Development of a world class lucerne breeding program

Summary
Lucerne is Australia's most important perennial forage legume, with about 3.5 million hectares sown, and has potential for expansion in New South Wales (NSW) and Queensland (QLD). Considerable genetic diversity exists within the *Medicago sativa* complex, which includes subspecies sativa and falcata, for a wide range of agronomic traits, including forage yield, pest and disease resistance, winter activity, drought tolerance and deep rootedness, grazing tolerance, tolerance of acid soils and others. This project identified germplasm exhibiting these traits at research nodes.

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Conclusions
The importance of lucerne in Australia continues to grow. This is due, in part, to previous achievements made by the Australian lucerne breeding programs. Australia has three public lucerne breeding programs that release industry leading varieties to market and conduct innovative basic research into developing new traits that target lucerne production in Australia and specifically benefit the grains industry.

In this project, background research has been carried out that will allow development of improved lucerne cultivars which will increase the productivity and utilisation of lucerne in Australia. Genetic maps of autotetraploid lucerne, linked to the Medicago truncatula map, have also been generated, and important agronomic traits have been integrated into the map. This will facilitate flow of technology generated in M. truncatula, which has been completely sequenced, to lucerne. The project has demonstrated the potential to develop more narrow-based synthetics or semi-hybrids, affording better protection of intellectual property generated in cultivars. This may facilitate greater investment into strategic research aimed at developing lucernes carrying new traits important to Australian agriculture.

The achievements of this project include release of two new candidate lines by Department of Industry and Investment, NSW (DII NSW); development of a trial network that included Victorian and Queensland locations and agriculture departments; the commercialisation of varieties by the University of Queensland (UQ); release of new dormancy 7 by the South Australian Research and Development Institute (SARDI); and development of new breeding lines by all three breeding nodes. These clearly demonstrate that this project met all its targets, despite funding restrictions, and developed new information for growers. Selection of traits for mitigation of effects of climate change will lead to development of better breeding lines by the programs in future endeavours. The availability of markers to the breeding programs (if negotiated) will allow for implementation of marker-assisted selections in Australian breeding programs. All these achievements can only lead to Australian growers reaping the benefits of this research into the future.

New lucerne cultivars are being developed with a range of important traits for the grains industry including grazing tolerance (ease of management), acid tolerance and improved nodulation (increased nitrogen fixation and improved adaptation and performance on acidic soils), disease resistance (improved ease of establishment and reduced transfer of diseases to crops in rotation), and seed size (improved establishment through increased vigour and sowing depth). Many of these traits will require additional breeding and evaluation over the next three to five years to reach commercial maturity.

The maintenance of these programs is important to agriculture in Australia, as highlighted by the recent identification of a potential new aphid biotype and the savings to industry that may be made if early identification and management of the problem are used to reduce the severity of the outbreak.

Recommendations
Before greater productivity and utilisation of lucerne in Australia is seen, the traits identified in this project will have to be incorporated into cultivars carrying multiple disease and pest resistances. This work will have to be carried out before the research outputs achieved in this project can be delivered to industry. This can be done through the conventional breeding of broad based synthetics, although this approach does not afford protection of intellectual property. If we are to achieve
greater productivity and utilisation of lucerne, then the strategic research undertaken in this project should continue. Unfortunately, funding sources for this to happen have not been forthcoming.

SARDI will continue to breed lucerne for a range of traits including large seed size, grazing tolerance, acid tolerance, and multiple disease resistance with an emphasis on diseases that affect lucerne and crops grown in rotation. The core breeding will be funded from lucerne royalty sales, with additional funding sought to fund specific industry requirements.

SARDI recommends that industry fund a short-term project to identify and develop management options for a new blue-green aphid biotype if a current SARDI experiment confirms a breakdown of resistance.

DII-NSW recommends that industry recognises the value lucerne brings to grain growers and the contribution of lucerne in minimising the effects of greenhouse gases such as nitrous oxide through biological nitrogen fixation and better soil health through incorporation of organic matter and carbon. Therefore it is recommended funding is provided for the programs to develop lucerne system packages and varieties with innovative traits to enable growers to minimise ground cover issues and maximise benefits to grain growers across Australia.

**Outcomes**

The lucerne plant has the capacity to deliver considerable benefits to the Australian grains industry. It has the capacity to arrest declining protein levels in wheat through its ability to fix atmospheric nitrogen. Lucerne rotations also provide greater livestock productivity in mixed farming operations because it is one of the most nutritious fodders available. It also has the capacity to manage rising water tables through its deep root system (up to 6 m). Lucerne also offers a disease break for all of the cereal rusts, and many of the cereal root diseases such as take-all and *Rhizoctonia*.

