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Soil biology review and project prioritisation for the feedbase investment plan

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Abstract

Soil health has been identified as a key theme area of the Feedbase Investment Plan.

Based on a review of an MLA discussion paper, a “strawman” discussion paper was developed which suggested priority research topics in the soil biology area. The “strawman” was tested at an interactive workshop with producers, advisors and researchers from which emerged a series of key objectives for a single project specification to be developed to address soil-borne root diseases in sub-clover.

Soil borne root disease in southern Australian pastures was agreed to represent a significant productivity constraint, particularly during the autumn-winter period and therefore an opportunity to assist producers lift productivity considerably if soil-borne diseases could be adequately managed.

University of Western Australia and SARDI were confirmed as the best candidates to undertake the work.

Project specifications were developed for two soil biology projects and are provided. One for the main project to cover objectives indicated at the soil biology workshop and a second to complement the first project should additional funds become available.

Subsequently, the project development in soil biology needed to account for potential overlaps with parallel research planning on “Plant Health” within the Feedbase plan development. There was also overlap with the Legume Performance theme, particularly with respect to productivity and persistence of legume species in pastures.

A project specification and accompanying contract schedule was developed for soil biology research which addressed the defined soil biology research objectives and also contributed to some elements of the “Plant Health” research priorities.

A second project specification was developed to complement the first project should additional funds become available.

Suggestions for complementary PhD projects are provided.

Executive summary

"Soil Health" has been identified as a key theme area within MLA's Feedbase investment plan.

An internal discussion paper was drafted which collated elements of previous reviews, former soil biology research and researcher feedback to form a basis on which to plan new research within the "Soil Health" space.

This project was commissioned to:

- Review the discussion paper in relation to MLA's proposed Feedbase investment plan
- Prepare a "strawman" discussion paper proposing suggested priority research investment areas for soil biology as a basis for an interactive workshop to agree on research priorities
- Propose project investments for soil biology under two funding scenarios – 1. Up to $1m over 5 years; 2. Should additional money become available.
- Propose the research organisation for the recommended projects
- Provide a description of the project outputs matched against the outcome stated in the Feedbase planning documents.

The "strawman" paper was presented to the interactive workshop comprising producers, advisors and researchers and it was agreed that the primary focus of soil biology research should be on better managing soil born root pathogens that affect sub-clovers in pastures.

Evidence from MLA's recent soil biology research program indicated that root damage may potentially constrain pasture yields by from 18% to 37% (mean 26%) during autumn-winter. Given the widespread occurrence of root damage found and the fact that the autumn-winter period is a critical time of feed shortage, it is possible that this estimate indicates a significant problem for livestock enterprises across southern Australia.

Simpson R and Richardson A (2009) estimated the impact of root damage to grazing system productivity and enterprise profitability by simulation using Ausfarm and the GRAZPLAN pasture and animal models. For the purpose of the simulations it was assumed that root damage constraints only applied to autumn-winter production. A farm stocked conservatively (12 sheep/ha) and consequently achieving a relatively low income, was predicted to experience a significant but comparatively low financial loss (16%) when restrictions to autumn-winter pasture growth were moderate (i.e. <20% reduction in autumn-winter growth), but would suffer a 54% constraint on income if autumn-winter pasture growth were reduced by 40%. At higher stocking rates (15-18 sheep/ha) with potentially higher farm incomes, the impact of restricted autumn-winter pasture growth was more obvious with ~25% constraints on income for a 20% reduction in autumn-winter pasture growth and massive 58-84% constraints on income for a 40% reduction in autumn-winter pasture. Indeed, at pasture growth reductions above 20% the value of lifting stocking rate to improve net farm income was predicted to be progressively lost as a consequence of root damage and a grazier's financial situation may even deteriorate seriously if root damage were constraining autumn-winter pasture growth by 30% or more.

This evidence was supported by other research and literature reviews (e.g. Barbetti et al, 2005).

The soil biology workshop determined that the main objectives to be addressed in soil biology research were to:

1. Determine the farm management factors that influence the expression of soil-borne root diseases in sub-clover pastures.

2. Evaluate practical management techniques aimed at reducing the pasture productivity loss during autumn-winter induced by soil-borne disease

3. Characterise the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots.

4. Broaden the quantification of pasture productivity constraints induced by pasture soil-borne pathogens across agro-ecological zones

5. Develop molecular assays to fill gaps in experimental tools needed to adequately conduct research into, and monitor, soil borne disease in pastures.

Any new project was also seen as an opportunity to build research and knowledge capacity in pasture pathology in light of the near retirement of the most experienced scientists.

The University of Western Australia (Dr M Barbetti) and the South Australian Research and Development Institute (SARDI – Drs R Ballard, A McKay) were identified as the best groups to undertake the work.

A project proposal (***Effective management of soil-borne disease threats to southern Australia pastures***) was developed by Dr M Barbetti (UWA) which addressed all the soil biology research objectives and which was aimed at meeting funding scenario 1. This project placed emphasis on chemical and cultural (non-breeding) control measures of soil-borne diseases in sub-clover as well as understanding the interaction between expression of disease symptoms and the presence of dominant pathogens and pathogen complexes.

A complementary project (***Using applied DNA methods to identify practices that improve sub-clover root health)*** proposal was also developed by SARDI to fit with funding scenario 2 and which placed more emphasis on variation in how sub-clover cultivars vary in their responses to cultural control measures developed within the scenario 1 project. This project also offered the scope to use novel molecular DNA assays of soil-borne pathogens to characterise the relationships between root pathogen levels in soils before the season break and the expression of disease during the critical during autumn-winter period. Resolving such an issue has the prospect to provide producers and advisors with diagnostic tools to estimate the risk to pasture productivity based on pre-season levels of pathogens in soils.

During the course of developing the soil biology projects, the potential for overlap or conflict in project development in the parallel theme of “Plant Health” was acknowledged.

A "Plant Health" final report and subsequent workshop suggested the following research priorities under the "Plant Health" theme which had potential to overlap with the soil biology project objectives:

- ***Rhizobium function*** – "identification of the impact of changes in farm practices, especially herbicide impacts on fixation"
- ***Diseases*** – "The evaluation of non-breeding approaches to fungal and viral disease management, including cultural practices"

It was accepted that the soil biology project "***Effective management of soil-borne disease threats to southern Australia pastures"*** fulfilled the requirements of the "Plant Health research priority relating to disease management.

This soil biology project also covered aspects of the impact of soil-borne disease on rhizobium function and nitrogen fixation but not specifically the effect of pasture herbicides.

This overlap, and convergence, of "Plant Health" and Soil Biology" research priorities resulted in a merging of the two budgets such that the soil biology projects meet also the aspirations of the Plant Health program.

It is proposed that a PhD project be developed to provide particular focus on the interactions between soil-borne disease, herbicides and cultural practices on rhizobium function. This meets the need of the "Plant Health" research priority and also adds a further dimension to the soil biology work with respect to effective nitrogen fixation in sub-clover pastures.

A further PhD is proposed in basic science to determine the biochemical mechanisms of pathogen resistance and tolerance in sub-clover plants, if and how that differs with variety and the interactions of these factors with rhizobium symbiosis. This project will complement the soil biology and "Plant Health" themed projects and will also help identify specific breeding objectives to develop sub-clover varieties with superior pathogen resistance and tolerance.

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1 Background

"Soil Health" has been identified as a key theme area within MLA's Feedbase investment plan.

An internal discussion paper was drafted which collated elements of previous reviews, former soil biology research and researcher feedback to form a basis on which to plan new research within the "Soil Health" space where the emphasis is on soil biology.

A key element of MLA strategic investment plan was to seek priorities that support "Productive and Sustainable Pastures and Soils". Key areas of focus were to be:

- *Productive and well managed soils* – resolving soil based limitations to pasture production – focus on soil nutrients (P&N), soil biology (including soil C) and soil physical limitations.
- *Pasture agronomy* - emphasis on producing more and better pastures for longer – focus on better legumes and performance (including N fixation), productive low P pasture systems, plant P efficiency, persistence of new-sown pastures, NRM impacts
- *Healthy pastures* – emphasis on pasture plant health for better performance and resilience – focus soil borne root pathogens of forage legumes and grasses, the interaction between mineral nutrition and beneficial and pathogenic soil organisms, plant resistance/tolerance to disease.

MLA supported a significant research program on soil biology research during the period 2003-2008 which left a significant legacy of achievements on which to build future research investments in soil biology. The program was terminated at the completion of the first of a three phase, long-term, investment strategy.

This project was commissioned to develop new project specifications for at least 2 projects on soil biology which would underpin productivity improvements in sub-clover pastures in southern Australian pasture systems.

The project was to be developed in parallel with a "Plant Health" theme identified within the Feedbase Investment Plan to cover pests, diseases and rhizobial function in pastures.

The project was also to be developed cognisant of a new project about to begin (B.PUE.0103 – Root Disease constraints in pasture productivity) which sought to focus on phosphorus nutrition in relation to soil borne diseases.

The author of this report was engaged by MLA as Program Manager for the MLA Pasture Soil Biology Program which was active between 2003-2008.

2 Project objectives

The objectives of this project were to:

- Review the discussion paper in relation to MLA's proposed Feedbase investment plan
- Prepare a "strawman" discussion paper proposing suggested priority research investment areas for soil biology as a basis for an interactive workshop to agree on research priorities

- Propose project investments for soil biology under two funding scenarios – 1. Up to $1m over 5 years; 2. If additional money should become available.
- Propose the research organisation for the recommended projects
- Provide a description of the project outputs matched against the outcome stated in the Feedbase planning documents.

During the course of the project, the project brief was varied to facilitate the margining, in part, of the "Soil Biology" and "Plant Health" themes to incorporate research priorities identified in the "Plant Health" theme. These related specifically to:

- ***Rhizobium function*** – "identification of the impact of changes in farm practices, especially herbicide impacts on fixation"
- ***Diseases*** – "The evaluation of non-breeding approaches to fungal and viral disease management, including cultural practices".

The additional tasks included ensuring that the identified "plant Health" priorities were incorporated into the soil biology projects, developing and negotiating a contract schedule for at least one project (main project – scenario 1) and to develop a complementary PhD project in which rhizobial function in sub-clover as affected by herbicides was incorporated.

3 Method

A review was conducted of a 22 page discussion paper prepared internally by MLA (Driver 2011). This paper sought to align MLA's research portfolio in soil science and pasture systems with the corporate strategic plan and national R&D strategies and summarise a range of relevant reports reviews and programs that had relevance to MLA's soil science area of interest. The objective was to determine if this paper provided sufficient justification for research investments in the soil science area a detailed review of prospective research investments in soil biology.

A paper was then developed in the form of a "strawman" to propose level investments in pasture soil biology as a discussion paper. This paper was to set out the author's opinion of priority projects which would contribute to an MLA goal of increasing meat production by at least 2.5%. without compromising sustainability.

The "strawman" discussion paper was tested at a workshop comprising producers, advisors and scientists with the aim to refine the "strawman" and determine an agreed investment plan at project level in soil biology for use by MLA and partners.

Following the workshop, the author embarked on negotiations with the nominated scientists to develop project specifications for both scenario 1 and 2 as required by the brief.

During this period, it became apparent that the "Plant Health" theme research priorities overlapped with those of "Soil Biology". A process of consultation and negotiation was undertaken to merge the two themes with respect to soil borne disease management and rhizobial function consistent with an increased investment pool.

A draft contract schedule was developed for the project identified for scenario 1 and project outlines developed for suggested PhD studies aligned with the merged "Soil Biology" and "Plant Health" themes.

4 Project outputs

4.1 Discussion paper review

The discussion paper (Driver 2011) gave a strong overview of recent and current reports, reviews and programs pertaining to MLA's potential interests in soil health. The recommendations from this discussion paper placed emphasis mainly on:

- Monitoring tools for soil "condition" as it relates to soil carbon and nutrient dynamics
- Models for ecosystem function in agricultural land-use based on metagenomics
- Tools and models for soil biological function and resilience
- Shared data to develop metadata standards for landscapes and land use.

The aspirational suggestions from this paper focused more on general soil condition and were remote from practical farmer-useful "what to do" solutions.

Hence the author's conclusion was that there should be more research investment focus, at least with the limited funds available, on practical solutions for farmers to deploy and at developing research tools to assist and accelerate that research.

It was concluded that there was no need for a specific review of research opportunities in pasture soil biology as the recent Pasture Soil Biology Program which concluded in 2008 left a legacy of:

- A knowledge and Opportunity Audit (Gupta and Ryder, 2003) which formed the basis of developing the Pasture Soil Program portfolio of projects and remains relevant.
- A series of reports, project outputs and recommendations from the MLA Soil Biology Program on which to develop new projects in 2012. See a summary in Hannam, 2008 – Pasture Soil Biology Program Final report)
- A business case which proposed a further series of project investments as phase 2 of the Pasture Soil Biology Program to follow the success of phase 1.

[**Note:** Phase 2 of the Pasture Soil Biology Program was not supported by MLA. The overall plan envisaged three phases:

- Phase 1 – develop new research tools (DNA assays) to facilitate quantitative research of soil biological organisms and function. Much of this was achieved along with some studies on characterising soils under different management for beneficial and pathogenic soil borne organisms.
- Phase 2 – use the research tools to investigate and understand the dynamics and interactive influences favouring beneficial or root pathogenic organisms in pasture soils.
- Phase 2 and 3 – Develop proven and practical pasture management tactics to enhance beneficial and constrain pathogenic organisms in pasture soils to underpin pasture productivity improvements, particularly during autumn-winter when root pathogens have their most significant impact.

That subsequent phases of the planned soil biology research were not supported meant that the achievements in phase one were not capitalised upon, although the legacy was sufficient to develop any further investments in soil biology that MLA may consider]

The Strawan Discussion Paper for Soil Biology

A "strawman" discussion paper was developed as a basis for developing new research projects in pasture soil biology aligned to MLA's R&D priorities for deliverables that support "Productive and Sustainable Pastures and Soils" in which pasture productivity improvements underpin a goal of 2.5% increase in meat production without compromising sustainability.

The key areas of MLA's R&D focus include:

- *Productive and well managed soils* – resolving soil based limitations to pasture production – focus on soil nutrients (P&N), soil biology (including soil C) and soil physical limitations.
- *Pasture agronomy* - emphasis on producing more and better pastures for longer – focus on better legumes and performance (including N fixation), productive low P pasture systems, plant P efficiency, persistence of new-sown pastures, NRM impacts
- *Healthy pastures* – emphasis on pasture plant health for better performance and resilience – focus soil borne root pathogens of forage legumes and grasses, the interaction between mineral nutrition and beneficial and pathogenic soil organisms, plant resistance/tolerance to disease.

The "strawman" provided two project outlines as prospective areas of research activity to achieve MLA's strategic goals through improvement in soil health.

Based on a significant body of evidence (both historical and recently in the 2003-2008 pasture soil biology program), emphasis was placed on minimising the impact of soil-borne root pathogens in reducing pasture plant productivity in the medium-high rainfall zones.

The rationale was that the dominant economic issue for producers is carrying stock through periods of feed shortage, usually the autumn-winter period. Better carrying capacity at this time results in improved pasture utilisation in spring and enhanced producer profit. Hence the focus on this is likely to have the most immediate benefit to producers if solutions can be found.

The autumn-winter period is when soil borne pathogens have their most significant impact on production and persistence of key pasture species and where recent evidence suggests that between 25-58% losses in potential farm income is possible due to poorer pastures compromised by root damage from soil-borne disease

Hence the focus of the strawman proposals was to develop practical management tactics for producers to minimise this productivity loss. Two themes were proposed:

- Managing soil borne pathogens affecting important pasture species (e.g. sub clover)
- Develop diagnostic, monitoring and benchmarking tools for researchers, advisors and producers to measure key soil borne organisms to quantify responses to pasture and soil management strategies.

Further research ideas were also proposed for consideration:

- Some basic science to define the biochemical mechanisms of tolerance and resistance to soil borne pathogens in key pasture species.
- Understand carbon and nitrogen cycles in soils through developing molecular techniques to measure soil biological function for these two key processes

A background paper was also produced to provide some basic understanding of soil biology under pastures and also to outline the achievements and aspirations of the recent pasture soil biology program. The aim here was to inform development of any new projects in pasture soil biology with the knowledge of what has recently been done.

The papers are provided in Appendices 7.1, 7.2.

The strawman and discussion paper were distributed to PICs partners for comment before distribution to participants in an interactive workshop aimed at agreeing on the research priorities in soil biology for 2012.

All participants were encouraged to provide any feedback prior to the workshop.

4.2 Pasture Soil Biology Workshop

An interactive workshop was held on 7th February 2012 at which a forum of producers, researchers and advisors with interests or experience in soil biology discussed the strawman and suggested research priorities.

The workshop agenda sought to review the strawman under funding scenario 1, agree priorities and achievable objectives for the limited budget available, agree researchable questions, agree planned outcomes and drill down to project specification detail to the extent possible including indicative budget and who(persons/groups) are best able to undertake the work.

The workshop produced the following outcomes:

Hypothesis: ***Soil borne root pathogens constrain pasture productivity***

This assertion was based largely on a significant literature base and recent evidence from a novel field bioassay technique applied to pastures.

Research Needs: (These are to be designed with a $200K/yr budget over 5 years)

- To determine the factors that influence expression of soil-borne root diseases in pastures
- To characterise the expression of root disease in pastures and the associations with soil borne pathogens on affected plant roots
- To develop new molecular assays to fill gaps in experimental tools needed to adequately conduct research into soil borne diseases of pastures.
- Broaden quantification of pasture productivity constraints induced by soil borne pathogens across agro-ecological zones.

Key Research Groups

Researchers at **SARDI** and **UWA** were identified as having the best expertise and capability to undertake this work.

Notes: The following directions were suggested:

- That the primary focus should be on subterranean clover in permanent pasture situations

- That in developing soil biology projects, there be overt linkages to other projects (P vs root damage, relevant fields sites, the legume stability component of the Feedbase investment plan and perhaps pasture variety testing sites)

A range of additional research opportunities were also identified should additional finance become available. These include:

- Expand the research indicated above to grasses and medics
- Undertake pathogen ecology (complex) studies to complement the above project
- Undertake functional soil-biota community analysis (This will require a separate project specification)
- Understand the effects of water management (e.g. drainage) on expression of disease and soil biological community structure
- Determine the soil profile stratification of soil borne pathogens on roots to depth
- Determine the effects of agricultural chemicals on soil borne diseases and soil biological community structure
- Evaluate the prospects of practical chemical control of soil borne diseases in pastures
- Enhance knowledge on the mechanisms and management options to induce resistance to root pathogen infections on pasture plant roots.
- Link delivery from soil biology projects to the “Soil Health” network

Terms of reference for a project specification on soil biology were developed:

Main Project

Deliverables

- A project specification defining a research opportunity on soil borne root pathogens of pastures and the factors affecting the expression of disease and pasture production loss.
- The project specification to include:
  - Project title
  - Purpose and objectives
  - Justification
  - Proposed experimental approach and details
  - Budget (limited to $200K/yr over 5 years in addition to “in-kind”)
  - Personnel and research organisations
  - Intellectual property contributions

Considerations

- Initial focus on subterranean clover in permanent pastures of southern Australia
- Determine the factors that influence expression of soil-borne root diseases in pastures
- Characterise the expression of root disease in pastures and the associations with soil borne pathogens on affected plant roots
- Identify and develop new molecular assays required to fill gaps in experimental tools needed to adequately conduct research into soil borne diseases of pastures.
- Broaden quantification of pasture productivity constraints induced by soil borne pathogens across agro-ecological zones.

