.63	-0.49	-0.57	0.03	0.03	-0.14	-0.34	0.10	-0.32	-1.06	-2.33
GSC6	-0.88	-2.39	-0.37	-0.58	0.09	0.13	-0.23	-0.53	0.14	0.09	-0.48	-0.83
HYOLA50	-0.39	-0.95	-0.09	-0.83	0.03	0.12	-0.31	-0.34	0.12	0.37	-0.99	-2.78
HYOLA75 (1)	-1.03	-1.45	-0.73	-0.97	0.02	0.06	-0.28	-0.54	0.18	0.24	-0.99	-1.51
HYOLA75 (2)	-0.47	-0.68	-1.51	-2.41	0.05	0.02	-0.32	-0.33	0.04	-0.90	-1.27	-1.39
KAROO	-0.67	-1.46	-0.82	-1.39	0.06	0.19	-0.08	-0.24	0.15	0.49	-0.66	-2.06
MONTY	-0.12	-0.38	-1.44	-1.78	0.01	0.03	-0.38	-0.47	0.10	0.08	-0.67	-2.32
RIVETTE	-0.46	-0.56	-0.56	-1.31	0.07	0.08	-0.26	-0.54	0.07	0.35	-1.38	-2.44
RT057 (1)	-0.90	-1.34	-2.98	-3.50	0.01	0.06	-0.15	-0.73	0.03	-0.13	-0.56	-2.79
RT057 (2)	-1.09	-1.17	-2.16	-2.97	0.01	0.15	-0.25	-0.64	0.06	0.06	-2.51	-4.20
RT123	-0.96	-1.48	-3.83	-4.83	-0.18	-0.07	-0.52	-1.19	0.06	0.23	-1.49	-5.40
RUBY	-0.07	-0.20	-0.65	-1.20	0.02	0.09	-0.17	-0.43	0.07	-0.96	-1.24	-0.97
SARDI607	0.03	-0.24	-1.81	-2.19	0.06	0.04	-0.65	-0.88	0.05	0.14	-3.01	-3.33
TARCOOLA (1)	-0.30	-0.68	-0.75	-1.30	0.00	0.14	-0.39	-0.56	0.03	-0.44	-0.58	-2.04
TARCOOLA (2)	-0.24	-0.77	-0.44	-1.13	0.00	0.02	-0.42	-0.71	0.14	-0.14	-0.98	-1.42
TRANBY	-0.53	-1.39	-0.42	-0.69	0.02	0.01	-0.36	-0.42	0.11	0.13	-0.41	-2.41
44C73	-0.45	-1.41	-1.25	-1.12	-0.07	-0.07	-0.22	0.39	0.05	-0.16	-1.02	-1.56
46C76	-0.43	-0.49	-0.28	-0.61	-0.06	0.09	-0.07	-0.26	0.12	0.71	-3.99	-10.37

11.9 Appendix 9: Shatter screening 2006/07

Evaluation of *B. napus* crosses F1/F2 for shattering during 2006/07, TERI

Line	Plant type (F/M/I)[a]	50% flowering	Maturity	Shattering (Visual)[b]	Shattering (% shattering)[c]
Monty		56	144	3	51
Monty x R9903	F	51	145	6	30
Monty x GS17	F	51	145	1	72
Monty x EM05	F	63	145	3	56
RR005		56	144	3	52
RR005 x R9903	F	56	144	4	49
RR005x GS17	F	73	144	7	16
RR005x EM05	F	73	144	7	14
Trigold		62	146	7	20
Trigold x R9903	F	62	146	6	30
Trigold x GS17	F	54	146	7	20
Trigold x EM05	M	70	146	6	26
Tranby		61	147	6	28
Tranby x R9903	**F**	**61**	**147**	**5**	**37**
Tranby x GS17	F	54	147	5	32
Tranby x EM05	F	68	147	6	29
GM05		52	147	6	27
GM05 x Monty	F	52	147	6	29
GM05 x RR005	F	67	147	5	33
GM05 x Trigold	F	67	147	5	34
GM05 x Tranby	F	72	147	5	32
R9903		56	148	6	26
R9903 x Monty	F	51	148	5	31
R9903 x RR005	**F**	**51**	**148**	**6**	**29**
R9903 x Trigold	F	51	148	6	29
R9903 x Tranby	F	51	148	6	21
GS17		51	142	7	15
GS17 x Monty	F	51	142	6	23
GS17 x RR005	F	52	142	7	20
GS17 x Trigold	F	52	142	6	30
GS17 x Tranby	F	51	142	6	30
Surpass x R9903 (F_2)	**F**	**49**	**143**	**7**	**15**
Trilogy x R9903 (F_2)	**F**	**57**	**143**	**7**	**15**
Average		58	145	6	30