In subtropical Queensland, where most of the cropping is conducted on heavy cracking clay soils, there is no real viable alternative to lucerne, which shows adaptation to a wide range of soil types.

While these points above constitute mainly economic benefits, use of lucerne has the capacity to reduce soil erosion and to increase the long-term sustainability of our farming systems.

By improving the adaptation and productivity of lucerne through the activities of this project, there will be increased utilisation of lucerne in farming systems, leading to greater realisation of the benefits described above.

**Achievements/Benefits**

It has been well established that there are considerable economic, environmental and social benefits from greater use of lucerne in farming systems. However, modelling has indicated that there is considerable scope to increase the use of lucerne in Australia, particularly in western New South Wales and Queensland. If greater utilisation and productivity of lucerne is to be achieved in Australia, additional traits will need to be introduced into Australian cultivars. These traits include: inherent higher yield potential, increased winter activity, larger seed size to facilitate establishment, grazing tolerance, acid soil tolerance and improved water use efficiency. DNA marker assisted selection research has been conducted in this project to facilitate breeding of improved lucernes expressing these traits. The following achievements have been made at each node:

University of Queensland: Germplasm with more than 40% larger seeds than Australian cultivars has been sourced, and the trait introduced into adapted genetic backgrounds. UQ/GRDC-owned lines which outyielded Sequel by 20% after 11 harvests at Gatton were identified. Progress has been made in marker assisted selection, with two papers published in Theoretical and Applied Genetics providing map locations for resistances to *Colletotrichum trifolii* (races 1, 2 and 4) and *Stagonospora meliloti*. The use of male sterility to generate single cross hybrids and assess heterosis was also investigated and published (refer to Australian Journal of Agricultural Research (AJAR) 2008, 59: 999-1009), providing a potential mechanism by which intellectual property can be better protected than Plant Breeders’ Rights (PBR).

NSW Department of Industry and Investment: Candidate lines which are higher yielding than the check varieties are being multiplied and developed for release into the market. These varieties will also have pest and disease resistances which will lower chemical inputs. Acid soil tolerant germplasm will provide new unique materials for growers, particularly where lucerne has not been available so far as a feed option. New crosses will deliver future products suitable for the Australian environment and provide tools to mitigate effects of drought to a certain extent. A presentation by the DII-NSW breeder at the Northern American Alfalfa Improvement Conference (NAAIC) clearly placed the breeding efforts in Australia regarding lucerne and the
development of persistent, high yielding varieties. This was well appreciated by breeders and researchers from the USA and Europe. The rhizobia work and availability of rhizobia in Australia is also considered as significant by the US researchers who do not have access to lucerne appropriate rhizobia. Development of high yielding lucerne with persistence and grazing tolerance will remain a priority for NSW. DII-NSW is also keen to continue collaboration with the Department of Employment, Economic Development and Innovation (DEEDI) (formerly QDPI&F) if funding sources are available. Data sent by QLD from two local trials has been valuable.

**SARDI:** The SARDI lucerne breeding program has used this funding to identify and select germplasm with new traits: grazing tolerance, higher winter activity, large seed size and disease resistance. SARDI has developed screening techniques for root lesion nematode, *Fusarium, Pythium* and *Rhizoctonia.* The program has independently funded the evaluation of these new breeders lines in 28 full breeding trials that have been sown in collaboration with commercial partner Heritage Seeds over the past two years (three in QLD, four in Northern NSW, two in central NSW, four in southern NSW, four in VIC, one in Western Australia, and 10 in South Australia). The trial sites cover a range of farming systems and rainfall environments (300 mm annual rainfall at Point Pass, SA to 800 mm rainfall at Coleraine, VIC). Evaluation of this new germplasm will continue in future years as more seed becomes available. The program also independently screens for abiotic stress tolerances including improved root growth with acid and aluminium tolerance, and improved nodulation at low pH. In August 2009, SARDI scientists believe they found a new blue green aphid (BCA) biotype with more virulence and higher fecundity (result currently being confirmed). SARDI has developed markers for blue green aphid resistance and is in the process of developing a marker for spotted alfalfa aphid (SAA). SARDI has also improved its capacity to isolate and culture *Phytophthora* and anthracnose.

A continuous grazing trial at Turretfield, SA, has validated the improvement in sheep grazing tolerance made in SARDI Grazer. This cultivar was developed in GRDC project DAS347 with collaboration from the Department of Agriculture and Food in WA. The level of grazing tolerance in SARDI Grazer is significantly greater than all other commercial cultivars including Stamina GT; a cultivar developed in the USA for grazing.

**Other research**

The research conducted in the project was of a strategic nature, and funding beyond the two years of this project is needed to deliver the outputs to industry.