- Linkages with other relevant research activities developing within the Feedbase Investment Plan to be defined.

Complementary Research Projects – to the main project

Should additional investment become available, other projects may be considered for investment. These are listed in the workshop outcomes (above)

Deliverables

- Summary project specifications defining the complementary research opportunities in pasture soil biology for up to three priority projects.
- The project specifications to include:
  - Project title
  - Purpose and objectives
  - Justification
  - Overview of proposed experimental approach(es)
  - Indicative budget
  - Suggested personnel and research organisations

Considerations

- Define linkages to the research objectives of the main project
- Define linkages with other relevant research activities developing within the Feedbase Investment Plan to be defined.

4.3 Project Specifications for Soil Biology Research 2012

4.3.1 Scenario 1 – Effective Management of Soil-borne Disease Threats to Southern Australia Pastures (also draft contract schedule)

A project specification was developed by UWA (in consultation with the author) with the following objectives:

To enable producers to reliably enhance the productivity from their pastures by c. 25% by:

1. Determining the farm management factors that influence the expression of soil-borne root diseases in sub-clover pastures.
2. Identifying effective and practical management techniques that reliably reduce the pasture productivity loss during autumn-winter induced by soil-borne root disease.
3. Characterising the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots.
4. Broadening the quantification of pasture productivity constraints induced by pasture soil-borne root pathogens across agro-ecological zones.
5. Developing molecular assays to fill gaps in experimental tools needed to adequately conduct research into, and monitor, soil borne diseases in pastures.
6. Securing pasture pathology skills and expertise for the future

The project specification is shown in full in appendices (7.3).

In overview, glasshouse and field experiments will be conducted to evaluate and select the most promising biocide, pasture cultural practices and nutrition treatments that producers can adopt to better manage soil-borne pasture diseases. Regional, environmental and edaphic factors that influence the dominance of particular pathogens and pathogen complexes in relation to the expression of disease (symptoms and sub-clover productivity) will be evaluated to help better formulate management strategies. This will include examination of the interaction between root disease and rhizobial function in sub-clover plants.

Further, molecular assays which are useful to identify and quantify pathogens present in soils and roots will be expanded and validated as research tools.

The project will also focus on training a pasture pathologist – soil biologist as an investment in maintaining and enhancing the knowledge on soil borne disease management which will be available to producers and future research.

A draft contract schedule was developed for this project was developed to help facilitate contracting this project by 1 July 2012. This is shown in the appendices (Appendix 7.4).

4.3.2 Scenario 2 – Using Applied DNA Methods to Identify Practices that Improve Sub-clover Root Health

An additional project specification was developed by SARDI (in consultation with the author) to complete the brief of this project. This project specification is available should additional funds become available.

The full project specification is provided in the appendices (Appendix 7.5). It is designed to be complementary to the "scenario 1" project. This project places greater emphasis (than in the scenario 1 project) on genetic variation among sub-clover varieties in resistance and tolerance to soil borne pathogens, pasture age and synergies with practical disease control measures evaluated in this project and identified in the scenario 1 project.

In brief summary, the project seeks to use applied DNA assays to simultaneously measure several pathogens in the soil and plant roots in grazed pastures to:

- Better understand how contemporary sub clover cultivars with different resistance to root pathogens respond to management practices used to reduce the impact of soil borne diseases across different environments, seasonal conditions and pasture age.
- Characterise relationships between pathogen levels in soil before the break and root disease levels during autumn/winter in key agro-ecological zones.
- Identify important root pathogens for which molecular assays have not yet been developed to expand the suite of DNA assays such that they can be used to reliably explain variation in plant root health. This data can then be used to develop pathogen risk profiles for use by advisors and producers(similar to current use of these assays in estimating productivity loss potential from cereal soil borne diseases).

The project will collect and archive soil extracted DNA for possible future studies on soil microbial community analysis including structure and function to assess impact of farm management on soil borne disease.

There is an opportunity for this project to be directed to greatly enhance the reach of the scenario 1 project in terms of extending the use of DNA assays of soil borne pathogens to a wide range of pasture environments (including benchmarking sites within the "Legume Performance and Proportion" theme) to characterise the nature of the pathogen complexes present, evaluating biological constraints with strategic use of field bioassays and also identify unusual pathogen complexes worthy of further direct pathology examination within the scenario 1 project.

The project will seek to develop a pasture research capability to help address future RD&E needs of the livestock industry by mentoring a scientist in pasture soil biology who is equipped with a modern pathology skill set focussed around the use and interpretation of molecular assays.

These project plans have been developed with the assumption that these projects will link with:

- Project B.PUE.0103 “Root Disease Constraints in Pasture Productivity” which is focussed on the interaction between pasture phosphorus nutrition and root disease.
- Projects funded by the MLA “Plant Health” theme of the Southern Feedbase Investment Plan.
- Projects funded by the Feedbase Investment Plan on “Pasture legume performance and proportion”.

4.4 Additional Project Suggestions

The following project ideas are offered which would be complementary to the main research project:

PhD – Herbicide and Root Disease Impacts on Rhizobium Function in Sub-clover

This project would study the interactions between soil borne root disease, herbicide use and rhizobium function in sub-clover. The candidate would operate under the supervision of Dr Martin Barbetti (UWA) and use both the glasshouse techniques and field sites used in the Scenario 1 project to facilitate a unique learning experience and also share relevant data to expand the collective output from the soil biology work.

This would also address the specific “plant health” theme research priority on the effects of herbicides on rhizobial function

The project could consider the use of herbicide in crop-pasture mixed farming systems as well as herbicide use in permanent pastures. It may be possible to also consider the impact of pasture insecticides on rhizobial function if it is thought that the student could cope with the broader brief.

It is suggested that consideration of soil borne disease be given some importance as herbicides can weaken non-target pasture species making them more vulnerable to disease as well as compromising root growth and rhizobial function. There may also be beneficial effects of rhizobium inoculation on limiting root disease.

The planned outputs would be knowledge to:

- Define the interactions between root disease, herbicides and rhizobial function
- Define the impacts of these interactions on root growth, seedling performance, rhizobial function and nitrogen fixation potential
- Identify intervention strategies to optimise rhizobial function in relation to herbicide selection and root disease management

PhD – Biochemical Mechanisms of disease resistance and tolerance in sub-clover

The aim here would be to identify biochemical mechanisms for resistance and tolerance to some important soil-borne root pathogens and examine how these mechanisms interact (and impact on) symbiotic rhizobia prevalence and activity associated with sub-clover plants.

This is a basic science project with higher risk, but with profound potential to influence both pasture management strategies to manage disease and plant breeding programs for disease resistance.

It would seek to discover if and perhaps how selected chemicals (key active molecules) may influence resistance and tolerance characteristics and robustness in disease environments. This may provide a practical management tactic to help manage soil borne diseases in established pastures and the concepts may have application to foliar diseases and viruses as well (also part of the “Plant Health” theme research priorities.

It is suggested that this PhD student be under the supervision of Dr Ulrike Mathesius (ANU) with co-supervision from Drs Barbetti (UWA) and Simpson (CSIRO). Dr Mathesius has expertise in studying plant physiological responses to treatments including chemical signalling between plant roots and soil borne organisms.

This project would also relate to the “Plant Health” theme research priorities on the importance of rhizobial function in sub-clovers to support nitrogen fixation with interactions from soil borne disease.

The planned outputs would be knowledge to:

- Develop breeding objectives for resistance and tolerance to disease in sub clover
- Develop systemic foliar applied bio-chemicals that can enhance resistance and tolerance to pathogens
- Better manage symbiotic N fixation potential in sub-clover in interaction with soil-borne disease

5 Success in achieving objectives

The objectives of the initial brief for this project have been achieved - two project specifications on soil biology which are linked to MLA’s strategic research objectives on soil health within the Feedbase Investment Plan.

However, during the course of the project, it became obvious that parallel research planning activities under the themes of “Plant Health” and “Legume Performance and Proportion” represented considerable overlap with the “Soil Biology” research project planning.

The “Plant Health” theme implied factors (disease, rhizobia) that affect pasture plant roots and which are key elements of the soil biota.

The “Legume Performance and Proportion” theme should also be affected by soil biology through root borne pathogens (disease) affecting both plant performance and seed bank maintenance which can compromise persistence of key pasture species and therefore their proportion within pasture swards.

Hence the project brief was extended to interact with the “Plant Health” theme to ensure that conflicts were avoided and that complementarities were secured. This has been achieved for at least the “Plant Health” theme.

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7 Appendices

7.1 Strawman

PASTURE SOIL BIOLOGY RESEARCH

A "STRAWMAN" – PROJECT SUGGESTION

Improving Pasture Productivity by Minimising Soil Biological Constraints

Introduction

This document seeks to provide the basis for developing a research project in pasture soil biology which will meet MLA's R&D investment priorities for deliverables that support "Productive and Sustainable Pastures and Soils". Improved pasture productivity that leads to 2.5%pa increase in meat production, without reducing sustainability, is sought.

Key areas of focus are:

- *Productive and well managed soils* – resolving soil based limitations to pasture production – focus on soil nutrients (P&N), soil biology (including soil C) and soil physical limitations.
- *Pasture agronomy* - emphasis on producing more and better pastures for longer – focus on better legumes and performance (including N fixation), productive low P pasture systems, plant P efficiency, persistence of new-sown pastures, NRM impacts
- *Healthy pastures* – emphasis on pasture plant health for better performance and resilience – focus soil borne root pathogens of forage legumes and grasses, the interaction between mineral nutrition and beneficial and pathogenic soil organisms, plant resistance/tolerance to disease.

New projects are now being developed in the ***soil biology/soil health*** area that will contribute solutions, tools or support for producers to better manage soil biological health for improved pasture (and therefore livestock) productivity.

There are two funding scenarios proposed by MLA for the soil biology work:

1. What should be the priority research to be undertaken with a budget of c. $200K per year over 5 years? (This suggests that a single project is likely).

2. What priority research projects should be considered if additional funding becomes available?

The Strawman (funding scenario 1)

Two project outlines are suggested below which seek to target soil biological constraints to pasture establishment and persistence as primary areas of risk and cost to producers.

Overview

Key Focus – management tactics for minimising the impact of soil borne pathogens on productivity of targeted pasture plants in medium/high rainfall permanent pasture zones

Deliverables – calibrated molecular assays for research, monitoring, benchmarks; practical soil/pasture management tactics for biological constraints; microbial-ecology knowledge; capacity building.

Primary Suggestion

Managing soil biological constraints to pasture plant health and productivity

Theme – management strategies for minimising the impact of soil borne pathogens on productivity of targeted pasture plants.

Primary Targets

Pathogens – recent reviews suggest that the following pathogen organisms should be primary targets – *Phytophthera clandestina, Pythium irregulare, Aphanomyces eutichies, Rhizoctonia spp, Fusarium spp*. Nematode pathogens may also be important.

Plants – sub clover (annual legumes), Lucerne, ryegrasses, selected perennial grasses.

Rationale –

- A dominant economic issue for producers is carrying stock through periods of feed shortage, usually the autumn-winter period in southern Australia
- Simpson and Richardson (2009) have estimated from recent evidence obtained in field bio-assays that seedling loss and root damage during this autumn-winter pasture could be resulting in between 25% and 58% loss of potential net farm income. Hence the potential economic return from research investment in reducing the impact of soil borne pathogens is significant. Much of this is “sub-clinical” in that visual symptoms are not obvious so the “loss’ is not recognised. There is also considerable historical evidence demonstrating large pasture growth responses to biocide applications (see companion paper on soil biology)

- A major investment is required by producers for pasture renewal but concerns over poor persistence of introduced pasture species can be a disincentive and significant cost to producers. It may be that persistence is linked to soil borne pathogens affecting seedling germination and growth, seed-bank maintenance as well as the resilience of pastures to grazing.

- Pasture plant roots compromised by root damage due to pathogens are likely to be less able to access and take-up soil released and fertiliser nutrients leading to nutrient-use inefficiency (this is a key MLA issue identified in the Feedbase Investment Plan and subject of a new project on phosphorus acquisition by plants)

- It is likely that root pathogens slow or prevent (possibly in parallel with compromised nutrient access/uptake) recovery of pasture roots and this slows regrowth of pasture herbage for livestock consumption. Grazing pressure and frequency may also interact with these responses. There is a need to know the pathogen, nutrition and grazing thresholds that best facilitate optimum recovery of pasture roots to underpin regeneration of grazing herbage for livestock consumption.

- There is a need to innovate practical management responses to biological constraints that can be deployed soon rather than relying on the traditional approach of breeding plants for resistance/tolerance to specific organisms which had a very long lead-time even with adequate financial support. Breeding should be seen as a valuable contributor to the holistic solution with longer-term payoffs but also needs to be supported with complementary agronomic research to optimise genetic advantages developed.

- The emergence of DNA assays for soil borne organisms, functional groups and plants (roots) provide a new set of research tools capable of accelerating discovery and measurement of key biological responses in soils. It is important to note that these assays must be calibrated to define the meaning and importance of the data measured.

- Use the project as an opportunity to appoint an early-mid career pasture focussed scientist (biologist) to help consolidate and integrate existing and new knowledge on soil biology, soil borne pathogens, beneficial soil organisms plus traditional and emerging technologies (including molecular assays) for measuring aspects of soil biology and plant health. Such a person could then ensure that the knowledge from retiring experts is retained as a platform on which to develop new knowledge.

Experimental Elements For Consideration

The project may comprise one or more of these elements or variants of them or even others not listed.

- Establish the genetic diversity in tolerance and resistance among current and emerging pasture legumes and grasses to the major root pathogens
- Evaluate the possibility of endophytes contributing to the control of soil borne pathogens. Consider companion planting – selected grasses with legumes and effects on soil borne pathogens. Consider beneficial organisms as well.
- Evaluate the potential for nutrients, other than P, such as K, S, Mg, Zn, Cu to reduce the impact of soil borne pathogens on pasture legumes.
- Evaluate interactions between beneficial rhizosphere organisms (perhaps also including mycorrhiza) and pathogen infection
- Stock-take molecular assays that have been developed for specific pasture soil borne organisms and specific plant roots. Contract development of new molecular assays to fill perceived gaps.
- Extend the pasture bioassay technique developed by Simpson and Richardson (2009) across agreed agro-ecological zones and pasture types to quantify the opportunity cost of lost pasture production due to soil borne pathogens. Use these sites to identify causal organisms and calibrate the molecular assays by identifying the level of pathogens that represent risk to plant yield loss.
- Develop dedicated experiments to explore the interactions between mineral nutrition of pasture plants, soil borne disease tolerance and the impact of grazing. Use molecular assays for plant roots to explore root recovery dynamics in relation to grazing and disease incidence.
- Use long-term historical trial sites, current research sites (with particular emphasis on mineral nutrition under grazing by both sheep and cattle) as experimental laboratories to explore the interactions between grazing, pasture type, mineral nutrition and soil borne diseases to gather knowledge on the scale, diversity, impact, variation and management options associated with potential pasture performance. Include some consideration of how to estimate the potential growth of pastures in various environments as bench marks of performance. Use this aspect also to collaborate

with other scientists and co-operator farmers as part of the extending the knowledge emerging from this program.

- Develop bioassays in association with the molecular and root assays to form surrogate tests for resistance and tolerance to soil borne diseases in relevant pasture species and emerging cultivars. Genetic diversity may be considered here as well.

The basis of this project is underpinned by the idea that healthy soils ► plant health ► animal health ► producer profitability

Alternate Suggestion (in brief)

Research and monitoring tools for soil biology parameters under pastures

Theme – develop diagnostic, monitoring and benchmarking tools for researchers, advisors and producers to measure key soil biological parameters (organisms and function) in response to pasture and soil management strategies.

Background –

- MLA's feedbase R,D&E investment plan is being developed in a series of concurrent planning activities aimed at defining investment priorities and project detail for MLA to consider. Some examples include:
    - *Plant Health* – a science review of biotic factors affecting pasture growth including foliar and root diseases, invertebrate pests, beneficial soil microbes (e.g. rhizobium) etc
    - *Legume Performance and Proportion* – suggesting some focus on benchmarking leading producer pastures; enhancing legume composition through manipulation via grazing, soil ameliorants and nutrition, herbicides; improving legumes in crop rotations - species and mixtures, seed ecology, weeds; better legume species and varieties – renovation, legume alternatives, establishment technologies; links with producer groups.
- There is potential for some overlap in project design to develop in this process and so perhaps the soil biology project could focus more on providing a linkage or support function perhaps with an aim to establish and calibrate diagnostic and monitoring tools and benchmarks for soil biological and plant health in pasture soils.
- This project suggestion provides for sharing relevant experimental sites and producer benchmarking properties for which soil biological health would be an important

contributor. It also anticipates establishing dedicated experiments at selected sites where warranted or specific researchable questions become clear.

- It is anticipated that data would be shared (under suitable arrangements) which will broaden the scope to develop, evaluate and calibrate chosen molecular assays across diverse pasture systems and management practices.
- In this case, the experimental objectives are less well defined (at the outset) and contingent on attaching to new (and perhaps existing) projects within the Feedbase R&D portfolio which would benefit from some investigation into soil biology. The synergy benefit could be to add value to each of the collaborative projects and also provide an opportunity to help support development of relevant measuring tools and benchmarks for soil biology parameters as suggested in this theme.
- Hence for the planning of a project such as this at the workshop, there should be a focus on what could be done in this context, what measurements would be most valuable and what experimental approaches would be required. Then as relevant new Feedbase R&D projects are defined, the definition of the soil biology contribution could be negotiated.

Rationale –

- The emergence of modern technologies to analyse DNA as a means to identify and quantify specific organisms in soil or around roots, measure root responses and measure soil biological functions for essential soil processes provide an opportunity to greatly accelerate the capacity to research soil biology under pastures.
- These assays, however, require significant calibration to determine meaning so that they can be used in research and monitoring contexts. To develop such tools will broaden and deepen the opportunity to create an investigative platform to better understand soil biology, its responses to soil and pasture experimental treatments and to implement workable tactics to favour soil biological health in a measurable way.
- Access to, and cooperation with, a diverse range of projects across contrasting grazing systems, pasture types, soils, environments will provide opportunity to gather relevant soil biology data for comparison, baseline establishment and measurement of treatment responses, calibration and validation of chosen measurements.
- Cooperation with a number of research projects will offer opportunity to educate researchers, advisors and producers on soil biological aspects of interest.
- It is suggested that the primary focus remain on soil biological constraints for the reasons indicated earlier although additional assessment of soil biological functional capacity for key processes and some examination of beneficial organisms may be warranted.