[a] *More resemblance to Female (F) / Male (M) / Intermediate (I)*

[b] *Visual observation:1-high shattering, 9-low shattering*

[c] *% shattering = (no. of pods shattered/ total no. of pods on mainshoot) x 100*

11.10 Appendix 10: Shatter screening 2007/08

Evaluation of F_2/ F_3/ BC_1/ BC_2 *B. napus* crosses for agronomy and shattering during 2007/08, TERI

Line	Plant type	50% flowering	Days to Maturity	Shattering	
				Visual obs.	% shattering
Monty		52	131		
Monty x GS17(BC1)	I	52	131	7	40
X GS17(F2)	I	52	131	7	46
x R9903(BC1)	F	52	131	8	44
x R9903 (F2)	F	52	136	8	47
x EM05 (BC1)	F	52	136	8	50
x EM05 (F2))	F	54	136	8	39
RR005		71	137	8	45
RR005 x R9903(BC1)	F	69	137	7	33
x R9903(F2)	F	69	138	8	21
x GS17(BC1)	F	79	138	8	34
x GS17(F2)	F	79	138	8	52
x EM05 (BC1)	I	78	138	8	45
x EM05 (F2)	I	78	142	8	38
Trigold		69	140	7	33
Trigold x R9903(BC1)	F	69	140	8	50
x R9903(F2)	F	69	140	7	37
x GS17(BC1)	I	67	140	7	46
x GS17(F2)	I	67	140	8	44
x EM05 (BC1)	I	67	140	8	50
x EM05 (F2)	I	67	140	7	31
Tranby		65	138	9	45
Tranby x R9903(BC1)	**F**	**65**	**138**	**7**	**48**
x R9903(F2)	**I**	**68**	**138**	**7**	**50**
x GS17(BC1)	I	58	138	8	47
x GS17(F2)	I	58	138	8	20
x EM05 (BC1)	I	58	138	8	35
x EM05 (F2)	I	58	138	8	38
EM05		56	141	8	42
EM05 x Monty (BC1)	I	56	141	7	48
x Monty (F2)	I	56	141	8	45
x RR005(BC1)	I	56	141	8	54
x RR005(F2)	I	72	143	8	52
x Trigold (BC1)	I	72	143	8	37
X Trigold (F2)	I	54	143	8	48
x Tranby (BC1)	I	54	143	7	47
x Tranby (F2)	I	72	143	7	45
R9903		72	151	8	43

R9903 x Monty(BC1)	I	52	151	7	40
x Monty (F2)	I	52	151	7	30
x RR005(BC1)	**I**	**52**	**151**	**7**	**43**
x RR005(F2)	**I**	**52**	**151**	**7**	**44**
x Trigold (BC1)	I	52	151	7	59
X Trigold (F2)	I	52	151	7	42
x Tranby (BC1)	I	52	151	7	37
x Tranby (F2)	I	52	151	7	33
GS-17		48	148	8	30
GS-17 x Monty(BC1)	I	48	148	8	51
x Monty (F2)	I	48	148	8	48
x RR005(BC1)	I	48	148	8	44
x RR005(F2)	I	48	148	8	28
x Trigold (BC1)	I	48	148	8	40
x Trigold (F2)	I	48	148	7	40
x Tranby (BC1)	I	48	148	7	67
x Tranby (F2)	I	48	148	7	42
Surpass x R9903 (BC_2)	**I**	**52**	**152**	**7**	**36**
x R9903(F_3)	**I**	**52**	**152**	**8**	**46**
Trilogy x R9903 (BC_2)	**I**	**52**	**152**	**8**	**49**
x R9903 (F_3)	**I**	**52**	**152**	**8**	**49**
Average		59	143	8	43