A potential biotype of blue green aphid (BCA) has been recently discovered by SARDI scientists. A BCA collected from Urrbrae in South Australia has greatly increased virulence and fecundity. If widespread, this aphid could be devastating for lucerne growers in Australia. A breakdown of resistance has been identified in cultivars bred by all programs tested including SARDI, DII-NSW, UQ, WL (Waterman and Loomis), Forage Genetics and Pioneer. Resistance in highly resistant cultivars, SARDI Five and WL414 (usually 40-50% resistant), was reduced to less than 10%. Importantly, mortality in seedlings, which is usually rare, was between 20-80% for all populations tested. Experiments are now underway with a larger number of cultivars (to be completed by September 30) to confirm this result before making a public announcement. Research will be urgently required to (1) phenotype aphids from around Australia to provide early indications of aphid diversity with regards to virulence and fecundity, (2) identify genetic differences in aphid DNA, and (3) conduct larger scale aphid surveys using the molecular markers identified to provide a quick and cheap assessment of the new aphid biotype distribution. Plant breeders will then need to quickly develop and release new resistant breeding populations to assist with integrated pest management.

**Intellectual property summary**

UQ: all background intellectual property brought into this project is co-owned by UQ and GRDC. Material provided to J. Irwin by University of Wisconsin-Madison has been gifted to the researcher, so UQ has full freedom to operate on project outputs. UQ/GRDC background intellectual property has been kept isolated from the other parties to this agreement in the absence of a long-term, full collaboration being achieved.

SARDI has not used third party property and therefore has freedom to operate. Commercialisation of new cultivars follows a long-term successful partnership with Heritage Seeds. New cultivars have PBR protection and cultivars with novel traits and breeding methods may additionally be covered with a patent.
DII-NSW. All background intellectual property is co-owned by DII-NSW and GRDC. Commercialisation of new candidate line identified will be through Seedmark.

**Additional information**

Further information is provided in an attachment to this report.

**Updated results for new BGA biotype**


**Updated results for performance of SARDI Grazer**


**Grazing tolerant lucerne**

One of the main constraints to lucerne adoption is the perception that it requires controlled rotational grazing management for good persistence. Because of this, lucerne is considered by many to be a specialist pasture, and not a species that can be grown across large areas of the farm. New varieties have been bred with improved tolerance to grazing, and can now be managed with more flexible grazing.

Lucerne is a perennial plant that uses energy stored in its taproot to grow. Once the new growth is about 20 days old, it starts to put energy back into its root system. The plant is usually grazed or cut for hay 30 days after cutting and this triggers new growth from the stored energy. This is a cyclic pattern of energy storage, use and replenishment. Rotational grazing will always be the most productive way to utilise lucerne, resulting in high yields and improved livestock production. Unfortunately it isn't always possible to graze lucerne this intensively because of fencing or watering points, or other livestock issues (mob size, area of sown lucerne, etc).

A new lucerne cultivar called SARDI Grazer has been developed to tolerate more persistent grazing. SARDI recommends a flexible grazing rotation of around 6-8 weeks grazing followed by 6-8 weeks recovery.

The two paddock rotation requires much less infrastructure and labour inputs, and should make lucerne more accessible to more farmers. The two paddock rotation is also close to traditional management for perennial grasses such as cocksfoot and phalaris, making this lucerne a more compatible option for sowing in mixtures.

SARDI Grazer was developed by selecting plants that survived 18 months of intensive continuous grazing from sheep. This extreme level of grazing is not recommended for farmers, but was used as a breeding tool to kill most of the plants as quickly as possible and leave only the most grazing tolerant individuals surviving for breeding. The surviving plants were crossed together to form seed of the next generation, which was then selected again for tolerance to continuous sheep grazing. This process was repeated once more, so that the resulting parent plants in the new variety had three generations (or cycles) of selection for tolerance to long spells of continuous sheep grazing (table 1).

<table>
<thead>
<tr>
<th>Trial</th>
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<tr>
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<td>2003-06</td>
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<tr>
<td>Katanning Continuous Grazing trial, WA</td>
<td>2003-06</td>
<td>3</td>
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<tr>
<td>Turretfield Continuous Grazing trial, SA</td>
<td>2007-10</td>
<td>4</td>
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**Table 1. History of SARDI grazing tolerant lucerne trials**
SARDI Grazer is a result from almost 30 years of breeding, and was developed from selections of third generation grazing tolerant plants at Turretfield (alkaline clay loam) and Katanning (deep acidic sand) lucerne sites. The performance of SARDI Grazer at Turretfield in 2007-10 is shown in Figure 1.