- A broad project such as this offers opportunity to develop expertise in diverse aspects of soil biology under contrasting conditions and so would provide fertile ground for post-graduate training in soil biology, plant pathology etc in relation to pasture systems.

Experimental Elements for Consideration

The project may comprise one or more of these elements or variants of them or even others not listed.

- Extend the pasture bioassay technique developed by Simpson and Richardson (2009) across shared sites to estimate the impact of soil borne pathogens in the different environments. Use this as an opportunity to "train" researchers and others to utilise the bioassay and interpret information from it.
- An example may be to test leading producer properties used for production benchmarking to determine if there are sub-clinical but significant opportunity costs due to soil biological constraints. In the developing legume stability project, a survey of legume production on leading producers' farms is proposed.
- Develop pathogen risk levels in pasture soils to establish trigger points for management action
- Develop benchmarks for soil biological diversity and function for key selected targets e.g. P, N and C transformations in relation to grazing system, season, environment
- Begin developing models of soil biological measurements in relation to pasture performance to help identify knowledge gaps and support extension activities.
- Provide a way to engage with leading producers across diverse systems
- Provide a training ground for PhDs – tools, knowledge, innovation.
- Develop expertise in pathology and ecology of soil borne organisms in pasture systems.
- Provide a pathway to "harvest" and consolidate information on soil health, plant health and pasture performance etc within a systems framework

Scenario 2 – Additional funds become available (quantum unknown!)

Should additional funds become available, the following projects (provided in brief overview) are suggested for consideration. Alternatives may be proposed.

Background

There are a number of focus areas that could be considered for additional funding with soil biology research:

- *Production* – infers research to enhance pasture productivity and producer profitability. This has been the focus of scenario 1.
- *Discovery* – infers basic research with high risk but aimed at “break-though” knowledge
- *Environment* – infers a focus on soil biological functions such as C sequestration and turn-over, mineral nutrient dynamics, impacts of soil biology on key environmental and sustainability parameters etc.
- *Delivery* – infers education of potential users of research outputs (researchers, advisors, producers) and may infer activities to adapt research outputs to conditions different to those in which they were developed.
- *Capacity* – infers building human capacity to grow industry knowledge and expertise to support future research and extension in soil biology as well mentor students in relevant disciplines.

It is expected that aspects of *Delivery* will be attached to any project that emerges for investment and that a delivery plan will be a key part of project milestones.

In scenario 1, reference is made to the need to build *Capacity* in the soil biology area and ensure knowledge and skills associated with contemporary and near-retiring expertise are retained and built upon.

Hence the suggestions for investments should additional funds become available are focussed on *Discovery* and *Environment*. Some ideas are offered in brief below for consideration, although it is expected other suggestions will emerge at the workshop.

Discovery

Some complementary research to the Scenario 1 project is suggested which will help develop under-pinning knowledge to support practical outcomes in minimising the impact of soil borne root pathogens on pasture productivity.

Suggestion – Mechanisms of clover and grass plant tolerance/resistance to soil pathogens

Some basic science is suggested aimed at defining and influencing mechanisms plants deploy to either resist (resist infection and build up) and tolerate (remain productive despite the presence) of pathogens on roots. This is a critical area of knowledge development which has the potential to underpin breeding programs for resistance/tolerance characteristics of new legume and pasture species; evaluate the resistance/tolerance of contemporary varieties, understand what management practices compromise resistance/tolerance and develop management tactics to favour enhanced resistance/tolerance.

This may require some focus on the dynamics of plant root exudates and the reaction of beneficial rhizosphere organisms which assist with reducing the impact of soil borne pathogens on plant performance.

Interactions – Plant Roots and Soil Microbes in the Rhizosphere	**Sought Outcomes**
• Basic research into understanding interactions at the root surface with clearly defined researchable questions of importance to plant productivity, disease resistance/tolerance and interactions with beneficial soil organisms.	• Effect of plant type and stress on rhizosphere exudates and interactions with soil organisms defined • Mechanisms of resistance/tolerance defined. • Plant selection/breeding traits for enhanced performance defined

Environment

Background

A rapidly evolving issue relates the Carbon Farming Initiative which seeks to manage greenhouse gas emissions from agricultural systems and foresees the prospect of soil carbon sequestration and abatement activities being considered in potential carbon trading markets. It allows for farmers to earn carbon credits by storing carbon and reducing greenhouse gas emissions which can then be sold to businesses wanting to offset their emissions.

The CFI scheme has now commenced. The government has proposed that the CFI would credit land sector abatement whether or not it is recognized towards Australia’s protocol targets (Kyoto or otherwise). Eligible abatement activities (relevant to soil management) may include:

- Emission avoidance – methane or nitrous oxide from soil, animal excreta
- Sequestration of soil carbon including biochar
- Reduced fertilizer use and manure management

Activities should:

- Take regional NRM plans into consideration
- Include the optional information on bio-diversity and other co-benefits.

Further to the above, soil carbon is an important contributor to soil health in Australian rangeland and improved pasture grazing systems. One of the major threats to sustainability of Australian grazing lands is the depletion of soil organic carbon (SOC) particularly where fertilizer inputs are not economically feasible.

Recent work in Australia (including MLA investment in the detailed spatial study of the Wambiana long term grazing trial and Soil Pasture Biology Program Final report 2008) points to contrasting responses of soil carbon stocks to grazing management which can be monitored at the paddock scale, and it shows a strong interaction between stocking rate and soil type on SOC stocks.

Soil carbon availability drives or constrains soil biological function in Australian soils. A range of soil carbon fractions can be measured (organic and inorganic) but it is usually recognised that turnover rates of soil organic carbon (SOC) pools, which is considered for C sequestration and soil health, impacts permanence, leakage and market based considerations. The spatial and temporal dynamics of carbon availability (seasonal, field-based, comparing management regimes, within pasture & pasture/crop systems) in different soil C pools, are linked to their impact on key soil biological functions such as nutrient mineralization and loss, microbial bio-diversity, pathogen survival, pesticide degradation and soil aggregate formation, and these differ depending on agro-ecological zone.

Driver (2011) has suggested an emphasis on the role and nature of soil borne organisms on ecosystems services in soils with particular reference to emissions mitigation, mediating nutrient availability and use-efficiency in addition to supporting pasture health and productivity. They, along with soil carbon, also contribute strongly to soil chemical and physical health.

In this context Driver (2011) suggests investment in soil science should focus on:

- “Improved capacity to measure and monitor soil condition as it relates to the capacity to sequester carbon or influence carbon dynamics and nutrient cycling and nutrient availability”
- “Support the development of standardised models for measuring soil ecosystem functions including emerging standards and technologies for soil and environmental metagenomics as they relate to agricultural land use”
- “Fit-for-purpose tools, models and predictive capabilities for soil function and resilience”
- “Contribute to the development and management of shared data sets and national coordination in areas of developing metadata standards for landscapes and land use”.

Suggestion –

There is clear inertia toward the CFI initiative and concerns about sustainable land management practices which focus attention on soil carbon dynamics and so the development of measuring and monitoring tools for soil biological function in relation to greenhouse gas emissions, carbon cycling and soil resilience under pasture systems is suggested.

Carbon and Nitrogen Cycles in Pasture Soils – Production and Environment	**Sought Outcomes**
• Use a combination of molecular microbial, soil function and chemical analytical techniques to assess pasture management, pasture type, soil type and season on: o The N and C cycles with special emphasis on greenhouse gas (N_2O, methane, CO_2) emissions, systems transformations, leakage and storage. o Fungi and bacteria microbial community diversity and function • Determine the temporal dynamics of carbon availability in association with key soil biological functions across agro-ecological zones, pasture systems and management practices	• Effect of grazing systems on soil biological functions defined • Grazing systems characterised for greenhouse gas emissions • Biodiversity of soil biota and functions evaluated • Knowledge developed to design and test ways to reduce emissions • Knowledge for enhanced C sequestration developed • Benchmarks for and monitoring tools for soil carbon and biota in providing ecosystem services.

Final Note – Purpose of the Workshop

The projects suggested above are described simply to stimulate thinking and contributions to ideas for the best investment that can be made in pasture soil biology of benefit to producers.

A workshop will be held on February 7th, 2012 (location to be confirmed) at which the objective will be to define specific project details within the allocated budget framework that MLA can accept and act on.

The purpose of the workshop is to discuss the proposals in this paper and agree, modify or if disagree then decide alternative proposals.

The objective is to design project proposal(s) in sufficient detail as to submit to MLA and possible co-investing partners for contracting and funding.

Identification of the key researchers and research groups that are most competent to undertake the work is also sought.

The project(s) should have potential to maximise the benefits to producers and lead to producer participation where practical and also emphasise strong collaboration and links to complementary research.

Those who wish to submit alternative ideas, please provide a project summary using the following format:

- ***Proposer Name and Organisation***
- ***Project Title***
- ***Purpose and Overview Description***
- ***Justification***
- ***Objectives***
- ***Experimental Details***
    - ***Background, rationale, justification***
    - ***Proposed methods (overview)***
    - ***Expected resources required (human, other, budget)***
    - ***Research group(s) who are most capable to do the work***

References

Driver F (2011) MLA Strategic Plan and National RDE Strategies – Soil Science Portfolio and Pasture Systems. Internal MLA Discussion paper.

Gupta VVSR and Ryder MH (2003) Soil Biology in Pasture Systems – Knowledge and Opportunity Audit. Meat and Livestock Australia project report BSC.007.

R J Hannam (2008) Pasture Soil Biology 2003-2008, Final Report. Meat and Livestock Australia project report B.SHP.0100.

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7.2 Soil Biology Background Paper

SOIL BIOLOGY RESEARCH FOR PASTURE SYSTEMS

Background Paper

Basic Concepts and Recent Research History

Introduction

Meat and Livestock Australia (MLA) has developed a new Feedbase Investment Plan to guide research investments to enhance productivity and sustainability of the Southern Australian red meat production systems based on pasture and complementary crop and shrub herbage feed sources.

“Soil Health” has been identified as one of a number of key theme areas for research in the investment plan as having the potential to contribute to optimum feedbase productivity and underpin livestock health and production. This broad theme may relate to many, often interacting, aspects of soils including physical, chemical and biological elements although “soil health” is often considered with emphasis on soil biological diversity and function.

MLA now seeks to consider the possible research opportunities in soil health, and soil biology in particular, and distil these into defined projects to be considered for investment.

This paper seeks to provide background information to support a workshop on defining new investments in soil biology research to project submission detail.

It contains some basic soil biology relevant to pasture systems for those not familiar with this in pasture systems and also summarizes the research conducted in recent MLA investments in soil biology research, what it sought to achieve, where it got to and what the thinking has been for project ideas to build on that initial effort.

The objective is simply to inform and perhaps broaden the thinking of those who attend the MLA sponsored soil biology planning workshop.

Some basic soil biology

The soil biota comprises a vast range of organisms ranging in size from the:

- microscopic such as bacteria and fungi (principal agents of nutrient cycling, plant root diseases and disease suppression);

- protozoa and nematodes (regulate bacteria and fungi populations and impact on nutrient mineralisation);
- through larger organisms such as mites and springtails (predators of smaller microbes and affect nutrient turnover);
- to larger organisms such as earthworms and dung beetles (fragment organic residues and help form stable soil aggregates).

These various groups of organisms are linked in a "food-web" which infers a predator-prey relationship between the organisms that regulates their relative proportion in the soil. Generally, soils with higher levels of organic matter inputs are both more active and have greater biological diversity than soils depleted of soil carbon.

Also, the total number of organisms, the diversity of species and their activity is affected by the soil condition induced by changes in factors such as soil moisture (daily variation and season), temperature, soil disturbance, soil ameliorants (e.g. lime), fertilizers, agricultural chemicals etc.

The commonly identified key functions of the soil biota include:

- mediate organic matter turn-over
- nutrient cycling within soils
- maintenance of soil physical structure
- plant disease transmission and suppression
- pollutant degradation or detoxification

It is important to note that soil borne organisms operate in at least two different domains:

- free living in the soil, collected in micro-sites rich in organic matter
- associated with a narrow zone close to plant roots (rhizosphere) in which micro-organisms interact with the release of organic materials from roots.

Soil biology in pasture systems

The level of understanding of soil biology under pastures is well behind that for mono-culture cropping systems due largely to the much greater complexity in pasture soil systems making it much more difficult to research. Some of the contributing factors to this greater complexity include:

- pasture swards contain mixed plant populations each contributing different forms of organic material (root exudates, roots and leaf/stem litter) to soil as food for the soil biota and so influence the temporal and spatial variation in soil borne organism populations and functions
- the impact of grazing animals in terms of the type of animal (sheep, cattle), the intensity and timing of their grazing, the nature and distribution of urine and faeces deposits on pastures and the physical impact on surface soils (e.g. compaction).
- The nature and composition of pastures vary considerably between agro-ecological zones, soil types and soil conditions such as pH and salinity which reduces the prospect of research findings being transportable broadly across pasture systems without validation.

Permanent pastures usually have no regular soil disturbance event (as do cropping soils frequently) and so carbon inputs from plant roots' litter and return of excrement are the major regulators of biological activity and turnover (succession) in soils under pastures. In crop-pasture systems, the major soil disturbance at the end of the pasture phase re-sets the microbial succession cycle as does soil disturbance associated with sowing new or renovating pastures.

Grazing has a particular effect on pasture soil biology because eating and trampling of pasture plant leaves and stems by livestock can, depending on grazing pressure, reduce carbon flow to roots for respiration maintenance and growth as well as symbiotic nitrogen fixation. Some root death may occur. The extent of root death, and their level and rate of recovery, is influenced by the intensity and frequency of grazing. Grazing also removes, or at least redistributes unevenly via manure deposits, leaf material that was potential organic matter resource for the soil biota near the grazed plants.

Another major factor influencing plant roots are root pathogens (mainly fungi and nematodes) which also destroy roots and/or reduce their ability to take up nutrients and water and so their productivity relative to growing season potential is reduced. Importantly, root pathogens can dramatically reduce the persistence of sown pasture species vulnerable to various root diseases. This alone can be a significant financial business risk to farmers seeking to sow new pastures, or renovate older ones.

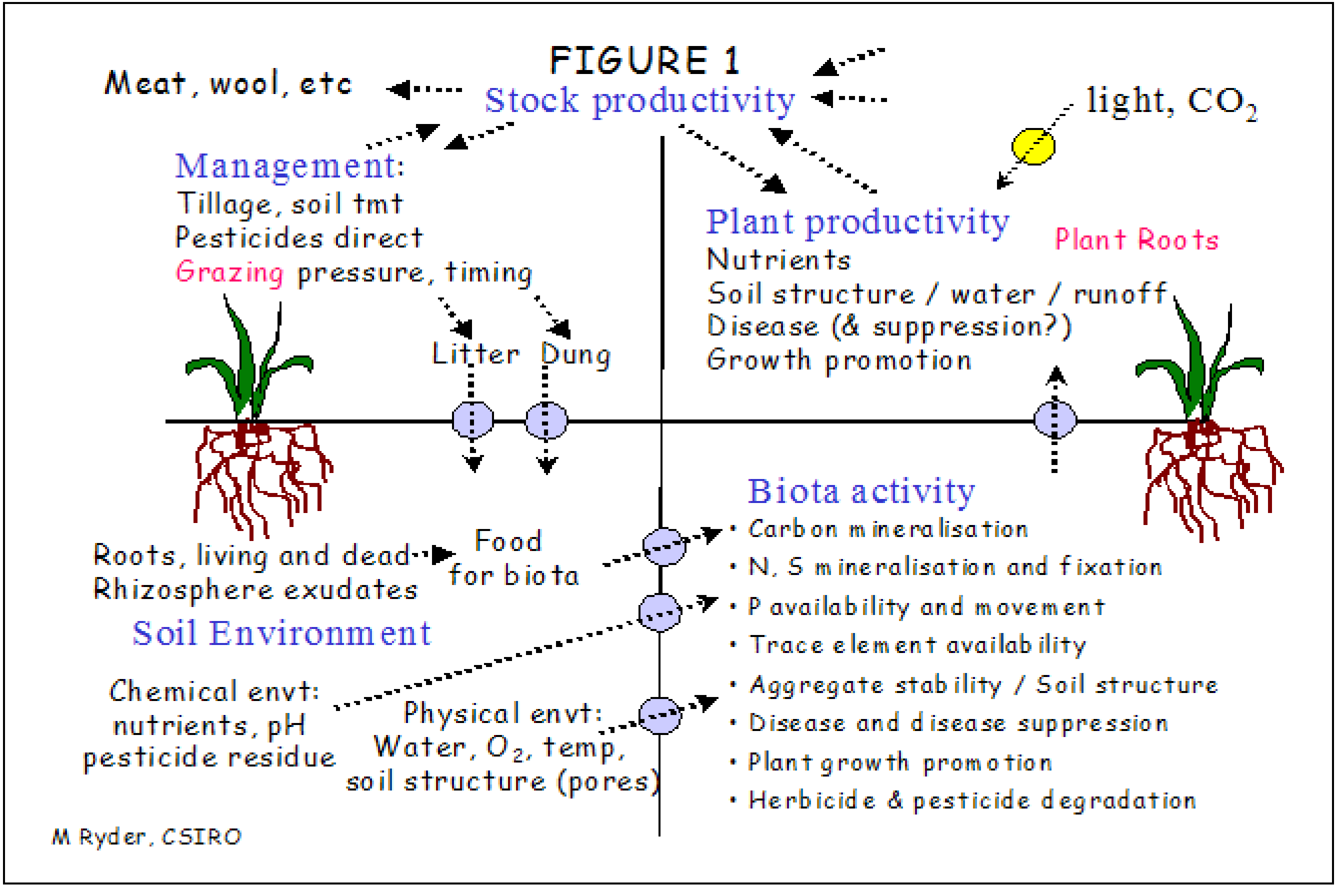
For example Simpson and Richardson (2009), using a unique field bioassay experimental technique, estimated that in clover-grass pastures root damage induced by root pathogens limited clover growth by 21-36% and ryegrass by 10-45% across a range of sites during the autumn-winter period which is a critical time of feed shortage in these systems. Also, these estimates translated into the conclusion that well managed pasture enterprises may be forgoing between 25 – 58% of potential net farm income due to seedling loss and root damage in their pastures caused by root pathogens.

