

IDENTIFICATION OF ANTHRACNOSE RESISTANCE IN YELLOW LUPINS AND ITS INCORPORATION INTO BREEDING LINES

Kedar Adhikari, Geoff Thomas, Bevan Buirchell and Mark Sweetingham

Department of Agriculture and Food, 3 Baron-Hay Court, South Perth WA 6151, Australia
Centre for Legumes in Mediterranean Agriculture, The University of Western Australia, Crawley WA 6009, Australia

Corresponding author's email: kadhikari@agric.wa.gov.au

ABSTRACT

Lupin Anthracnose (caused by *Colletotrichum lupini*) first occurred in commercial crops in Western Australia in 1996 and severely affected the lupin industry, particularly in the northern grainbelt of Western Australia. Subsequent studies led to the identification of good sources of resistance to the disease in *Lupinus angustifolius* and *Lupinus albus*. However, no reliable source of resistance was obtained in *Lupinus luteus* until 2002. Screening of accessions from USDA and Russian Lupin Research Institute, Bryansk, Russia yielded two lines with some resistance to Anthracnose. At the same time a group of sister lines in the West Australian lupin breeding program derived from a Hungarian parent P20856 also showed some resistance, giving three putative sources of resistance. Crosses were made between the resistant lines and other breeding lines. Consequently several breeding lines have been developed with a moderate level of resistance to the disease. This is the first report on development of Anthracnose resistance in *Lupinus luteus*. This level of resistance should provide enough protection to grow *Lupinus luteus* in southwest of Western Australia as this region does not have *Lupinus cosentinii*, a perpetual natural host of Anthracnose pathogen.

KEYWORDS

Colletotrichum lupini, *Lupinus luteus*, yellow lupin, Anthracnose resistance, breeding

INTRODUCTION

Yellow lupin (*Lupinus luteus*) has high demand as an excellent feed ingredient for aquaculture, poultry and pig industries due to the grain having higher protein content (38–40%) and higher sulphur amino acid content than narrow-leaved lupins. It is widely cultivated on acid sandy soils in northern Europe where it has been bred by regional programs in Germany, Poland, Belarus, Russia and Ukraine. Since the widespread prevalence of Anthracnose and unavailability of resistant cultivars, the area under yellow lupin has decreased significantly in Europe in favour of *Lupinus angustifolius*.

A renewed interest in yellow lupins developed in the 1990s in Western Australia (WA) after realizing that the crop was very resistant to *Pleiochaeta* root rot and brown spot, tolerant to aluminium toxicity and had a highly efficient mechanism of phosphorus uptake (French, 1999; French *et al.* 2001). Subsequent research led to the development of Wodjil and Pootallong cultivars for WA. However, the area under yellow lupin is negligible at present because of its lower yield, susceptibility to Anthracnose and aphid feeding damage. The crop has potential to be grown successfully in higher rainfall regions of southwest WA and Anthracnose resistance is a must if it is to be established as a crop in this area.

Lupin Anthracnose, caused by *Colletotrichum lupini*, is a devastating disease infecting all species of lupins. It is prevalent in many parts of the world where lupin is grown commercially except in the eastern States of Australia. Within WA it is an ongoing threat to lupin production particularly in the northern agricultural regions where blue lupins (*Lupinus cosentinii*) are a perpetual host of the pathogen (Thomas, 2003). The problem is less common in the southwest region of WA because of the lack of blue lupins. However, the pathogen is favoured by higher rainfall environments, therefore potential for significant yield loss exists in southwest and southern coastal regions as they are higher rainfall environments. Yield losses of up to 75% have been recorded in Anthracnose infected Wodjil lupins.

The disease can be controlled by using clean seed, seed treatment or repeated application of fungicides, but the most economical way of controlling the disease is through the use of resistant cultivars. Very limited knowledge is available on the occurrence of resistance in yellow lupins and the host-pathogen relationship.

This study was conducted to identify resistance to Anthracnose in *Lupinus luteus* and incorporate resistance into the breeding program for development of disease resistant germplasm. The paper will present results of disease screening trials, discuss the level of resistance from three different sources and their usefulness to the breeding program.

Table 1. Anthracnose reaction of *Lupinus luteus* lines derived from various crosses along with parental lines and controls when tested at Medina disease nursery in 2006 and 2007. The disease reactions are in 1-5 scale, where 1 is very resistant and 5 is extremely susceptible.