11.11 Appendix 11: Shatter screening 2008/09

Evaluation of BC_2 and BC_3 *B. napus* breeding populations during 2008/09, TERI

Line[a]	Emer-gence	Seedling vigour	Days to 50% Flowering	Days to Maturity	Shattering Visual Obs at harvesting	Shattering % 4 weeks post maturity
Monty x GS17	8	8	57	138	7	20
Monty x R9903	8	8	54	138	7	20
Monty x EM05	8	8	58	138	8	11
RR005 x R9903	8	8	50	138	8	18
RR005 x GS17	7	8	50	138	7	18
RR005 x EM05	8	7	73	143	6	21
Trigold x R9903	9	9	50	143	8	21
Trigold x GS17	8	8	50	143	7	22
Trigold x EM05	8	8	51	143	7	17
Tranby x R9903	**8**	**8**	**64**	**143**	**8**	**14**
Tranby x 6S17	7	7	73	143	6	31
Tranby x EM05	7	7	63	139	8	25
EM05 x Monty	8	8	59	139	7	8

EM05 x RR005	9	9	61	139	8	12
EM05 x Trigold	7	8	59	139	8	19
EM05 x Tranby	8	8	59	138	8	15
R9903 x Monty	8	7	50	138	8	10
R9903x RR005	**8**	**8**	**50**	**139**	**8**	**7**
R9903 x Trigold	8	7	50	139	8	15
R9903 x Tranby	8	8	48	139	8	18
GS17 x Monty	7	8	48	140	8	22
GS17 x RR005	7	7	51	140	8	11
GS17 x Trigold	8	8	63	142	8	16
GS17 x Tranby	8	7	75	142	8	15
Surpass x R9903	**9**	**8**	**49**	**142**	**8**	**8**
Trilogy x R9903	**8**	**7**	**77**	**142**	**8**	**24**
Average	8	8	57	140	8	17

[a]*R9903 = TERI(00)R9903; GS17 = TERI(00)GS17; EM05 = TERI(00)EM05*

[b]*Breeding populations for shatter resistance to be exchanged.*

11.12 Appendix 12: Yield of *B. napus*

Yield of *B. napus* lines (t/ha) averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
Aus	I	0.3-1.0 (ave 0.6)	Zhongshu-ang No.9, Fan168	0.8-1.6 (ave 1.2)	Ag-Spectrum, RR005	0.4-0.7 (ave 0.6)	
China	I		Yu178 Zhongyou-za No.8		Mystic, RR0001, RQ001-02M2, Oscar, RQ011, Lantern		GSL2 Neelam
India	I	1.3-3.0 (ave 2.3)	Ding474, 30920	1.2-2.1 (ave 1.70)	Ag-Spectrum,	1.7-2.0 (ave 1.9)	GSL2
Aus	II	0.5-1.2 (ave 0.8)	06-6-3792, 05-P71-11	0.9-1.4 (ave 1.1)	RT125, RT117	0.8-0.9 (ave 0.8)	
China	II	1.3-2.9 (ave 2.1)	ZY002	1.0-2.4 (ave 1.6)	BLN3245, BLN3630	0.7-1.4 (ave 1.0)	GSC5
India	II	0.8-2.5 (ave 1.5)	06-6-3771, 06-6-3792	1.3-2.7 (ave 2.0)	BLN3354, BLN3575, BLN3630	1.4-1.6 (ave 1.5)	GSC5

11.13 Appendix 13: Oil content of *B. napus*

Oil content of *B. napus* lines averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
Aus	I	37-41 (ave 39)	03-p74-3 P3083	38-44 (ave 41)	Purler RQ001-02M2 Surpass400	39-40 (ave 39)	GSL1
China	I	35-44 (ave 40)	P617	36-48 (ave 41)	Av-Sapphire	40-44 (ave 41)	Neelam
India	I	37-43 (ave 40)	03-p74-3, P624	39-43 (ave 41)	RR002	39-41 (ave 40)	GSL1
Aus	II	37-42 (ave 39)	ZY002, 05-P71-11	38-42 (ave 41)	BLN3350, BLN3189, BLN3562	38-41 (ave 39)	TERI (00)R9903
China	II	41-52 (ave 47)	ZY014	40-49 (ave 45)	RT123, RT076, BLN3630	41-44 (ave 43)	GSC5
India	II	34-42 (ave 37)	ZY007	35-47 (ave 42)	RT108	-	-

11.14 Appendix 14: Glucosinolate content of *B. napus*

Glucosinolate content of *B. napus* lines averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
Aus	I	9-38 (ave 13)	P624	5-11 (ave 8)	RR002	48-52 (ave 49)	-
China	I	16-69 (ave 28)	Yu178, P3083, Zhongyou-za No.8, P624	19-32 (ave 26)	Lantern, Rivette, Oscar	105-123 (ave 111)	-
India	I	-	-	14-39 (ave 21)	RR013	-	-
Aus	II	5-15 (ave 10)	ZY003	6-16 (ave 9)	BLN3348	21-23 (ave 22)	-
China	II	16-50 (ave 25)	ZY014	18-32 (ave 26)	BLN3353	42-55 (ave 49)	-
India	II	14-23 (ave 13)	06-6-3725, ZY006	7-38 (ave 19)	BLN3343, BLN3189, RT076	-	-