It is also worthy of note that the rhizosphere surrounding roots is colonised by a range of bacteria (and fungi) which feed on the mucilage, soluble sugars, amino acids and discarded root cells released from plant roots. These produce compounds which can affect root and top growth as well as substances that can stimulate or inhibit other microbes, including root pathogens.

While soil biological activities under pastures contribute strongly to pasture plant productivity, they, along with soil physical and chemical conditions, also have important impacts on eco-system functions. Examples include:

- Movement of nutrients and pollutants such as agrochemicals through soils to ground water and across landscapes
- An accumulation of decomposing organic material exacerbates release of greenhouse gasses such as methane and nitrous oxide.
- Soil stability against erosion

A general overview of the complexity of soil biology activity, function and influences in pasture systems is shown below in a diagram developed by M Ryder (CSIRO) in Gupta and Ryder (2003):



The MLA Pasture Soil Biology Program 2003-08

MLA's most recent investments in pasture soil biology commenced in 2003 with co-investment from AWI and GRDC. The vision for this program was for it to be an initial phase of long-term investment in both applied and basic science with an overarching strategy to:

- Develop and validate research tools to investigate key aspects of pasture soil biology
- Use these tools to understand biological systems in pasture soils in relation to pasture management across agro-ecological zones
- Develop practical management solutions to manipulating pasture soil biology to enhance soil and plant health and productivity
- Develop knowledge and research capacity in light of the aging research expertise in key soil biological disciplines, particularly for pastures. Of particular interest was to develop at least one person who could absorb existing knowledge from the current experts and bridge that with modern molecular measurement and interpretation approaches and provide a focal point for future research and knowledge in pasture soil biology.
- Leave a comprehensive legacy of research and monitoring techniques supported by adequate knowledge to underpin and accelerate development of farmer-friendly soil health assessment tools and practices for managing pasture soil biology and plant health for improved productivity.

A brief overview of the nature, achievements and aspirations of the Pasture Soil Biology Program are outlined in the figures below:

- Figure 1 – provides an overview of the planned long-term strategy for pasture soil biology research and its key components as viewed at the inception of the program
- Figure 2 – summarises the key achievements over the first phase of the program.
- Figure 3 – provides an overview of the planned investment strategy for phase 2 of the program. The program was terminated at the completion of phase 1.

The aim of producing these figures is to simply provide an overview of what was previously identified as key research areas in pasture soil biology and what has been achieved to date with a view to inform consideration of how best limited research funds can be deployed in the future.

Figure 1

OVER VIEW OF PASTURE SOIL BIOLOGY RDE STRATEGY - 2003

OBJECTIVE: ***Soil Biological Health for at least 10% Improvement in Pasture Productivity***

Applied Research

Characterise & quantify soil biological constraints

- Identify dominant pathogen and beneficial organisms
- Quantify pasture productivity constraints
- Use nutrition and biocides to estimate water use efficiency potential
- Cover main pasture agro-ecological zones
- Use field sites to link applied and basic research.

Evaluate pasture management strategies on critical soil biological functions

- Pasture management vs shifts in soil biota indicators.
- Consider all macro-management options such as grazing, pasture species, nutrition, soil ameliorants, tillage, irrigation, weed management etc

Manipulation of key biota drivers for improved pasture productivity

- Test pasture ameliorants and management strategies to drive positive shifts in soil biota. Based on above research.
- Aim for disease suppression and optimum nutrient turnover.
- Develop disease control measures

Manipulate soil biological processes for successful pasture-crop-pasture transitions

- Test intervention strategies as indicated in basic studies.
- Emphasis on setting up crops for transition to pastures

Strategic Basic Research

Develop molecular assays as research tools to study soil organisms important to pastures

- DNA based tools to identify and quantify populations of beneficial and detrimental organisms.
- Develop rapid test for root ID, function and composition analysis

Understand soil biology at the root surface

- Pasture root rhizosphere conditions vs population shifts in beneficial and detrimental organisms
- Pasture management effects on root rhizosphere condition and biota populations

Understand soil biological processes

- Free living soil and rhizosphere organisms in nutrient cycling and uptake.
- Soil biota role in chemical breakdown
- Soil biota role in amelioration of acid soils

Understand critical biological processes for successful pasture-crop-pasture transitions

- Characterise soil biological conditions for optimum transitions between crop and pasture phases

Planned Outcomes

- Rapid assay techniques for diverse soil biological groups – research tools
- Basis for developing diagnostic tests for key organisms or groups for producers to use
- Consolidate pasture pathology expertise
- Soil microbiology capacity building

- Reporter organisms or compounds to aid decision support on critical pasture management strategies.
- Basis for developing diagnostic tests for disease suppression.
- Basis for developing root disease control strategies.
- Basis for seedling survival strategies in pasture establishment

- Nutrient retention for efficiency and reduced environmental impacts.
- Match nutrient availability to plant demand.
- Reduced agrochemical pollution
- Potential natural acidity management

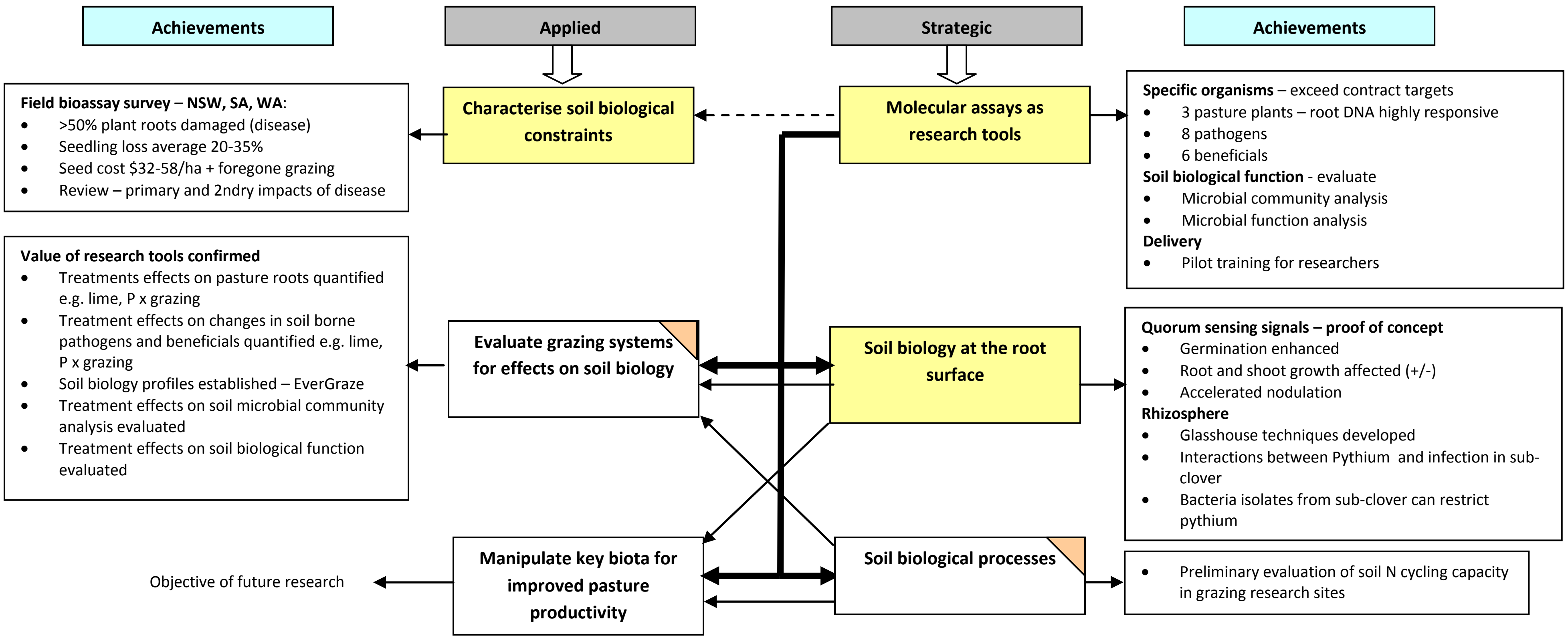
- Reduced risk in pasture-crop-pasture transitions.

Yellow indicates initial investment strategy

Figure 2

PASTURE SOIL BIOLOGY STRATEGY PHASE 1 - PROGRES 2003 – 2007

OVER-ARCHING STRATEGY – ENDORSED BY REVIEW

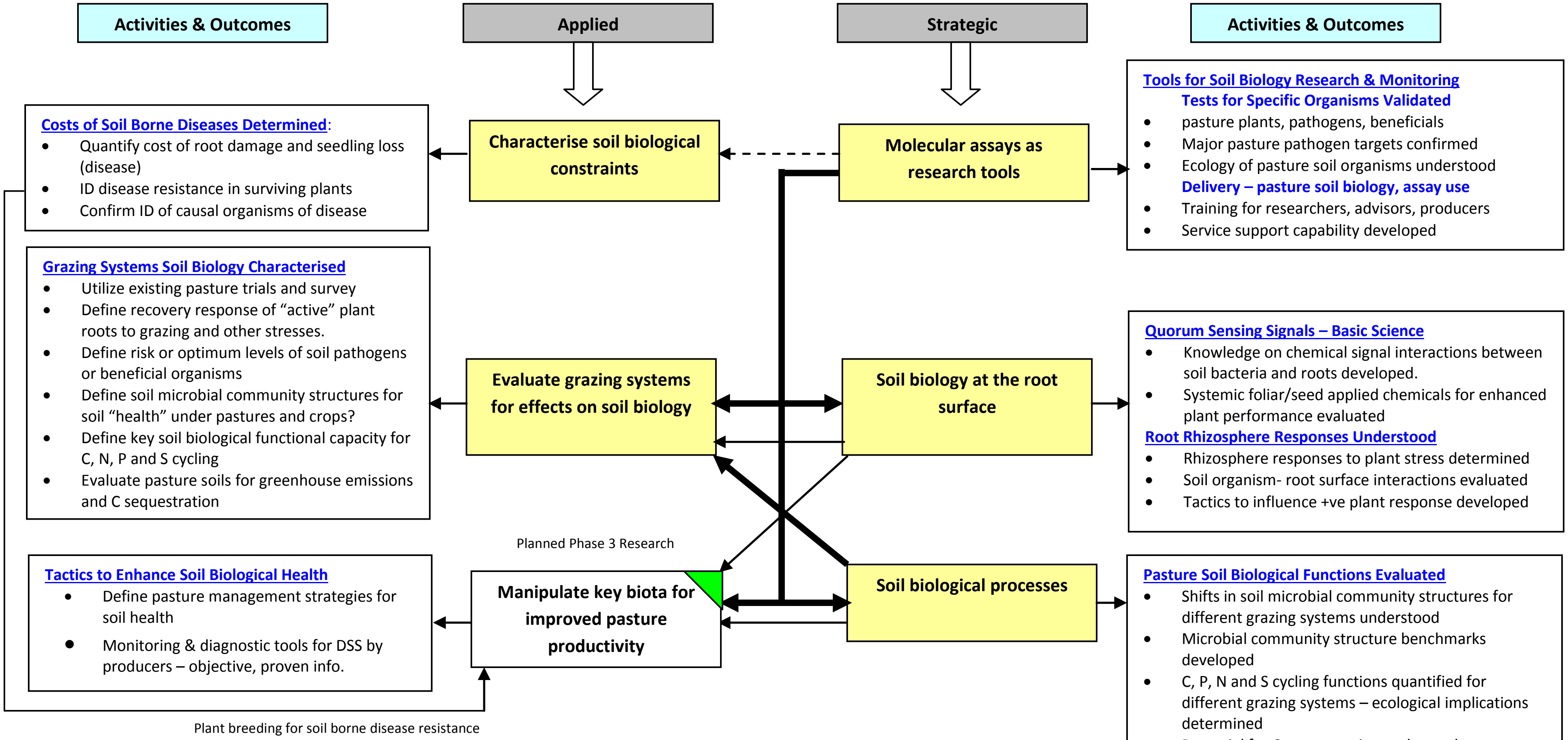


Notes:

- Over-arching investment strategy was endorsed by a program review by Professor J Irwin and Dr A Rovira
- Activities depicted in [yellow box] areas were initial investment priorities. Activities have begun in other areas [orange corner] ahead of strategy.

Figure 3

PLANNED ACTIVITIES & OUTCOMES 2007 – 2012 – Phase 2



Notes:

- Activities depicted in [yellow box] are phase 2 investment priority areas. Activities planned to begin – [green triangle]
- Over-arching Strategy – **develop research tools ► understand systems ► develop practical management solutions**
- Key Benefit – vastly accelerated understanding of pasture soil biology to capture productivity gains from improved soil health

A review of the above program (Irwin and Rovira, 2007) endorsed the overall research strategy of a combination of applied and strategic research in the areas identified. Their key recommendations were:

- That the work on DNA probe development and validation (understanding what the measurements mean) continue for key root pathogens along with the expansion of the field bioassay work and assisted with the appointment of a plant pathologist.
- That the validation of the DNA probes continue to make optimum use of long-term pasture management trials
- That the strategic elements of the program continue with particularly with reference to:
    - Quorum sensing signals between plant roots and soil biota with more focus on beneficial growth promoting and disease suppressing rhizosphere bacteria
    - Release of organic compounds from the roots in response to grazing.
- That a new area of strategic research be considered on the diversity of soil biological functional groups involved in carbon turnover. Also that the validation of a bioassay for soil health developed by M Ryder (CSIRO) be supported.
- That due to a shortage of trained pathologists relevant to pasture soil biology research, two PhD scholarships in the field of pasture soil biology be supported

To further inform consideration of what might be appropriate research activities in future pasture soil biology research, below is a brief outline of a range of project ideas which were identified in a business case developed for MLA in 2008 for consideration for investment in phase 2 of a soil biology program. Please note that strong co-investment from a range of stakeholders was envisaged to add to any direct MLA investment.

The project ideas are categorised in terms of productivity, discovery, environment, delivery and capacity building viz:

Project Suggestion	Expected Outputs/Outcomes
Production	
Ecology & Management of Root Damage in Pastures • Field bioassays across numerous sites using the DNA assays to identify causal organism and also help calibrate the level of pathogens that represent risk of plant yield loss	• Extent and severity of pasture yield loss defined • Seasonal soil biology dynamics characterised • Management tactics for pasture/soil health defined
Molecular Assays as Research Tools – Production • Development and validation of assays for organisms and plant roots important in pasture and pasture/crop systems – correlate with factors affecting production and environmental impacts • Evaluate assays for general soil biology parameters which relate to soil health • Educate and engage with researchers on their use in research and monitoring	• Molecular assays for pathogens, beneficial organisms and roots developed and validated • Treatment effects on soil biology in field experiments (new and established) defined • Knowledge on application of assays developed • Researcher education
Lucerne establishment and productivity – Mallee and contrasting areas • Resolve the risks of establishing Lucerne phases in cropping systems which recent surveys have shown to be affected by soil borne pathogens affecting establishment, survival and productivity. • Place some emphasis on management to optimise crop-pasture-crop transitions in mixed farming systems.	• Impact of disease on Lucerne establishment quantified • Pathogens identified for DNA assays • Lucerne lines evaluated for disease resistance • Lower risk of Lucerne establishment

Root Growth in Difficult Soils - PhD • Use the molecular assays for plant roots to study how plant roots penetrate hostile soils • Link to plant improvement programs to evaluate emerging varieties for better root penetration and survival in these soils – feedback to breeders on important traits.	• Fundamental knowledge on root behaviour in difficult soils vs management • Adaptation of plant varieties for rooting depth • New root research tool proven and available
Beneficial Endophytes to Protect Pasture Plants from Soil Borne Disease • Identify systemic endophytes as seed treatments for pasture seed to confer early vigour and resistance/tolerance to soil borne pathogens. Consider foliar applications to established plants • Link with research into chemical signalling in the rhizosphere of target plants	• Beneficial systemic and beneficial organisms identified to control seedling disease • Practical disease control measure to reduce the risk of pasture establishment
Soil Ameliorants – Impacts on Soil Biota -PhD • Evaluate conventional and unconventional soil ameliorants (e.g. lime, charcoal, “organic” lobby recommendations etc) on soil biota as measured with the molecular assays.	• Fertilisers and “organic” soil ameliorants objectively evaluated for soil biology and pasture effects • Objective info to provide advice to producers

<table>
<tr><td colspan="2">Discovery</td></tr>
<tr><td>Interactions – Plant Roots and Soil Microbes in the Rhizosphere
• Basic research into understanding interactions at the root surface with clearly defined researchable questions of importance to plant productivity, disease resistance/tolerance and interactions with beneficial soil organisms.</td><td>• Effect of plant type and stress rhizosphere exudates and interactions with soil organisms defined
• Plant selection/breeding traits for enhanced performance defined</td></tr>
<tr><td>Quorum Sensing Signals
• Basic science investigating the signalling between plant roots ad soil microbes with a view to identify systemic signalling chemicals which can benefit plant performance and resilience.</td><td>• Beneficial systemic signalling chemicals identified for plant health
• Novel plant treatments for enhanced production and resilience determined</td></tr>
<tr><td colspan="2">Environment</td></tr>
<tr><td>Nitrogen and Carbon Cycles in Pasture Soils – Production and Environment
• Use a combination of molecular microbial, soil function and chemical analytical techniques to assess pasture management, pasture type, soil type and season on:
o The N and C cycles with special emphasis on greenhouse gas (N_2O, methane, CO_2) emissions, systems transformations, leakage and storage.
o Fungi and bacteria microbial community diversity and function
• Link with the effort on production and interactions with other organisms.</td><td>• Effect of grazing systems on soil biological functions defined
• Grazing systems characterised for greenhouse gas emissions
• Biodiversity of soil biota and functions evaluated
• Knowledge developed to design and test ways to reduce emissions</td></tr>
<tr><td>Carbon Inputs & availability in Pasture Systems
• Place special emphasis on micro-ecological studies of carbon turnover, microbial community analysis and soil health in relation to pasture systems and pasture-cropping systems. Objective to develop basis for soil health benchmarks.</td><td>• Temporal dynamics of carbon availability defined and linked to soil biological function, ecology and management
• Effects of grazing systems on C sinks and leakages defined</td></tr>
</table>

Delivery	
Researcher Outreach – Support for Molecular Assays • Provide a pool of funds for researchers of current projects to access to begin using the molecular assays within their project for which existing budgets are prohibitive. Use the fund to leverage engagement by researchers with the technology.	• Pool of funds for researchers to use DNA assays • Mechanism to link researchers to soil biology research
Producer Engagement • Design and implement small projects in partnership with producers with specific topics of interest in pasture soil biology. • Link to PIRD projects • Contrasting pasture systems • Link to field bioassay project	• Producer involvement in objective soil biology research • Producers educated in soil biology and new technology • Producer advocates developed
Capacity	
Pasture Plant Pathologist – Soil Biologist • Engage an early career pasture pathologist to develop skills in traditional and molecular aspect of soil pathogens in soil and also develop skills in general pasture soil biology • This position should offer a significant career prospect and should be seen as a key plank in developing research capacity in this area for the next 10-15 years.	• Essential capacity to develop research into soil biological constraints and soil biology • Resource for all projects • Industry focal point for progressing soil biology and pathology research and extension.