Lines	Pedigrees	2006	2007
01D055-36	98D013-2/P28716	3.0	3.3
01D055-4	98D013-2/P28716	3.0	3.4
01D055-43	98D013-2/P28716	3.1	3.1
01D055-34	98D013-2/P28716	3.1	3.2
01D055-57	98D013-2/P28716	3.1	3.4
01D055-1	98D013-2/P28716	3.1	3.4
01D055-48	98D013-2/P28716	3.1	3.5
01D056-13	96D001-18-12/P28716	3.1	3.7
01D055-22	98D013-2/P28716	3.3	3.3
01D055-61	98D013-2/P28716	3.3	3.3
01D055-13	98D013-2/P28716	3.3	3.4
01D055-73	98D013-2/P28716	3.3	3.5
01D055-16	98D013-2/P28716	3.4	3.5
01D055-15	98D013-2/P28716	3.5	3.5
01D055-52	98D013-2/P28716	3.5	3.6
01D055-45	98D013-2/P28716	3.5	3.8
01D056-6	96D001-18-12/P28716	3.6	3.8
01D055-9	98D013-2/P28716	3.6	3.9
97D001-11-3	P20855/Wodjil	3.5	3.8
97D002-22-3	P20856/Wodjil	3.6	3.8
97D002-26-16	P20856/Wodjil	3.5	3.8
97D002-26-25	P20856/Wodjil	3.6	3.9
97D002-26-7	P20856/Wodjil	3.6	3.9
97D002-30-3	P20856/Wodjil		3.7
P20855			3.7
P20856		3.5	3.7
P28348		3.6	3.8
P28716		3.3	3.5
P28865		3.5	3.5
Tanjil		2.0	1.5
Kalya		2.8	2.6
Belara		3.8	3.8
Pootallong		4.3	4.4
Wodjil		4.5	4.3
LSD		0.3	0.3

MATERIALS AND METHODS

LUPIN GERmplasm COLLECTION

In a quest to find a source of resistance to Anthracnose in *Lupinus luteus*, about 200 lines from the lupin germplasm collection centre at South Perth in WA were tested for Anthracnose resistance in New Zealand disease nursery in 1996/97. In 2000, an additional 100 *L. luteus* lines were tested in field conditions near Geraldton, WA, and 33 *L. hispanicus* lines were tested in glasshouse conditions at South Perth.

OVERSEAS GERmplasm

In 2000 two *L. luteus* lines were introduced from USDA collection and tested for Anthracnose resistance in glasshouse conditions using the techniques outlined by Cowling *et al.* (2000). One of the lines, PI168539 (hereafter referred to as P28716) showed some resistance and was crossed with other domesticated lines in 2001. The progenies were advanced to F₅ using single pod descent method. Single plants were selected from F₅ in 2004 and subsequent F₇ and F₈ populations were screened for disease resistance in 2006 and 2007

in small plot disease nurseries at Medina, WA (Table 1). High disease pressure was created by artificial inoculation and timely irrigation as outlined by Adhikari *et al.* (2006).

In 2003 nine *L. luteus* lines were received from the Russian Lupin Research Institute, Bryansk. These lines along with P28716, three WA breeding lines and controls were screened against Anthracnose in 2004 at Medina, WA (Table 2). P28716 is a very late flowering line and struggles to flower without vernalisation even in winter months, so this entry was always vernalised for three weeks at 6°C before planting in the disease nursery.

In 2005 an F₇ population derived from an Hungarian cv. Gyulatanya (P20856) was screened for resistance at Medina, WA. This population was previously untested and the resistance status of P20856 had not been determined.

Two crosses were made between the three putative sources of resistance (Bryansk x USDA, Bryansk x Hungary). Another two crosses were made between resistant (USDA, Hungary) and susceptible parents (Table 3). Approximately 120 F₂ progenies from each cross along with parental lines were screened against Anthracnose in the Medina disease nursery. At the time of pod set on the main stem, each plant was assessed for presence of Anthracnose lesion severing main stem. The percentage of plants surviving in each population was determined. Any plant with minor infection but without severing of branches or inflorescence was considered resistant.

RESULTS AND DISCUSSION

GERMPLASM FROM LUPIN GERMPLASM COLLECTION

No resistant germplasm was identified from the Lupin Germplasm Collection despite screening a total of 300 *L. luteus* lines in New Zealand and Geraldton and 33 *L. hispanicus* lines at South Perth. The *L. hispanicus* lines were tested because it is a very close relative of *L. luteus* and these species can be hybridised conventionally.

GERMPLASM FROM USDA

P28716 introduced from USDA showed a high degree of resistance in the initial glasshouse screening. It was a very late line and remained in the rosette stage in the absence of vernalisation in glasshouse conditions, even in winter. The internodes failed to elongate and the plant seemed very resistant even after repeated inoculations. In the next season it was vernalised and tested for Anthracnose where its level of resistance was determined to be moderate. This was the first confirmed report of Anthracnose resistance in *L. luteus*.