11.15 Appendix 15: Fatty acid composition of *B. napus*

Fatty acid composition of series II *B. napus* lines (range and averages)

Country	Series	Erucic acid	Oleic acid	Polyunsat (ave)	Monounsat (ave)	Saturated (ave)
Aus[a]	II	0.1-0.3 (ave 0.1)	60-71 (ave 64)	27	65	7.3
China[a]	II	0.1-24.1 (ave 2.3)	28-65 (ave 60)	28	65	7.4
India[a]	II	1.7-4.6 (ave 3.2)	53-60 (ave 57)	27	65	7.6

[a] *Testing in Australia (2008/09)*

11.16 Appendix 16: Seed weight of *B. napus*

1000 seed weight of *B. napus* lines averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
India	I	2.3-3.4 (ave 2.9)	03-p74-4	2.2-2.7 (ave 2.5)	Purler	3.0-3.2 (ave 3.0)	Neelam
Aus	II	3.2-5.3 (ave 4.4)	ZY015, 05-P36R	3.0-4.7 (ave 3.7)	CBBoomer	-	
India	II	1.9-3.7 (ave 2.6)	ZY002	1.9-3.6 (ave 2.5)	BLN3245	3.0-3.3 (ave 3.1)	GSC5

11.17 Appendix 17: Yield of *B. juncea*

Yield (t/ha) of *B. juncea* lines averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
Aus	I	0.4-0.5 (ave 0.4)	CBJ-001	0.5-0.8 (ave 0.7)	JN28, JO009	0.2-0.8 (ave 0.6)	RH819
China*	I		CBJ-004, Xinyou4		JM16, JO006, JN31		Vaibhav, Prakesh, GM1
India	I	1.6-2.7 (ave 2.2)	Xinyou4, MPIR	0.3-1.1 (ave 0.8)	JO006, JR049, JN28	0.6-1.7 (ave 1.2)	RL1359, RH30, RH819
Aus	II	0.0-0.7 (ave 0.4)	B. juncea 2, Jinshahuang	0.5-0.8 (ave 0.7)	JM06011, JM06009	0.4-0.9 (ave 0.7)	Vasundhra Urvashi
China*	II		Ekla, Manasihuang		JM06012, JM06013	-	-
India	II	1.0-1.5 (ave 1.2)	Datonghuangyoucai	2.0-3.1 (ave 2.5)	JM06003	1.8-2.7 (ave 2.2)	JM-1

**XAAS*

11.18 Appendix 18: Days to 50% flowering for *B. juncea*

Days to 50% flowering of *B. juncea* lines averaged over two seasons in each country

Country	Series	Chinese lines	Australian lines	Indian lines
		Range	Range	Range
Aus	I	121-139 (ave 129)	103-126 (ave 112)	66-107 (ave 101)
China	I	(ave 55)	(ave 54)	(ave 53)
India	I	54-99 (ave 70)	58-96 (ave 72)	54-76 (ave 65)
Aus	II	106-117 (ave 112)	84-100 (ave 94)	90-97 (ave 93)
China	II	(ave 57)	(ave 54)	-
India	II	93-112 (ave 103)	53-60 (ave 56)	42-67 (ave 56)

11.19 Appendix 19: Oil content of *B. juncea*

Oil content of *B. juncea* lines averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
Aus	I	34-39 (ave 36)	TABP-15 Xinyou4	37-39 (ave 38)	JN033	32-35 (ave 34)	
China	I	25-31 (ave 29)	CBJ004, Xinyou9	30-35 (ave 32)	JN033, JM18, JM16	26-30 (ave 28)	Seeta, PBR97
India	I	39-40 (ave 40)	TABP-15, Xinyou4	36-40 (ave 38)	JR049	37-40 (ave 39)	Vardan, RLM619

Aus	II	38-43 (ave 40)	Jinshahuang Yilihuang	36-39 (ave 37)	JM06015	33-37 (ave 35)	
China	II	21-34 (ave 28)	Hetianyoucai	26-31 (ave 30)	JM06013, JM06001, JM06015	-	
India	II	33-42 (ave 38)	B. juncea 2	36-45 (ave 39)	JM06004	37-41 (ave 39)	GM-2