Also see Appendix 1 for a summary of the suggestions by Gupta and Ryder (2003) developed in a knowledge and opportunity audit commissioned by MLA as a basis of planning their pasture soil biology research program.

Additional information

As part of the planning for the initial Pasture Soil Biology Program, MLA commissioned a knowledge and opportunity audit as a means to identify research possibilities in pasture soil biology (Gupta and Ryder 2003). A summary of their major recommendations is as follows:

(Priority in bold, marked*)

A. Setting the scene: diagnosis of problems

A1*. Diagnose pasture productivity constraints using **Water Use Efficiency** measurements to compare actual pasture productivity to **potential productivity**, and determine the nature of the constraints (both for research and on-farm).

NOTE: This work will set the target for pasture productivity and enable quantification of opportunities to improve current levels of pasture production.

Where current productivity is below potential and where the nature of the constraints is not known, ascertain whether the primary limitations are physical, chemical or biological. If there are major biological constraints, specific causes and treatments should be investigated.

Methods: fumigation and deep ripping trials, corrected for nutrition; measuring water use through soil profile.

A2*. Determine the likely positive contribution of changing management of the system (e.g. increased carbon and other inputs) in achieving potential pasture production, via improved soil biological functions such as nutrient cycling, nutrient use efficiency and disease suppression.

NOTE: This work may require several growing seasons, but will provide results that **cannot** be obtained via research towards **Recommendation A1**. In contrast to the identification of major constraints, which can often be done in one growing season.

B. Driving soil biological activity

Carbon inputs & availability

B1*. Soil carbon availability drives or constrains soil biological function in Australian soils. Determine the temporal dynamics of **carbon availability** (seasonal, field-based, comparing management regimes, within pasture & pasture/crop systems) and link this to key soil biological functions. Depending on agro-ecological zone (e.g. mallee cf western slopes) the key biological functions will differ (nutrient mineralization and loss, pathogen survival, pesticide degradation, soil aggregate formation). Management regimes include grazing systems, pasture composition, pasture renovation and soil ameliorants.

B2. Determine the role of grazing management in carbon dynamics, in relation to soils as carbon sinks and global greenhouse gas budgets. (Research in the USA, for example, is far ahead of Australia, and we cannot "import" these results).

Water availability & temperature:

B3. For key soil microbial processes (e.g. mineralization of N or development of disease suppression), define the number of microbially-optimal days based on conditions (soil type, soil moisture & temperature, based on weather data) found in different agro-ecological zones.

Aim: to provide information for prediction of soil biological function (**Link to A**).

C. Grazing management

C1*. Plant re-growth can be used as an indicator for resumption of grazing. Test the link between plant re-growth and key soil and rhizosphere biological processes. Does this method of scheduling grazing also deliver greater benefits from soil biological activity? (i.e. long-term sustainability of soil biological processes such as nutrient cycling). (Applied research).

C2.* *Appropriate grazing pressure could stimulate pasture re-growth via rhizosphere biological processes*.
Investigate rhizosphere processes over a range of grazing pressures that may lead to positive feedback on microbial mineralization of nutrients and their availability to plants (resulting in more rapid pasture re-growth). (Basic research).

C3*. Different grazing systems and grazing pressures lead to differences in soil biological functions (e.g. proportion of beneficial to deleterious organisms, nutrient cycling, disease expression or disease suppression). Determine the links between the composition and activity of soil biota under different grazing systems (linking soil biodiversity to function). (Basic research).

C4.* Determine the impacts of grazing management on soil food web dynamics, importance of various trophic groups in different agro-ecological zones and links to biological functions (dryland, lower rainfall). For example, food web dynamics in relation to synchronization of nutrient mineralization with plant demands. When plant demand is low, nutrients can be lost. If demand and supply of nitrogen do not match, excess accumulation of N could contribute to soil acidity problems.

C5*. *Minimize nutrient losses from grazing systems, especially in higher rainfall areas.* (Inappropriate grazing pressure could result in nutrient loss from the system via denitrification and by leaching).

Determine the role of grazing pressure in soil nutrient loss (especially N, but also P in high rainfall areas). Determine regulators of biological nutrient mineralization and loss, as affected by pasture composition: how can soil biota activity minimise the loss of nutrients from pasture soils?

Aim: recommend grazing management to improve efficiency of resource use & reduce off-site impacts.

C6. Investigate the relationship between carbon inputs (seasonal, above- and below-ground) and biological processes associated with soil nutrient cycling within a field-based grazing system.

Aim: to determine the balance between input of nutrients versus nutrients provided by soil biological activity, to gain more benefit e.g. mineralized nitrogen, also P availability in calcareous soils.

D. Management of pasture – crop transitions

Management of the transition from crop to pasture:

D1*. Seedling establishment and growth are important in the establishment of pasture and are affected by, for example, pathogen and nutrient status of soils. Develop measures of the status of key soil biotic activities in the transition from crop to pasture, to determine the impact of cropping phase management (e.g. nutrient cycling and availability, disease suppression). This is to provide information for farmers to maximize benefits from improved pasture soil biology.

Management of the transition from pasture to crop:

D2. Develop methods to evaluate a pasture soil prior to the next crop. Pastures provide a biologically-based benefit for the next cropping phase. For example, knowledge on disease potential, disease suppression potential, nutrient supply potential and soil aggregate stability at the end of a pasture phase will assist in deciding management practices for the next crop.

E. Soil-Borne Pasture Diseases: their Diagnosis and Control

E1*. Determine the major soil-borne plant pathogens for non-legume pasture plants (mainly grasses, including perennial and native grasses; region-specific; following from diagnosis of biological constraints). (Link to **A1**, determining constraints for pasture production).

E2*. Develop and deploy disease control measures, including chemical and biological treatments, for major soil-borne pathogens. Field-testing and assessment of potential for commercial development of bio-agents that induce systemic resistance to disease in pasture legumes.

E3*. Develop diagnostic DNA probes for the most important pathogens (fungi, nematodes; new research tools and methods for on-farm management of diseases). To be useful, this must be linked to information on the effects of environmental factors on disease expression.

E4*. Investigate the potential for development of disease suppression by promoting native microbial communities in pasture soils (i.e. control of disease by soil biological and/or physical factors while pathogen is present).

F. Removing negative impacts: Pesticides and pollution

F1*. Establish the capacity of soil macrofauna such as dung beetle species to reduce pollution of water by pathogens and organic material (carbon and nutrients) that move from pastures into water catchments. (Determine compatibility with agro-chemical use).

F2. Determine the effect of agrochemicals on specific biota e.g. effect of anthelminthics on soil macrofauna, aiming to minimise collateral mortality.

F3. Determine the effect of new generation herbicides on plant disease expression and nitrogen fixation.

G. New Management options: System inputs and Pasture renovation

G1*. Determine the full beneficial effect of pasture fertilizer inputs on soil biological activity (extent and duration of change in biological function), both directly and via improved plant growth. Consider this research alongside determining the benefits of greater carbon inputs.

G2. Pasture renovation to overcome soil compaction problems: compare the effect, benefit and cost of two contrasting approaches, i.e. soil physical disturbance versus changed grazing management, in different regions, for their ability to improve pasture soils and pasture productivity, especially via macrofauna activity.

NOTE: In addition to established methods in soil biology, the application of new tools to investigate soil biota and their activities will be valuable for progress. The use of these tools, based on advances in molecular biology and biochemistry, should aim to contribute to the research goals and priorities suggested in this report.

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7.3 Project Specification – Scenario 1

PART 2: FULL APPLICATION FORM

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Effective Management of Soil-borne Disease Threats to Southern Australia Pastures

Background of Research Work

1. Purpose

The recent Soil Biology Workshop held in February 2012 highlighted both the need and opportunities to curtail current severe losses from soil-borne diseases (and as outlined in detail in Appendix 1 attached) such that legume based pasture productivity across southern Australia is greatly increased. It was agreed at this workshop that this would best be achieved by focusing on the managing soil-borne diseases in the main legume component, subclover, and hence the focus on subclover in this application. This proposal addresses the producer outcome to "Increase feed productivity and sustainability". Specifically, this project targets improved management practices associated with grazing systems that effectively manage soil-borne diseases to enhance production efficiency and profitability for all livestock producers across southern Australia.

2. Critical feed shortages coincide with period of most severe soil-borne disease

Crucially, meat producers inherently face critical feed shortage in across the autumn-winter months, coinciding with the main soil-borne pathogens attack and consequent damage from massive pre- and post-emergence damping-off and seedling root disease, inflicting well documented losses up to 40-45% in the most disease prone areas; consisting up to 30-90% of seedlings failing to emerge, 10-40% of surviving seedlings succumbing to post-emergence, and with a 35-70% reduction in growth of survivors (see Barbetti et al. 2005, Barbetti and Jones 2011, Barbetti et al. reviews of 2005, 2007 and MLA report of 2005 by Barbetti et al.). This feed shortage is likely the major constraint to increasing livestock production and profitability. Any increase in extra carrying capacity developed during this autumn-winter period from improved disease management will not only improve overall carrying capacity but also reduce the current high degree of uncertainty faced by meat producers in being assured of sufficient feed over this period. Perhaps of greatest significance is the well documented rapid deterioration and lack of persistence of subclover pastures across southern Australia following attack by soil-borne pathogens (Johnstone and Barbetti 1986; Barbetti et al. 2005, Barbetti and Jones 2011, Barbetti et al. reviews of 2005, 2007 and MLA report of 2005 by Barbetti et al.). The combination of soil-borne disease particularly targeting and restricting seedling emergence and growth, along with poor performance of surviving plants throughout the remainder of the season (O'Rourke et al. 2009), results in severe decline in seed banks depriving pastures of the ability to persist, further increasing both the risks and opportunity costs and removing any incentive to invest in pasture improvement.

Recently, Simpson et al. (2011) used field-based plant bioassays at 17 locations across southern Australia to show that 9-93% of subclover seedlings failed to emerge at 14 locations, and with post-emergence losses ranging up to 32% and moderate to severe root disease on any surviving plants at all sites. Prior to this research, there were many studies to demonstrate the impact of soil-borne diseases across southern Australia where root rot has seriously adversely affected the production from subterranean clover pastures over at least the past 4 decades (e.g., Johnstone and Barbetti 1986; Barbetti et al. 2005, Barbetti and Jones 2011, Barbetti et al. reviews of 2005, 2007 and MLA report of 2005 by Barbetti et al.) and a summary of these impacts across southern Australia is detailed Appendix 1, Section 2. Conservatively, the combined annual losses from major soil-borne diseases of legume based pastures across southern Australia likely exceed $400 m annually, the majority of this being in relation to subclover pastures (Barbetti, unpubl.).

3. Most important soil-borne pathogens

Pre-emergence damping-off in subclover caused by *Pythium irregulare* and pre- and post-emergence damping-off combined with severe root disease in subclover pastures caused by *Phytophthora clandestina* constitute the over-riding biotic constraints to productive subclover pastures across southern Australia. A range of other pathogens are also very important and damaging, including one or more species of *Aphanomyces, Fusarium*, *Phoma* and *Cylindrocarpon*, with *Aphanomyces trifolii* recently being highlighted as pathogen of corresponding importance to *Pythium* and *Phytophthora* (O'Rourke et al. 2010). Soil-borne pathogens operate in various interacting combinations and complexes with each other to maximise damage to plants, and understanding these interactions is critical both to interpreting molecular DNA tests and development of successful control strategies.

4. Need for further quantification of impact of soil-borne diseases

Except for the recent studies by Simpson et al. (2011) and O'Rourke et al. (2009), the impact and related causes of severe disease have been largely ignored for some decades. This is despite field observations indicating that the situation in terms of relevant pathogens (e.g., addition of *Aphanomyces trifolii*) and disease expression has changed dramatically over recent decades highlighting the need for a more relevant/recent reassessment of incidence and impacts (losses and economic importance) of major soil-borne diseases in subclover across southern Australia. Such information is fundamental both to defining regions and situations of highest priority for management intervention and research priorities for subclover pastures. It is evident that many subclover pastures have and continue to fail to persist or be productive, particularly in high rainfall regions >500mm in situations where annual losses up to 40-50% occur.

5. Opportunities for managing soil-borne diseases from altered farm practices

i. Chemicals

There exist significant but rarely exploited opportunities for utilizing low cost chemical seed treatments to ensure successful stand establishment when sowing pastures (see MLA reports of Barbetti et al. 2005 and Barbetti and Jones 2011; Barbetti et al. reviews of 2005, 2007). For example, Taylor et al. (1983) demonstrated increases in Victoria from disease control by fungicides of nearly 60% in May and more than 95% in October in subclover herbage production. There are many cheap potential fungicide treatments that target a wide range of soil-borne pathogens associated with damping-off that would be appealing for widespread uptake and usage by producers. These not only include simple and cheap fungicidal seed treatments that could ensure successful stand establishment of pasture legumes, but also foliar applications to protect roots in established seedlings, together ensuring and/or re-establishing pastures that persist. Benefits would be extensive particularly in the high rainfall regions (>500mm), where pre- and post-emergence damping-off by *Pythium* and *Phytophthora* alone currently result in pastures sometimes failing to persist or be productive and where annual losses of up to 40-45% losses occur in the most disease prone areas.

ii. ***Farm cultural practices***

There are several potential yet rarely exploited opportunities for utilizing manipulation of cultural practices for soil-borne disease management in pasture legumes (see MLA reports of Barbetti et al. 2005; Barbetti and Jones 2011; Barbetti et al. reviews of 2005, 2007). While the low level of utilisation of cultural management measures by meat producers arises partly from lack of awareness of their benefits, this is also partly a consequence of additional measures not having been identified or evaluated for their potential benefits. This is despite historical information clearly suggesting roles for practices such as controlled grazing (e.g., can reduce losses from *Phytophthora* root rot in subclover by up to 55%) and soil disturbance (e.g., can greatly reduce levels of damping-off and root disease in subclover for up to three years).

iii. ***Nutrition***

Soil fertility affects the severity of disease on subclover, by influencing root physiology and host resistance. Better utilization of applied fertilizer and vastly improved management of root diseases of subclover have been demonstrated from utilising nutrient amendments. There is significant potential for improved management of major soil-borne pathogens of subclover from correction of common nutrient deficiencies. This is highlighted by current research at UWA that demonstrated reductions in root disease of the order of 27-45% were obtainable from improving the nutritional status of soils under subclover pastures. There are significant opportunities for such practices to be integrated into a sustainable approach to successfully manage root disease in subclover pastures (O'Rourke et al., unpubl.).

6. Environnemental influences on soil-borne pasture diseases

Environmental factors such as rainfall (soil moisture) and soil temperature have a marked effect on both the root disease severity on subclover from individual pathogens and on the interactions that occur between different root pathogens. Understanding differences in root disease symptoms and severity that occur between different areas across southern Australia as a consequence of fluctuating soil temperature and moisture conditions (particularly from influence of climate change) is fundamental to relevant interpretation of SARDI molecular DNA tests. Environmental differences across southern Australia determine distribution of the most important soil-borne subclover pathogens such *Phytophthora* and its races (You et al. 2006) and *Pythium* (Barbetti unpubl.). Environmental factors are major factors affecting both the root disease severity in subclover from individual pathogens and on the complex interactions that occur between the different root pathogens.

7. Why address soil-borne disease constraints to pasture productivity

Mitigating soil biological constraints to pasture productivity will result in very significant increases in pasture persistence, pasture production both in quantity and reliability of feed production, and nitrogen fixation. Further, Barbetti (2007) highlighted that reducing the impacts of soil-borne disease enhances efficient utilisation of pasture inputs; pasture persistence; pasture palatability; plant nutritional value; seed set and viability; and pasture composition in favour of legumes and enhances resilience to grazing. Please note that while it is acknowledged that a desirable long term approach to minimise losses is by understanding the mechanisms by which pasture plants resist infection and colonisation of plant roots, and by improving our understanding of the genetics of resistance in desirable pasture species, such an approach is a much longer term proposition and outside the scope of this proposal.

8. Securing future pathology skills for pastures

Currently, Barbetti remains the last remaining full time pasture plant pathologist in Australia with recognised expertise in soil-borne pasture diseases and with combined competency skills across the range of fungal and oomycete soil-borne pathogens involved. He has four decades of successful IDM outcomes and package delivery for the benefit of Australian pasture-based industries across the disease spectrum of annual and perennial legumes and grasses for southern Australia. This expertise will be lost once Barbetti retires in ~ 5 years' time. This project offers the unique opportunity to train a pasture pathologist by employment of a postdoc pathologist as the main researcher on this project. In this way, existing pasture pathology knowledge of Barbetti will be captured in the most cost-effective way (i.e., no additional cost to MLA).

9. Project management

UWA will be the lead organisation and take full responsibility for project management and reporting. UWA will subcontract to SARDI for molecular components as outlined and other government agricultural organisations and/or with extension consultants as required for assistance with some field aspects outside of WA and as detailed in this application.

Project Description

1. How this project arose

From the Soil Biology Workshop held in Melbourne on 7th February 2012, the following five areas were agreed as the main objectives needing to be addressed, viz:

1. Determine the farm management factors that influence the expression of soil-borne root diseases in subclover pastures.
2. Evaluate practical management techniques aimed at reducing the pasture productivity loss during autumn-winter induced by soil-borne root disease.
3. Characterise the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots.
4. Broaden the quantification of pasture productivity constraints induced by pasture soil-borne root pathogens across agro-ecological zones.
5. Develop molecular assays to fill gaps in experimental tools needed to adequately conduct research into, and monitor, soil borne diseases in pastures.

Objectives 1-4 will be directly addressed as outlined below by this project application. The field plant disease and pathogen samples needed in order for Objective 5 to be addressed will all be supplied from experiments addressing objectives 1-4 above undertaken through this project. All plant, pathogen and soil samples and relevant information needed by SARDI will be sent to SARDI who will then develop a new molecular DNA test for *Aphanomyces trifolii* for which there are currently no molecular DNA tests developed, to address Objective 5. This is essential if the molecular DNA tests are to have the capacity to meet the requirements in terms of both research and monitoring in relation to soil-borne pasture diseases. Estimated cost of $50,000 for the development of this new molecular DNA probe has been budgeted in this project in year 1, as have been the costs for delivery and molecular analyses of all other field samples sent to SARDI at the appropriate stages for other stages and components of this proposal. In this project, there will be an emphasis on field experimentation, but with supporting glasshouse techniques utilised as appropriate and as detailed in the methods section.