A range of resistance was expressed in the F₇ and F₈ derived lines from crosses involving P28716, with more than a dozen lines providing intermediate level of resistance (Table 1). In this test there was high inoculum

pressure and susceptible controls, such as Wodjil and Pootallong, had 100% of plants infected with the majority having main stems severed. Lines, such as 01D055-34, 01D055-43 were better than other lines in both years. Their level of resistance was superior to the resistant parent P28716 indicating transgressive segregation. Resistance in other lines was substantially better than the narrow leafed lupin cultivar Belara. Belara is moderately susceptible to Anthracnose, however, we believe that this level of resistance, when used with appropriate management approaches, will provide enough protection in south west of WA. *L. luteus* being a native of higher rainfall areas of the Iberian Peninsula requires more moisture than *L. angustifolius* and has greater potential to be a crop in the southwest region of WA.

Table 2. Anthracnose reaction of *Lupinus luteus* lines introduced from the Russian Lupin Research Institute, Bryansk along with controls when tested at Medina disease nursery in 2004. The disease reactions are in 1-5 scale, where 1 is very resistant and 5 is extremely susceptible.

Entry name	Assessment date	
	24 Aug.	27 Sep.
P28865	3.0	3.5
P28866	3.5	3.8
P28860	3.3	5.0
P28862	3.3	5.0
P28864	3.3	5.0
P28861	3.5	4.0
P28863	3.5	5.0
P28867	4.0	4.0
P28868	4.0	4.0
P28716	3.3	3.3
Teo	4.0	4.5
Wodjil	4.0	4.8
Pootallong	4.3	4.8
Kiev Mutant	5.0	5.0
Tanjil	1.0	2.0
Belara	3.5	3.9
LSD	0.68	0.60

GERMPLASM FROM HUNGARY

The F₇ population derived from the Hungarian lines P20855 and P20856 also showed some resistance when compared to susceptible controls (Table 1). Similar level of resistance was found in parents P20855 and P20856 confirming the source of resistance in the progeny. Another line P28348 (Portugal) also had some resistance. However, the level of resistance expressed was lower than the lines obtained from the cross involving P28716, such as 01D055-34 and 01D055-43.

Table 3. Survival percentage of approximately 120 F₂ progenies derived from various *L. luteus* crosses tested in an Anthracnose nursery at Medina.

Genotypes	Pedigree	Anthracnose status	% Survival
06D029	P28865/97D002-26-16	Resistant x Resistant	53.4
06D027	P28865/01D055-36	Resistant x Resistant	47.8
06D006	01D055-36/94D016-6-4	Resistant x Susceptible	37.5
06D013	97D002-26-16/94D016-6-4	Resistant x Susceptible	20.2
01D055-36	98D013-2/P28716	Resistant parent	64.9
P28865		Resistant parent	43.5
97D002-26-16	P20856/Wodjil	Resistant parent	32.8
94D016-6-4	Teo-101/Juno	Susceptible parent	13.6
Belara			36.2
Kalya			87.1

GERMPLASM FROM RUSSIA

Of nine germplasm obtained from the Russian Lupin Research institute, Bryansk, Russia, seven showed some resistance at the early growth stages (Table 2). However, as plants advanced and the disease pressure increased, many of them became fully susceptible. These lines were mostly restricted branching types and despite having less stem infection in the early assessment (24 Aug.), the later season infection resulted in complete absence of pods, as reflected in higher disease scores (27 Sep.). P28865 and P28866 were the best of the Russian lines, the former being slightly better than the latter. P28865 exhibited a similar level of resistance to P28716 at the final assessment in this disease nursery.

F₂ POPULATION TEST

F₂ progenies derived from resistant (R) x resistant (R) crosses gave higher level of resistance than resistant (R) x susceptible (S) crosses as reflected in the higher survival rate (Table 3). Within the R x S crosses, the survival rate of the progeny differed depending upon the level of resistance in the resistant parent (when crossed to the same S parent). Progenies from the cross 06D006 (USDA source) had greater survival than progeny from the cross 06D013 (Hungarian source). 01D055-36 (USDA source parent) in the former cross had a better resistance than 97D002-26 (Hungarian source parent) in the latter cross (Tables 1 and 3). These results indicated that the resistance is highly heritable and improvement in Anthracnose resistance can be achieved by choosing the most resistant parent for crossing.

Further studies are needed to determine whether different levels of resistance obtained from the parents are due to different genes. In this study the occurrence of susceptible progeny from R x R crosses does not imply that the resistance was due to two different genes. None of the parents were immune and many dead plants were observed in these resistant parents. More than 10% plants survived even in a fully susceptible line

94D016-6-4. What can be concluded from the test of the F₂ progenies is that the level of resistance in the progeny is very closely related to the parental resistance and that the resistance derived from the USDA source (01D055-36) was more effective than the Russian (P28865) and then the Hungarian (97D002-26).

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