11.20 Appendix 20: Seed weight of *B. juncea*

1000 Seed weight (g) of *B. juncea* lines averaged over two seasons in each country

Country	Series	Chinese lines		Australian lines		Indian lines	
		Range	Best lines	Range	Best lines	Range	Best lines
India	I	3.0-4.3 (ave 3.5)	CBJ001	2.1-2.7 (ave 2.4)	JN010 JO006	2.5-4.5 (ave 3.6)	PCR7 RH819 Varuna
Aus	II	2.1-2.8	Tunliuhuangjie Montara	1.8-2.7	JM06011	2.0-3.9	Urvashi
India	II	2.1-3.5 (ave 2.8)	Loiret, Montara, Haoyou11, Tunliuhuangjie	2.7-4.7 (ave 3.6)	JM06009, JM06012, JM06011	4.0-6.6 (ave 5.2)	Jagannath Laxmi

11.21 Appendix 21: Blackleg survival of *B. juncea*

Blackleg survival percentages of series I and II *B. juncea* lines in Australia

Institute	Series	Chinese lines		Australian lines		Indian lines	
		Range (%)	Range (%)	Best lines	Best lines	Range (%)	Best lines
NSW (2005)	I	-	-	30-74	JM18, JN032	16-77	Rohini
SA (2005)	I	-	-	56-87	JN031, JO006	29-84	Rohini
Vic (2005)	I	51-99	Xinyou5, MPIR, Xinyou4	63-99	JR042	4-91	RH781
WA (2005)	I	-	-	42-104	JN033, JM18	-	-
SA (2007)	II	14-80	Berry	46-87	JM06026, JM06002	52-82	GM3
SA (2008)	II	74-100	B. juncea 1	80-100	JM06011	70-97	Swarna Jyoti, GM-2
Vic (2007)	II	11-72	Hetianyoucai	44-76	JM06009, JM06013	34-95	Urvashi
Vic (2008)	II	18-86	B. juncea 2	17-86	JM06013	24-89	Urvashi
WA (2007)	II	-	-	32-100	JM06009, JM06001	21-38	GM1, GM2
WA (2008)	II	22-100	AmoraII, Yilihuang	0-93	JM06014	8-79	Laxmi

11.22 Appendix 22: Protocols for assessing individual agronomic traits

1 Emergence

Assessed as a visual rating on a 1 to 9 scale (1 poor emergence – 0-5 plants per m^2, 9 excellent emergence – 45-50 plants per m^2). The plots should be assessed for emergence at the 3-4 leaf stage. Plots that have patchy establishment and holes in only a part of the plot should be noted. The size of holes/patches should be noted in the field book and yield adjustments made after harvest.

2 Early vigour

Assessed as a visual rating on a 1 to 9 scale (1 poor vigour, 9 high vigour). This is a rating for plant growth at the cabbage stage of development. This rating is to give an indication of biomass production at the start of the season.

3 Start of flowering date

Assessed as 50% of plants with one or more flowers. Record the Julian calender date in the field book (i.e. 1st of January = 1, 2nd of January = 2 and so on to 365/366).

4 End of flowering date

Assessed as 90-95% of plants with no flowers left on them.

5 Plant height

Measured as the average height (cm) of plants in each plot at maturity. Presented as a mean height. Height measurement should be recorded when the plants have reached their maximum height. Height measurement should reflect majority of plot, not a small number of taller individuals.

6 Maturity

Assessed as the time when 40% – 60% of seeds are changing colour.

7 Shattering resistance

Assessed as a visual rating on a 1 to 9 scale (1 high shattering, 9 low shattering), measured prior to harvest. Plots of different maturity shall not be compared. Can only be measured on plots left standing.

8 Lodging resistance

Assessed as a visual rating on a 1 to 9 scale (1 on the ground, 9 no lodging). Each score represents approximately 10 degrees of lean.

9 Visual Disease

Assessed as a visual rating on a 1 to 9 scale (this may need to be modified for each different disease).

10 Seed size

Assessed as weight (grams) of 1,000 grains. Comparisons must be made with controls of similar maturity.

11 Plot yield

Measured as, or converted to, tonnes/ha (to 2 decimal places).

11.23 Appendix 23: Sclerotinia information package

11.24 Appendix 24: White rust information package