Further, the postdoc pathologist employed on this project will receive hands-on training across all aspects of dealing with soil-borne pathogens and the diseases they cause, expertise that will not only be available to this project, but expertise from this postdoc pathologist and Barbetti will be available to related MLA/AWI projects, projects that desperately need competent pathology support (e.g., Simpson project B.PUE.0103 “Root Disease Constraints in Pasture Productivity”; SARDI DNA testing; etc).

2. What we will do in this project
To meet the above 5 research objectives, this project will:

i. Focus on developing practical short term on-farm measures that producers can readily adopt; measures that will not only provide producers with real and practical options to curtail current losses from soil-borne diseases, but with options that provide producers adequate flexibility in choice of control options they can utilise, and while taking into account livestock prices, potential productivity of pasture(s), degree of feed shortage, etc. It will demonstrate cost-effective chemical seed treatments for effective control of pre- and post-emergence damping-off and seedling root disease in subclover and other legumes when sowing or re-sowing pastures. Further, it will identify and demonstrate the benefits to meat producers from modified cultural (i.e., on-farm management) practices (e.g., cultivation, grazing, manipulation of pasture composition, etc) for reducing severity and impact of major soil-borne diseases in pastures across southern Australia. *(Objectives 1 and 2)*

ii. Focus on defining the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots and the influence of environmental factors on disease expression. The different symptoms on roots and the relative impacts of different individual pathogens and pathogen complexes will be defined, providing an avenue for SARDI to realistically interpret molecular DNA tests in a practical and relevant way for producers. This includes provision of *Aphanomyces trifolii* samples for development of a new molecular test for this pathogen, and also provision of additional samples of the range of major pathogens and their complexes for SARDI to validate the success of the new molecular DNA pathogen probe for *Aphanomyces trifolii* and/or the application and interpretation of molecular DNA test results such that producers receive accurate and meaningful interpretations of their on-farm situations in terms of both pathogens present and expected soil-borne disease expression. These molecular DNA tests will be utilised to quantify on-farm impacts of chemical and cultural management technologies arising from this project *(Objectives 1,2,3 and 5)*

iii. Focus to broaden quantification of pasture productivity constraints induced by pasture soil-borne root pathogens across the agro-ecological zones of southern Australia. While there is much historical knowledge, recent studies [e.g., Simpson et al (2011) and O’Rourke et al. (2009)] suggest that the historical situation of the 1980’s and 1990’s likely no longer represents the current situation in terms of soil-borne disease impacts occurring (e.g., due to changes in pathogen races as for *Phytophthora,* recent identification and confirmation of importance of *Aphanomyces trifolii*). This project will determine current incidence and impacts (losses and economic importance) of major soil-borne diseases in subclover. *(Objective 4)*

iv. All plant, pathogen and soil samples and relevant information needed by SARDI will be sent to SARDI to develop a new molecular DNA test for *Aphanomyces trifolii*. This is essential if molecular DNA tests are to have the required capacity to deliver in terms of both research and monitoring in relation to soil-borne pasture diseases. Estimated cost of $50,000 for the development of this new molecular DNA probe has been budgeted in this project in year 1, as have been the costs for delivery and molecular analyses of all other field samples sent to SARDI at the appropriate stages for other stages and components of this proposal as outline under other objectives (*Objective 5*).

In addition, this project will also train/transfer, within the broader context of pasture agronomy, breeding and science, broad spectrum soil-borne pathology skills, such that the long-term capacity of pasture pathology is maintained nationally to ensure future productivity of the Australian livestock feed-base and an ongoing capacity to deal with future soil-borne disease and any other disease challenges. This will be achieved by locating and training and developing high level plant pathology skills in a postdoc pathologist who is highly motivated and committed long-term to ensuring the welfare of the Australian livestock feed-base. This has been added as an additional objective to the specific objectives listed below for this proposal.

3. Linkages to this project – This project will:

i. Link with to provide pathology support to and add to the outcomes of project B.PUE.0103 "*Root Disease Constraints in Pasture Productivity*" which is focussed on the interaction between pasture phosphorus nutrition and root disease. This project will provide the required pathology backup so desperately needed for that project.

ii. Link with to provide pathology support to and add to the outcomes of relevant projects that emerge from the MLA "*Plant Health*" theme of the *Southern Feedbase Investment Plan*.

iii. Link with to provide pathology support to and add to the outcomes of project theme in the *Feedbase Investment Plan* on "*Pasture legume performance and proportion*" as that project has direct relevance to legume productivity and seed bank persistence.

iv. Develop and consolidate new and existing knowledge and skills on pasture soil-borne disease pathology by mentoring the appointed postdoc pathologist to this project. In addition to Barbetti, this postdoc pathologist will be supported by Megan Ryan at UWA in relation to development of novel pasture species and associated mycorrhizal and rhizobial associations; and by Phil Nichols at DAFWA and CLIMA in relation to the breeding, selection and agronomy of pasture species with improved disease resistance. Barbetti, Ryan and Nichols have comprehensive local, national and international networks with other plant pathologists and pasture researchers and all three are involved in undergraduate teaching of pasture systems and the training of postgraduate PhD candidates on pasture issues - the postdoc pathologist will obtain additional skills, experience and opportunities from this. Nichols has been a pasture legume plant breeder national plant breeding programs, including the National Annual Pasture Legume Improvement Program (NAPLIP) and the FFICRC salinity tolerance project, and has extensive national and international pasture breeding networks and strong links to the Australian pasture seed industry. Ryan has a leadership role in the FFICRC and links to the FFICRC provide additional access to other important collaborative pasture breeding and selection programs at SARDI (including the lucerne program), DPI Vic and DPI NSW. Together, these approaches ensure that the postdoc pathologist will not only deliver on the proposed project but will be trained to a high level to meet both current and future pathology requirements for securing the feedbase productivity for the livestock industry.

Objectives and outcomes of project

1. Overall purpose and objective

This project is built upon a solid national focus, one that will provide the critical knowledge that meat producers, extension consultants, agronomists and researchers need to manage soil-borne diseases in subclover pastures and in order for them to be able to increase their sustainability, productivity and profitability. It will also eliminate the current production uncertainly, by providing producers with a range of management options that ensure certainty of success in managing soil-borne disease threats. This will not only ensure sustainable pastures that are reliable and persist, but that they have high productivity across the critical autumn-winter period. This will also ensure ongoing productivity from having pastures that not persist throughout an individual season but across multiple years. In particular, it will provide the practical pasture management techniques, giving meat producers flexible options to curtail current losses in pasture productivity caused by soil-borne root pathogens and thereby increase farm profitability with existing pastures. Finally, this project will ensure that the necessary pathology skills and expertise for dealing with soil-borne disease threats to the feedbase are maintained for future generations of producers.

2. Specific objectives of this project

7. Determine the farm management factors that influence the expression of soil-borne root diseases in subclover pastures.
8. Evaluate practical management techniques aimed at reducing the pasture productivity loss during autumn-winter induced by soil-borne root disease.
9. Characterise the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots.
10. Broaden the quantification of pasture productivity constraints induced by pasture soil-borne root pathogens across agro-ecological zones.
11. Develop molecular assays to fill gaps in experimental tools needed to adequately conduct research into, and monitor, soil borne diseases in pastures.
12. Secure pathology skills and expertise for pastures into the future.

3. Direct outcomes from this project

Direct outcomes in relation to objectives 1 and 2

Objectives 1 and 2 will develop and deliver practical on-farm measures that producers can readily adopt; measures that will not only provide producers with real and practical options to curtail current losses from soil-borne diseases, but with options that provide producers flexibility in choice of control options to utilise taking into account livestock prices, potential productivity of pasture(s), degree of feed shortage, etc. Examples include the following:

Chemicals: Outcome will be a range of effective and minimal cost chemical management options for meat producers, not only to provide assurance in establishing or re-establishing pastures, but also foliar applications to protect naturally regenerating pastures. Together, these chemical options will allow producers to minimise seedling root disease, particularly across high rainfall regions, and provide meat producers with greater flexibility in terms of disease management approaches available to them. In particular, it will provide producers with in-season opportunities to minimise disease and maintain both persistence and productivity in existing pastures throughout the season Widespread utilisation of chemical control options across high rainfall pastures could be expected to improve the productivity of the pasture feed-base component for meat producers, particularly when based on sown and re-sown pastures, of the order of 25%.

Manipulation of farm practices: The outcomes will be a range of effective and minimal cost cultural management options for meat producers to minimise the impact of soil-borne diseases on pasture legume productivity across all regions, particularly high rainfall >500mm areas. Further, as with chemicals, this will provide meat producers with greater flexibility and choice in terms of management options and approaches available to them pre-season and during the season. Widespread utilisation of cultural management options could be expected to improve the productivity of the pasture feed-base for meat producers across high rainfall regions of southern Australia in the order of 10%.

Direct outcomes in relation to objective 3

Outcome from addressing objective 3 will definition of relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots and how these relationships are strongly determined by environmental influences. This is not only essential in order to be able to define both the different symptom expressions on roots and the relative impacts of different individual pathogens and pathogen complexes in different regions and seasons, but provides the only sound basis for interpreting molecular DNA tests in a practical and meaningful way for producers. To this end, these studies will also provide the samples and materials needed for both development of new molecular tests by SARDI for the new important pathogen *Aphanomyces trifolii*, as well as provide SARDI with, for the first time, samples of individual and complexes of pathogens differentially expressing disease and upon which to develop, refine and then validate the molecular DNA testing. This is the only basis by which producers can be provided with accurate and meaningful interpretations of their on-farm situations, both in terms of pathogens present and also in relation to seasonal variations in terms of disease levels and the consequent disease impacts.

Direct outcomes in relation to objective 4

The outcome from addressing objective 4 will be quantification of importance and economic impact on pasture productivity of constraints induced by pasture soil borne root pathogens across the agro-ecological zones of southern Australia. It is evident that historical situations of the 1980's and 1990's no longer represent the current situation in terms of losses (e.g., due to changes in pathogen races as for *Phytophthora* and impacts of new pathogens such as *Aphanomyces trifolii*) and that quantification of losses is required.

Direct outcomes in relation to objective 5

The outcome from addressing objective 5 is development and availability of a new molecular DNA assay for *Aphanomyces trifolii*, currently a major gap in the experimental tools and precluding their effective use for conducting research into, and monitoring, soil-borne diseases in pastures. This additional capability is critical if molecular DNA tests are to have the capacity to meet the requirements in terms of both research and monitoring in relation to soil-borne pasture diseases and be an effective tool for quantifying roles of pathogens and benefits from application of on-farm management practices for management of soil-borne diseases.

Direct outcomes in relation to objective 6

The outcome from objective 6 will be the development and consolidation new and existing knowledge on pasture soil pathology by equipping the appointed postdoc pathologist on this project with the full spectrum of pathology skills needed to deal with soil-borne disease issues for current and future generations of meat producers. Such skills include abilities to conduct disease surveys, to isolate, identify and manipulate soil-borne pathogens, to determine disease impacts, define complex inter-pathogen interactions and consequences, and define both expression of diseases and their interactions with the host and wider environment, including nutritional status. The outcome is that, in a wider sense, such a person can provide the pathology support required by any and all pasture research programs, as required to develop IDM packages for the livestock industries, and as required to deal with current and future disease threats to the pasture feed-base.

4. Summary of overall direct outcomes from this project

Conservatively, the overall direct outcomes from this project are as follows:

i. Provision of the knowledge and tools for meat producers to substantially increase subclover productivity in permanent pastures by at least 25% through better understanding and practical management of soil-borne root diseases.
ii. Definition of extent that practical on-farm management practices can mitigate the expression of soil-borne root diseases of subclover and consequent plant performance.
iii. Delivery and demonstration of the most effective practical management options identified to producers in field sites across southern Australia for their capacity, reliability and impact in improving subclover productivity in the face of soil-borne disease challenges.
iv. Definition of relationships between the expression of disease on plants with the associations of particular soil-borne pathogens and how environmental influences determine the outcomes of these interactions and relationships - essential both to define both the different symptom expressions on roots and the relative impacts of different individual pathogens and pathogen complexes in different regions and across different seasons, and as the only basis for interpreting molecular DNA tests for producers
v. Quantification of the pasture productivity constraints induced by soil-borne disease of subclover across agro-ecological zones, also providing a focus for where and what type of future funding decisions are justified.
vi. Training of a new postdoc scientist with skills in soil-borne plant pathogens will ensure availability of these skills for producers into the future and is critical both to delivery of the outcomes listed above and also to advancing future research on pasture soil biology to continually improve management of soil-borne disease threats.
vii. Provision of the knowledge, pathogen isolates, diseased plant and soil samples, needed for SARDI to develop, refine and validate a package of molecular DNA assays of soil-borne pathogens that that can then be used as research and monitoring tools for better management of soil-borne diseases in pastures.

5. Additional outcomes for related pasture projects and programs

The project will also provide additional outcome benefits to related projects as follows:

i. Project B.PUE.0103 "*Root Disease Constraints in Pasture Productivity*" which is focussed on the interaction between pasture phosphorus nutrition and root disease will be provided with relevant pathology skills and expertise both through UWA and by periods of basing the postdoc pathologist in CSIRO labs in Canberra.

ii. Relevant projects that emerge from the MLA "*Plant Health*" theme of the *Southern Feedbase Investment Plan* will be assisted by provision of relevant findings, guidance and assistance from this project and, if required, further assisted by visits to these projects by the postdoc pathologist based at UWA.

iii. *Feedbase Investment Plan* on "*Pasture legume performance and proportion*" has direct relevance to legume productivity and seed bank persistence and will be provided with appropriate pathology guidance and assistance from this UWA project.

Methods

Objectives 1 and 2:

#1: Determine the farm management factors that influence the expression of soil-borne root diseases in subclover pastures

#2: Evaluate practical management techniques aimed at reducing the pasture productivity loss during autumn-winter induced by soil-borne root disease

Chemicals:
i), *Years 1 and 2:* Initially, we will test a wide range (minimum 15-20) of the best potential chemical seed treatment and/or seedling spray options (e.g., thiram, rovral, metalaxyl, propamocarb, various azole fungicides, phosphorous acid, and resistance-inducing compounds such as beta hydroxy acid, etc) for control of pre- and post-emergence damping-off and for control of seedling root disease during the autumn-winter period. Best prospects for testing will be identified from existing literature (Australia and overseas) and the local researcher knowledge base. There is certainty for success with locating and defining successful chemical treatments for this use because researchers in WA already have strong and successful track records in this area. To reduce costs and to rapidly identify the best chemical treatment prospects, these initial screenings of prospective chemical treatments will be undertaken in years 1 and 2 of the project in small field plots at Shenton Park Field Station in Western Australia, on land that has a history of several decades of subclover pastures. Reductions in root disease from chemical treatments will be assessed on the plants in each chemical treatment as will corresponding increases in plant growth.

ii), From these initial field testings in years 1 and 2, we will identify the most cost-effective chemical seed and/or foliar treatments for effective control of pre- and post-emergence damping-off and seedling root disease in subclover pastures.

iii), *Years 3 and 4*: Subsequently, in years 3 and 4, we will test the best three chemical prospects for seed treatments for control of seedling damping-off and the best of the foliar seedling spray treatments for control of root disease. We will do this by demonstrating their effectiveness nationally in eight on-farm trials (two each per WA, SA, Vic, NSW; each consisting of 3 chemical and a nil treatment comparison and with 4 treatment replications giving a total of 16 plots at each demonstration site) providing meat producers with on-farm demonstration of the effectiveness of these chemical treatments for ensuring both successful stand establishment and persistence (damping-off and seedling root disease) and ongoing productivity and persistence of surviving plants (root disease) in subclover pastures. Reductions in root disease from each chemical treatment will be assessed on plants across each chemical treatment as will corresponding increases in productivity. A composite sample from each of the 16 plots will be taken at 3 and repeated again at 9 weeks after commencement of the growing season and sent to SARDI to molecularly confirm and quantify the differences in pathogen population and density and define the pasture productivity increases obtained from these on-farm chemical treatments (total 32 samples for DNA testing each in years 3 and 4 at each field location = total DNA test samples of 512 over the two years and 8 sites). These field demonstration trials will be conducted in conjunction with Agronomists/consultants and in association with the pasture agronomy and extension personnel and programs across Australia. *Note: This is the reason for the particularly high operational costs listed for the third and fourth years of this project.*

Manipulation of on-farm cultural management practices:
i), *Years 1 and 2:* We will firstly, identify the best prospects for utilisation as cultural management measures (e.g., variety of subclover, grazing, cultivation, pH adjustments, application of rhizobial treatments, altered pasture composition, etc), for control of pre- and post-emergence damping-off and for control of seedling root disease during the autumn-winter period. Best prospects for testing will be identified from existing literature (Australia and overseas) and the local researcher knowledge base. There is certainty for success with locating and defining successful cultural management treatments for this use because researchers in WA already have strong and successful track records in this area. To reduce costs and to rapidly identify the best cultural management treatment prospects, these initial screenings of an estimated 10-15 different prospective cultural treatments will be undertaken in years 1 and 2 of the project in small field plots at Shenton Park Field Station in Western Australia, on land that has a history of several decades of subclover pastures. Reductions in root disease from these cultural treatments will be assessed on the plants in each cultural treatment as will corresponding increases in plant growth.

ii), From these initial field testings in years 1 and 2, we will identify the most promising and cost-effective cultural management treatments for effective control of pre- and post-emergence damping-off and seedling root disease in subclover pastures.

iii), *Years 3 and 4*: Subsequently, in years 3 and 4, we will test the best two cultural management treatment prospects for control of seedling damping-off and root disease. We will do this by demonstrating their effectiveness nationally in eight on-farm trials (two each per WA, SA, Vic, NSW; each consisting of 3 cultural management practices and a nil treatment comparison and with 4 treatment replications giving a total of 16 plots at each demonstration site) providing meat producers with on-farm demonstration of the effectiveness of these cultural management treatments for ensuring both successful stand establishment and persistence (damping-off and seedling root disease) and ongoing productivity and persistence of surviving plants (root disease) in subclover pastures. Reductions in root disease from each cultural treatment will be assessed on plants across each cultural treatment as will corresponding increases in productivity. A composite sample from each of the 16 plots will be taken at 3 and repeated again at 9 weeks after commencement of the growing season and sent to SARDI to molecularly confirm and quantify the differences in pathogen population and density and define the pasture productivity increases obtained from these on-farm chemical treatments (total 32 samples for DNA testing each in years 3 and 4 at each field location = total DNA test samples of 512 over the two years and 8 sites). These field demonstration trials will be conducted in conjunction with Agronomists/consultants and in association with the pasture agronomy and extension personnel and programs across Australia. *This is the reason for the particularly high operational costs listed for the third and fourth years of this project.*

Objective 3: Characterise the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots

Root rot of subclover clearly involves a complex of fungi which interact not only among themselves but also with the biotic and abiotic environment surrounding them – a complex combination of interactions that determines the final expression of root disease in pastures. A range of different fungi are normally associated with diseased subclover roots and no one pathogen has been able to reproduce the wide range of different field disease symptoms observed in different locations across southern Australia. At the current state of investigations, the most important pathogens associated with subclover root rot are, in order of importance; i), the many races of *Phytophthora clandestina*, ii), *Pythium irregulare*, and iii), *Aphanomyces trifolii*. While it is known that a number of different pathogens are present on diseased roots of pasture legumes, and that they do interact, the way in which such soil-borne pathogen interactions determine the final expression of root disease in pastures is unknown. This poses a problem precluding molecular DNA tests from being able to provide the meaningful feedback required by producers in relation to soil-borne disease occurring on their farms. Further, the expression of disease and the associations of soil borne pathogens with affected plant roots is strongly influenced by environmental factors such temperature, moisture and nutrition, and the level of host resistance in the affected subclover variety. Together these interactions and associations will determine the final expression of root disease in subclover pastures and provide both the basis and understanding required for meaningful application and interpretation of molecular DNA tests on pastures for producers.

Years 1-5: We will undertake wide-ranging investigations in this area to not only characterise the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots; but also to explain the differences in symptoms, disease severity, expression, etc. that occur between different regions as a consequence of variations in pathogen populations, environmental conditions and varieties with differing levels of host resistance. To do this we will undertake glasshouse studies (as this cannot be conducted in the field) to:

i. Characterise the relationships between the expression of disease on affected plant roots with the associations of individual and combinations of the soil-borne pathogens *Phytophthora clandestina*, *Pythium irregulare*, and *Aphanomyces trifolii*. This will involve testing 3 different levels of inoculum of each pathogen across all the possible pathogen x inoculum concentration combinations in factorial designed experiments each with a minimum 6 replications (i.e., minimum 108 pathogen combination treatments x 6 replications = 648 pots in total). Expression of root disease in each of these pathogen combinations will be assessed for all plants in each treatment as will corresponding effects on plant foliage and root productivity.
ii. Subsequent to completion of (i) above, then the influence of environmental factors such as temperature, moisture and soil type, and nutritional factors, upon the expression of disease on affected plant roots with the associations of individual and combinations of these soil-borne pathogens will be determined. This will involve testing the possible 2- and 3-way combinations of the 3 pathogens across 3 different temperature regimes and 3 different moisture regimes representative of autumn-winter temperatures across southern Australian pastures and across 3 different nutritional regimes and 2 different soil types in factorial designed experiments each with a minimum 6 replications (i.e., minimum 108 pathogen x environment combinations x 6 replications = 648 pots in total). Expression of root disease in each pathogen x environment combination will be assessed for all plants in each treatment as will correspondingly the effects on plant foliage and root productivity.
iii. *Year 3:* Once these relationships have been defined, representative samples from the experiments described in (i) and (ii) above that define the full range of characteristic expressions of disease on affected plant roots from the associations of individual and combinations of the soil-borne pathogens interacting with environmental factors will be sent to SARDI to enable calibration and validation of molecular DNA tests firstly to the disease expressions in these studies so they can subsequently be molecularly confirmed to enable better quantification of differences in pathogen populations in relation to field expression of disease for the different pathogens and their combinations across the geographical regions of southern Australia. It is estimated that approximately 60 molecular DNA tests will be needed for this purpose.
iv. *Years 4 and 5:* Subsequently, in years 4 and 5, studies will be undertaken to define relationships in the field between field expressions of root disease with the main environmental factors as occurs across the agro-ecological zones for southern Australia. This will also involve collection and collation of relevant environmental data across southern

Australia. Then available data will be incorporated into an integrated modelling package to simulate potential risk(s) associated with deployment of different host cultivars, for different seasonal conditions and for different geographic scenarios occurring across the southern Australian pasture belt. This modelling will be utilised to not only define the field influence of environment on disease expression across the agro-ecological zones of southern Australia, but, more importantly enable ability to define the relative risks for producers in different regions and across different seasons.

Objective 4: Broaden the quantification of pasture productivity constraints induced by pasture soil-borne root pathogens across agro-ecological zones

Year 3: We will quantify the pasture productivity constraints induced by pasture soil-borne root pathogens across agro-ecological zones from major soil-borne pathogens on subclover. We will do this by establishing 2 field trials each in NSW, Victoria, SA and WA (total 8 field trials across southern Australia) in year 3 to quantify the losses occurring at pre- and post-emergence and on seedling and mature plants throughout the growing season over two years as follows:

i. Establish field trial treatments utilizing the Wong et al. (1984) – Simpson et al. (2011) bioassay technique for pre- and post- emergence disease levels and associated losses in subclover (but incorporating additional treatments/replications such that sampling and assessment for root disease and rhizobial colonisation can continue throughout the season in a way such that while quantification is continuously undertaken it can be quantified for any particular time or period of the growing season.
ii. At each field site, all bioassay treatments will be established in both natural and dazomet fumigated soils at each field site, such that maximum potential productivity capacity in the absence of pathogens can be ascertained.
iii. At each field site, bioassay subclover varieties will consist of one known susceptible variety and one with an effective level of field resistance to root disease, in particular to *Pythium* and *Phytophthora*.
iv. At each field site, assess and sample bioassay field trials at emergence at 3 weeks after sowing for pre- and post-emergence severity and losses; for seedling root disease and losses at 9 weeks; and then for mature plant disease and losses at 20 weeks after sowing (2 varieties x ±fumigation x 4 replications = 16 field plots).
v. At each field site, isolations and identification of pathogens from diseased roots of plants will be made to define the exact cause(s) of the disease expressed and the associated quantified losses occurring at critical times of the season at each site.
vi. From each field site, samples will be sent to SARDI where DNA methods will be utilised to molecularly quantify pathogen(s) and their combinations occurring in subclover roots and to calibrate predicted levels from DNA tests to actual levels of root disease and root damage, and also pasture production, as expressed across treatments at each field location. To obtain this data, on each occasion when plants roots are assessed for seedling losses and/or root disease damage, soil and plant samples will be sent to SARDI for measurement of DNA of known subclover pathogens. A composite sample from each of the 16 plots will be taken at 3 and repeated at 9 weeks after commencement of the growing season and sent to SARDI to molecularly confirm and quantify the differences in pathogen population and density and define the pasture productivity increases obtained from these on-farm chemical treatments (total 32 samples for DNA testing in year 3 at each field location = total DNA test samples of 256 across the 8 sites in year 3).
vii. *Years 4 and 5:* Finally, utilising data from these field trials, we will use and apply appropriate modelling, as used previously, to provide both the dollar losses incurred from varying levels of soil-borne disease and associated yield and productivity losses.

All direct disease assessments and isolation of pathogens will be addressed and delivered through UWA. However, this field trial network will be conducted in association with pasture extension and agronomy programs across southern Australia for the establishment, maintenance and sampling of the trials outside of WA and with relevant government agencies, such as CSIRO in Canberra and SARDI in SA. *Note: This is deliberately planned for year 3 to be in conjunction with field trials outlined under objectives 1 and 2 to maximise effective management and running of both sets of field trials at the same time and locations. Despite this, this field component remains the reason for the particularly high operational costs listed for the third year of this project.*

Objective 5: Develop molecular assays to fill gaps in experimental tools needed to adequately conduct research into, and monitor, soil borne diseases in pastures

Years 1 and 2: This objective will ensure development of the required suite of molecular DNA assays needed to fill the existing critical gap in experimental tools needed to adequately conduct research into, and monitor, soil borne diseases in pastures. This will occur from the development of a new molecular DNA prove needed to test for *Aphanomyces trifolii*. This is absolutely vital to undertake if the molecular DNA tests are to have the capacity to meet the requirements in terms of both research and monitoring in relation to soil-borne pasture diseases and in quantifying the benefits of on-farm management practices. This will be achieved by collection and forwarding of relevant project pathogen and infested plant and soil samples for DNA extraction at SARDI who will undertake the required probe development.

Years 1, 2, 3 and 4: Development of the molecular probe for *Aphanomyces trifolii* will enable soil pathogen population analyses of changes in the structure and function in relation to the benefits from on-farm management and chemical applications from this project that demonstrate reduced soil-borne disease impacts for field components listed under Objectives 1, 2 and 4 and for glasshouse components listed under Objective 3. *Note: SARDI will invoice this project on a cost-recovery basis for $50,000 for development of this molecular probe for Aphanomyces trifolii and this has been budgeted in this project in year 1 against this objective. Additional costs of $62,100 in year 3 and $38,400 in year 4 have also been budgeted for molecular analyses of extensive field samples sent to SARDI and as detailed above under Objectives 1, 2 and 4 and for glasshouse sample outlined under Objective 3.*

Objective 6: Secure pathology skills and expertise for pastures into the future

Training and skilling a new pasture pathologist is an integral part of this project:

- *Firstly*, in terms of delivery of outcomes directly sought from this project.
- *Secondly*, in terms of developing these skills as a consequence of direct involvement in this project.
- *Thirdly*, in terms of developing these skills as a consequence working with related pasture projects.
- *Finally*, from the unique opportunity to capture the existing pasture pathology knowledge of Barbetti and others in WA.

The postdoc pathologist will collaborate and work closely with the new MLA-AWI project B.PUE.0103 that will examine interactions between root disease and plant P efficiency for subclover using the newly developed subclover core collection, providing both learning for the postdoc pathologist and provision to the MLA-AWI project B.PUE.0103 project of the needed pathology support from both the postdoc pathologist and Barbetti. In addition, the postdoc pathologist will have exposure by direct visits to other relevant projects that emerge from the MLA "Plant Health" theme of the Southern Feedbase Investment Plan; and the Feedbase Investment Plan on "Pasture legume performance and proportion", a project that will have direct relevance to legume productivity and seed bank persistence; and finally, from visits to related networks associated with these projects.

In addition to Barbetti, this postdoc pathologist will be supported by Megan Ryan at UWA in relation to development of novel pasture species and associated mycorrhizal and rhizobial associations and by Phil Nichols the pasture group at DAFWA in relation to the breeding, selection and agronomy of pasture species with improved disease resistance. Ryan's leadership role in the FFI CRC provides access to other important collaborative pasture breeding and selection programs at SARDI (including the lucerne program), DPI Vic and DPI NSW. Nichols, a pasture legume plant breeder on several national plant breeding programs, including the National Annual Pasture Legume Improvement Program (NAPLIP) and the FFI CRC salinity tolerance project, and has extensive national and international pasture breeding networks and strong links to the Australian pasture seed industry. Further, Barbetti, Ryan and Nichols all have comprehensive local, national and international networks with other plant pathologists, pasture agronomists, breeders, extension networks and other pasture researchers from which will add to the overall pasture pathology experience gained by this postdoc pathologist. Finally, Barbetti, Ryan and Nichols are also involved in undergraduate teaching of pasture systems and the training of postgraduate PhD candidates on pasture issues at UWA - the postdoc pathologist will obtain additional skills and experience and have further opportunities from this. Finally, the postdoc pathologist will have the opportunity to acquire skills in relation to development and interpretation of molecular DNA testing for root diseases.

Together, these approaches outlined towards involving and training a new pasture pathologist will ensure close alignment of the proposed area of research by the postdoc pathologist to meet both current and future pathology requirements of the livestock industry.

Interest (IP proportions)

MLA	64%
Research Organisation	36%

Milestones

Milestone Number	Achievement criteria	Due date
1	Postdoc pathologist appointed at UWA *(All Objectives, especially #6)*	3 months after project signing
2	Best chemical control measures identified from year 1 local field trials *(Objectives 1 and 2)*	End year 1
3	Best cultural control measures identified from year 1 local field trials *(Objectives 1 and 2)*	End year 1
4	Best chemical control measures identified from year 2 local field trials *(Objectives 1 and 2)*	End year 2
5	Best cultural control measures identified from year 2 local field trials *(Objectives 1 and 2)*	End year 2
6	New molecular probe developed for Aphanomyces (Objective 5)	End year 2
7	Impacts of chemical control measures successfully demonstrated in national field trials in year 3 *(Objectives 1 and 2)*	End year 3
8	Impacts of chemical control measures successfully demonstrated in national field trials in year 3 *(Objectives 1 and 2)*	End year 3
9	Relationships between the expression of disease and the associations of soil-borne pathogens characterised (Objective 3)	End year 3
10	Impacts of chemical control measures confirmed in national field trials in year 4 *(Objectives 1 and 2)*	End year 4
11	Impacts of chemical control measures confirmed in national field trials in year 4 *(Objectives 1 and 2)*	End year 4
12	Yield impacts of soil-borne disease quantified across national field trials (Objective 4)	End year 4
13	Relationships between disease expression and molecular DNA tests characterised *(Objective 3)*	End year 5
14	Final report to MLA	End year 5

Nominated Person(s)

Title/First Name/Surname	Dr Martin Barbetti
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Project Budget and Funding

Year 1

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 1
Postdoctoral Research Associate; Level A step 08; 28% on costs (100%, TBA). Barbetti (20%)	114,764 0	0 0	0 59,024	8,283 57,218	120,047 12,959 116,242
Travel	7,500				7,500
Operating	23,350				23,350
SARDI development of new DNA assay for *Aphanomyces*	50,000				50,000
Equipment	0				0
TOTAL BUDGET (excl. GST)	195,614	0	59,024	65,501	320,139

Year 2

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 1
Postdoctoral Research Associate; Level A step 08; 28% on costs (100%, TBA). Casual labour (40d) Barbetti (20%)	120,502 13,524 0	0 0 0	0 0 61,975	8,697 0 60,079	129,199 13,524 122,054
Travel	8,300				10,300
Operating	25,100				25,100
Equipment	0				0
TOTAL BUDGET (excl. GST)	167,426	0	61,975	68,776	328,177

Year 3

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 1
Postdoctoral Research Associate; Level A step 08; 28% on costs (100%, TBA). Casual labour (60d) Barbetti (20%)	127,732 22,294* 0	0 0 0	0 0 65,074	9,219 0 63,083	136,915 22,294 123,157
Travel	18,745*				18,745
Operating	132,600*				132,600
SARDI DNA tests (512, 60, 384; objectives 1 & 2, 3,4, respectively)	62,100				62,100
Equipment	0				0
TOTAL BUDGET (excl. GST)	363,471	0	65,074	72,302	500,847

**The reason for the particularly high operational, travel and casual labour costs listed for the 3rd year of this project are to cover costs of on-farm field trials across 4 states; firstly, in association with Objectives 1 and 2 to deliver cost-effective chemical and cultural management techniques for producers across southern Australia; and, secondly in association with Objective 4 to quantify field pasture productivity constraints.*

Year 4

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 1
Postdoctoral Research Associate; Level A step 08; 28% on costs (100%, TBA). Casual labour (80d) Barbetti (20%)	135,399 23,249* 0	0 0 0	0 0 68,328	9,772 0 66,237	145,171 23,249 134,565
Travel	21,745*				21,745
Operating	111,400*				111,400
SARDI DNA tests (512 objectives 1 & 2)	38,400				38,400
Equipment	0				0
TOTAL BUDGET (excl. GST)	330,193	0	68,328	76,009	474,530

**The reason for the particularly high operational, travel and casual labour costs listed for the 4th year of this project are to cover costs of on-farm field trials across 4 states in association with Objectives 1 and 2 to deliver cost-effective chemical and cultural management techniques for producers across southern Australia;*

Year 5

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 1
Postdoctoral Research Associate; Level A step 08; 28% on costs (100%, TBA) Barbetti (20%)	143,523	0 0	0 72,427	10,358 70,211	153,881 142,638
Travel	8,745				8,745
Operating	21,800				21,800
Equipment	0				0
TOTAL BUDGET (excl. GST)	174,068	0	72,427	80,569	327,064

Budget Summary

	Year 1	Year 2	Year 3	Year 4	Year 5	TOTAL
MLA	195,614	167,426	363,471	330,193	174,068	1,230,772
Research Organisation contributions	124,525	130,751	137,376	144,337	152,996	689,985

Contributors/Other Funds

Funding Breakdown (only if applicable)

Party	Amount of Contribution	Type of Contribution	Amount of Interest in Intellectual Property
MLA	1,230,772	Cash	64%
Other Contributors - UWA	689,985	Salary Barbetti + Overheads	36%
Research & Other Organisations (UWA in kind)	See note below *	Infrastructure support	

**UWA guarantees to provide all the necessary infrastructure support for the postdoc pathologist, including full access to laboratories, controlled-environment rooms, glasshouses and field experimentation sites, and meet all other indirect costs (phone, printing, technical and professional support, etc).*

Please also note:

i. ***That additional funding will also be sought from other sources (e.g. ARC) to further expand and build long-term capacity in pasture sciences, particularly pasture pathology;***
ii. ***Prof Hans Lambers, Head of the School of Plant Biology at UWA, has indicated strong support for the postdoctoral fellow towards a tenure track at UWA after the initial 5 year period funded by MLA, subject to satisfactory performance. This aligns well with Plant Biology plans to further expand its research base towards underpinning the long-term feed-base requirements for the animal industries of Australia. Other plans include enhancing research capacity in pasture breeding, agronomy and nutrition.***

Agent(s)/Subcontractor(s)

Name of Company **ABN** **Contact Person Title/First Name/Surname** **Mailing Address** **Phone Number** **Facsimile Number** **Email Address**	*Note: These details will be finalised for SA, Vic, and NSW once the exact extent and nature of the sampling and field on-farm experiments involved in this project across southern Australia has been negotiated with MLA. This project will involve at least one key contact person at each of SARDI in SA, DPI Vic in Victoria, and DPI NSW; and also involve private extension consultants to meat producers and producer groups across southern Australia.*

Communications and Delivery

Target groups

Group	Total Potential Number of People
Meat producers across southern Australia with subclover pastures	All
Private and state government extension and research personnel involved with subclover pasture productivity across southern Australia – including meat producer and grazing grower groups	All

Awareness/Participation/Adoption Objectives

Year following launch:	Awareness	Participation	Adoption
Year 1	10%	5%	0%
Year 2	20%	10%	5%
Year 3	60%	20%	10%
Year 4	80%	40%	20%
Year 5	95%	60%	30%

Communication Products/Tools/Services

Cost estimate included in budget? ☐ Yes ☐ NO No

Communication/Delivery Channels

Cost estimate included in budget? ☐ Yes ☐ NO No

Other Capacity Building

Training of new pasture pathologist to support and secure the feedbase from soil-borne diseases

Cost estimate included in budget? | YES | Yes | | No

Timelines

As outlined and detailed in project application for across the 5 years for research addressing each objective

APPENDIX to Project Specification Scenario 1

Comprehensive background to proposal

1. Critical feed shortages coincide with period of most severe soil-borne disease

The recent Soil Biology Workshop held in February 2012 highlighted both the need and opportunities to curtail current severe losses from soil-borne diseases (and as outlined in detail below) such that legume based pasture productivity across southern Australia is greatly increased. It was agreed at this workshop that this would best be achieved by focusing on the managing soil-borne diseases in the main legume component, subclover and hence the focus on subclover in this application.

Crucially, meat producers inherently face critical feed shortage in across the autumn-winter months. That the main soil-borne pathogens attack and do most damage during this period causing massive pre- and post-emergence damping-off and seedling root disease at this time of critical feed shortage compounds the adverse impact of these losses on pasture availability and productivity. This critical shortage of pasture feed during the autumn-winter period in southern Australian pasture systems is likely the major constraint to increasing livestock production and profitability. Any increase in extra carrying capacity developed during this autumn-winter period from improved disease management will not only improve overall carrying capacity but also reduce the current high degree of uncertainty faced by meat producers in being assured of sufficient feed over this period. Further, such improvement will also allow better overall pasture utilisation in spring, translating directly into substantially increased profits for meat producers. In addition, not only does soil-borne disease particularly target and restrict seedling emergence and growth, resulting poor pasture performance, it also results in severe decline in seed banks, further increasing the risks and opportunity costs of pasture renovation or any other investment into pasture improvement. Perhaps of greatest significance is the well documented rapid deterioration and lack of persistence of subclover pastures across southern Australia following attack by soil-borne pathogens (Barbetti et al. 2005, 2007). Finally, it has also recently been demonstrated by O'Rourke et al. (2009) for the first time that significant losses in subclover continue throughout the remainder of the season following attack in the autumn-winter period, restricting spring production as well.

Losses in pasture legumes from soil-borne diseases have been well documented for decades (see Barbetti et al. 2005, Barbetti and Jones 2011, Barbetti et al. reviews of 2005, 2007 and MLA report of 2005 by Barbetti et al.). For example, pre- and post-emergence damping-off results in pastures failing to establish or persist, or be productive, with annual average losses from major soil-borne pathogens up to 40-45% in the most disease prone areas. Conservatively, the combined annual losses from major soil-borne diseases of legume based pastures across southern Australia are likely to exceed $400 m annually, the majority of this being in relation to subclover pastures (Barbetti, unpubl.

2. Impact of soil-borne diseases across southern Australia

Recently, Simpson et al. (2011) used field-based plant bioassays to assess the potential for pre- and post-emergence loss of seedlings and for root damage affecting subclover during autumn-winter at 17 locations across a broad agricultural area of temperate southern Australia. Between 9 and 93% (median 21%) of subclover seedlings failed to emerge at 14 locations, and with post-emergence losses in the range 0-32% (median 7%) but with moderate to severe root disease on surviving test plants at all of the sites. These studies not only highlighted the extent of lethal damage, but that sub-lethal damage to pasture roots also constitutes a potentially large, but underestimated cost to production because it was so widespread and because the damage occurs during autumn-winter when pasture yield limits stocking rate. Prior to this research, there were many studies to demonstrate the impact of soil-borne diseases across southern Australia where root rot has seriously adversely affected the production from subterranean clover pastures over at least the past 4 decades (e.g., Johnstone and Barbetti 1986; Barbetti et al. 2005, Barbetti and Jones 2011, Barbetti et al. reviews of 2005, 2007 and MLA report of 2005 by Barbetti et al.) and a brief summary of these impacts is a follows:

Victoria: In Victoria, soil-borne disease on subclover was first recognized in 1960 (Anon. 1960) and is a serious problem which reduces the productivity of pastures (Taylor 1984, 1985; Taylor et al. 1984). A survey of subclover pastures in 1970 (McGee and Kellock 1974) showed that root rots were widespread in northern, southern and eastern Victoria, with more than 70% of plants in some pastures showing root rot symptoms. In some pastures up to 90% of plants in a stand were affected by the disease (Clarke 1983b). A subsequent study by Burnett et al. (1994) showed that decline in permanent pastures in north-eastern Victoria remains an on-going problem. Greenhalgh and Clarke (1985) used drenches of fungicides metalaxyl (against *Pythium* and *Phytophthora*) and benomyl (against *Fusarium*) in 1981 to study significance and etiology of root rot of subclover in dryland subclover pastures in Victoria. They examined sites located at Clunes in western Victoria north of Ballarat and demonstrated significant increases in subclover herbage production following fungicide application. Taylor et al. (1983) investigated root rot of irrigated subclover in northern Victoria, both its significance and the prospects of control. These studies demonstrated increases from disease control by fungicides of nearly 60% in May and more than 95% in October in herbage production from where the fungicide applications were made.

NSW: In NSW, a problem of poor re-establishment and poor forage and seed production in long-term subclover pastures has been recognized since the mid 1960's (Anon. 1965, 1968, 1969, 1970, 1971). Investigations by Stovold (1974a) showed that root rots were the most important factor in this observed decline of established subclover pastures. Hochman et al. (1990) examined the factors contributing to reduced productivity of subclover pastures on acid soils at four field sites with poor clover growth (Holbrook, Yerong Ck, Oberne, Lankey's Ck) in NSW and utilized multiple treatments including P, Mo, lime pelleted seed, lime additions, metalaxyl (against *Pythium* and *Phytophthora*), ryegrass competition, lime+Mg+trace elements. They demonstrated increases in herbage yields of the order of 10-12% from these treatments where root disease was reduced.

Western Australia: In WA, decline from root disease in subclover was first recognised by Shipton (1967b). Large areas of subclover in the south-west and south coastal districts have been affected by pasture decline due to root rot (MacNish et al. 1976; Gillespie 1983c), with heavy production losses resulting from severe root rot in situations where a big percentage of seedlings are killed even prior to emergence and where emerged seedlings die from root rot in the first few weeks of the growing season (Barbetti and MacNish 1983). Wong et al. (1985b) demonstrated that seedling losses in the field from damping-off could exceed 90%. Barbetti (1984e) showed an inverse relationship between the severity of rotting of the tap root system and plant size. The greatest reduction in plant size from root rot occurred from 6 or 7 weeks after emergence till 16 or 17 weeks into the growing season. Such reductions often exceeded 70% suggesting that production from pastures with severe tap root rot may be very poor. Wong et al. (1986a) described the inverse relationships between plant size and root disease for a range of soil-borne pathogens. Recently, O'Rourke et al. (2009) demonstrated that it was not just root disease on seedling and young subclover plants that was important, but that root disease on mature subclover plants can also be severe and significantly reduce productivity of subclover.

3. Most important soil-borne pathogens

Pre-emergence damping-off in subclover caused by *Pythium irregulare* and pre- and post-emergence damping-off combined with severe root disease in subclover pastures caused by *Phytophthora clandestina* constitute the over-riding biotic constraints to productive subclover pastures across southern Australia. A range of other pathogens are also very important and damaging, particularly *Aphanomyces trifolii* and *Rhizoctonia* spp., while in some situations one or more *Fusarium*, *Phoma* and *Cylindrocarpon* species can also be very damaging when acting as components of pathogen complexes with other soil-borne pathogens. Together, these pathogens constitute the main reason for pasture deterioration and decline. On a national basis, nematodes are considered of much lesser importance.

4. Need for further quantification of impact of soil-borne diseases

Except for the recent studies by Simpson et al. (2011) and O'Rourke et al. (2009), the impact and related causes of severe disease have been largely ignored for some decades. Field observations indicate that the situation has changed dramatically over recent decades and that a more relevant/recent reassessment is needed as there is a lack of sufficient information relating to incidence and impacts (losses and economic importance) of major soil-borne diseases in subclover across southern Australia. This information is fundamental to defining priorities for subclover pastures. It is apparent that many subclover pastures frequently fail to persist or be productive, particularly in high rainfall regions >500mm in situations where annual losses up to 40-50% occur.

5. Opportunities for managing soil-borne diseases from altered farm practices

5.1. Fungicides: There exist significant but rarely exploited opportunities for utilizing low cost chemical seed treatments to ensure successful stand establishment when sowing pastures (see MLA reports of Barbetti et al. 2005 and Barbetti and Jones 2011; Barbetti et al. reviews of 2005, 2007). For example, Taylor et al. (1983) demonstrated increases in Victoria from disease control by fungicides of nearly 60% in May and more than 95% in October in subclover herbage production. While Apron® is registered for control of damping-off, this chemical is comparatively expensive and targets only part of the pathogen complex associated with damping-off. There are many alternative fungicide treatments that not only would be more effective as they target a much wider range of soil-borne pathogens associated with damping-off, but also would be much cheaper, encouraging wide uptake and usage. There is also additional potential in that several cheap and potentially more effective possibilities simply have not been identified and/or evaluated for their potential roles in soil-borne disease management. It is clear that there are not only simple and cheap fungicidal seed treatments that could ensure successful stand establishment of pasture legumes, but would extend protection to roots and by provide additional benefits from reducing levels of root disease in seedlings. Benefits would be extensive particularly in the high rainfall regions (>500mm), where pre- and post-emergence damping-off by *Pythium* and *Phytophthora* alone currently result in pastures sometimes failing to persist or be productive and where annual losses of up to 40-45% losses occur in the most disease prone areas.

5.2. Farm cultural practices*:* There are several potential yet rarely exploited opportunities for utilizing manipulation of cultural practices for soil-borne disease management in pasture legumes (see MLA reports of Barbetti et al. 2005; Barbetti and Jones 2011; Barbetti et al. reviews of 2005, 2007). While the low level of utilisation of cultural management measures by meat producers arises partly from lack of awareness of their benefits, this is also partly a consequence of additional measures not having been identified or evaluated for their potential benefits. This is despite isolated historical information suggesting soil disturbance could reduce levels of damping-off and root disease in subclover for a period of three years; and that controlled grazing can reduce losses from *Phytophthora* root rot by up to 55% in situations where this disease is particularly severe.

5.3. Nutrition: It is evident that soil fertility affects the severity of disease on subclover, by influencing root physiology and host resistance. Both better utilization of applied fertilizer and improved management of root diseases of subclover by utilising nutrient amendments is possible, in particular from enhancement of plant growth and defences and also the provision of nutrient bases for the effective activity of the resident microbial populations that offer biological buffering against major pathogens. There is significant potential for improved management of major soil-borne pathogens of subclover from a better understanding of the full potential for manipulating host nutrition for improved disease control. Nutritional deficiencies such as those which occur in the old, leached soils across southern Australia are known to be able to reduce the natural resistance of plants to disease (Graham, 1983). It is however generally accepted that root disease on subclover is frequently exacerbated by nutritional stress imposed on the plant hosts. In summary, while correction of these deficiencies in most cases reduces the severity rather than the incidence of diseases, it still offers some opportunities for exploitation as plant nutrients can reduce the severity of diseases through enhancement of plant growth and disease tolerance. This is highlighted by current research in WA that highlights the potential that improvement of the nutritional status of the nutrient-impoverished soils can make in reducing root disease and the opportunities for such practices to be integrated into a more sustainable approach to the management of root disease in subclover pastures (O'Rourke et al., unpubl.).

5.4. pH amendment

Barbetti (1990) demonstrated that changes in the soil pH (from the addition of lime) offers significant opportunities for reducing pre- and post-emergence seedling losses and root disease and that addition of lime to acid soils shows significant potential for management of root disease. However, it was evident from the Plant Health Workshop on 29th February that adding lime generally does not improve productivity of pastures in eastern Australia.

6. Environnemental influences on soil-borne pasture diseases

Environmental factors such as rainfall (soil moisture) and soil temperature have been clearly shown to have a marked effect on both the root disease severity on subclover from individual pathogens and on the interactions that occur between or among the different root pathogens. Further investigations in this area is needed to explain the differences in root disease symptoms and severity that occur between different areas across southern Australia as this is fundamental to relevant interpretation of SARDI DNA tests. It is clear that the fluctuating soil temperature and moisture conditions so common across southern Australia strongly impact the expression of root disease in subclover. Historically, while there have been attempts to investigate the relevance and importance of environmental factors in relation to the severity of root diseases of subclover (e.g., temperature and moisture, Wong et al. 1984), the known range of soil-borne pathogens (and their complexes) primarily responsible for root disease has significantly altered over recent decades, as has the temperature and moisture conditions now occurring across southern Australia. Such climate changes as occurring in southern Australia are important as the distribution of the most important pathogens such *Phytophthora* and its races (You et al. 2006) and *Pythium* (Barbetti unpubl.) are determined by rainfall. Further, there are significant interactions between temperature and moisture in relation to expression and severity of root disease (Wong et al. 1984). In summary, environmental factors such as rainfall, soil moisture and soil temperature are major factors affecting both the root disease severity in subclover from individual pathogens and on the complex interactions that occur between the different root pathogens.

7. Why address these soil-borne disease constraints to pasture productivity

It is clear as outlined above that reduction of soil biological constraints to pasture productivity will result in very significant increases in pasture production, both in quantity and reliability of feed production, and nitrogen fixation as well as persistence of key species such as legumes. Barbetti (2007) has also highlighted that reducing the impacts of soil borne root pathogens in soils will enhance efficient utilisation of pasture inputs; pasture palatability; plant nutritional value; seed set, viability and persistence; pasture composition in favour of legumes and resilience to grazing.

It is acknowledged that a desirable long term approach to minimise losses due to soil borne diseases in pasture is by understanding the mechanisms by which pasture plants can resist infection and colonisation of plant roots, and by improving our understanding of the genetics of resistance in desirable pasture species. However, this approach is considered a much longer term proposition and outside the scope of this proposal.

8. Securing pathology skills for pastures into the future

Currently, Barbetti remains the last remaining full time pasture plant pathologist in Australia with recognised expertise in soil-borne pasture diseases and with combined competency skills across the range of fungal and oomycete soil-borne pathogens involved. He has four decades of successful IDM outcomes and package delivery for the benefit of Australian pasture-based industries across the disease spectrum of annual and perennial legumes and grasses for southern Australia. This expertise will be lost once Barbetti retires in ~ 6-8 years' time. There is, therefore, an urgent need to train a pasture pathologist and capture the existing pasture pathology knowledge of Barbetti, in order to safeguard the feed-base across Australia from major disease losses from soil-borne pathogens into the future. Further, this project offers the unique opportunity to not only train this pathologist, but to do so in the most cost-effective way by employment of this person as the main researcher on this project.

CAPACITY OF PRINCIPAL INVESTIGATOR TO DELIVER PROMISED OUTCOMES

Recent publications relating to pasture research – Martin Barbetti

Scientific Articles in Refereed Journals in the Last 7 Years in Relation to Pasture Research

1. Barbetti MJ. (2005). *Cylindrocarpon didymum* - a root pathogen of subterranean clover in the lower south west of Western Australia. *Australasian Plant Pathology* 34: 111-114.
2. Barbetti MJ, Allen JG. (2005). Association of *Fusarium* species, with potential for mycotoxicosis, on pods of annual *Medicago* in Western Australia. *Australian Journal of Agricultural Research* 56: 279-284.
3. Barbetti MJ, Nichols PGH. (2005). Field performance of subterranean clover germplasm in relation to severity of Cercospora disease. *Australasian Plant Pathology* 34: 197-201.
4. Barbetti MJ, Nichols PGH. (2005). New sources of resistance in *Trifolium subterraneum* to rust (*Uromyces trifolii-repentis*). *Australian Journal of Experimental Agriculture* 45: 1163-1166.
5. Barbetti MJ, Si P, Nichols PGH. (2005). Genetic basis for and inheritance of resistance to Race 1 and Race 2 of *Kabatiella caulivora* in *Trifolium subterraneum* ssp. *subterraneum* and ssp. *yanninicum*. *Euphytica* 144: 237-246.
6. You MP, Barbetti MJ, Nichols PGH. (2005). New sources of resistance in *Trifolium subterraneum* L. to root rot caused by two races of *Phytophthora clandestina* Taylor, Pascoe and Greenhalgh. *Australian Journal of Agricultural Research.* 56: 271-277.
7. You M, Barbetti MJ, Sivasithamparam K. (2005). Characterization of *Phytophthora clandestina* races on *Trifolium subterraneum* in Western Australia. *European Journal of Plant Pathology* 113: 267-274.
8. You M, Barbetti MJ, Nichols PGH. (2005). New sources of resistance identified in *Trifolium subterraneum* breeding lines and cultivars to root rot caused by *Fusarium avenaceum* and *Pythium irregulare* and their relationship to seedling survival. *Australasian Plant Pathology* 34: 237-244.
9. You M, Barbetti MJ, Nichols PGH. (2005). New *Trifolium subterraneum* genotypes identified with combined resistance to race 2 of *Kabatiella caulivora* and cross-resistance to fungal root rot pathogens. *Australian Journal of Agricultural Research* 56: 1111-4.
10. You MP, Simoneau P, Dongo A, Barbetti MJ, Hua Li, Sivasithamparam K. (2005) First report of an Alternaria leaf spot caused by *Alternaria brassicae* (Berk.) Sacc. on *Crambe abyssinicia* in Australia. *Plant Disease* 89: 430.
11. Barbetti MJ, Riley IT. (2006). Field application of *Dilophospora alopecuri* to manage annual ryegrass toxicity caused by the toxigenic bacterium *Rathayibacter toxicus*. *Plant Disease* 90: 229-232.
12. Nichols PGH, Evans PM, Craig AD, Sandral GA, Dear BS, Barbetti MJ, Si P, You MP. (2006). Napier subterranean clover (*Trifolium subterraneum* L. var. *yanninicum* (Katz. Et Morley) Zohary & Heller). *Australian Journal of Experimental Agriculture* 46: 1109-12.
13. Nichols PGH, Sandral GA, Dear BS, de Koning CT, Lloyd DL, Evans PM, Craig AD, Barbetti MJ, Si P, You MP. (2006). Urana subterranean clover (*Trifolium subterraneum* L. var. *subterraneum* (Katz. Et Morley) Zohary and Heller). *Australian Journal of Experimental Agriculture* 1112 46: 1105-1107.
14. You M, Barbetti MJ, Sivasithamparam K. (2006). Occurrence of *Phytophthora clandestina* races across rainfall zones in south west Western Australia. *Australasian Plant Pathology* 35:85-87.
15. Barbetti MJ. (2007). Resistance in annual *Medicago* spp. to *Phoma medicaginis* and *Leptosphaerulina trifolii* and its relationship to induced production of a phyto-oestrogen. *Plant Disease* 91:239-244.
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17. Nichols PGH, Loi A, Nutt BJ, Evans PM, Craig AD, Pengelly BC, Lloyd DL, Dear BS, Revell CK, Nair RM, Ewing MA, Howieson JG, Auricht GA, Howie JH, Sandral GA, Carr SJ, de Koning CT, Hackney BF, Crocker GJ, Snowball R, Hughes SJ, Hall E, Foster KJ, Skinner PW, Barbetti MJ, You MP. (2007). New annual and short-lived perennial pasture legumes for Australian agriculture - 15 years of revolution. *Field Crops Research* 104: 10-23.
18. Nichols PGH, Sandral GA, Dear BS, De Koning CT, Lloyd DL, Evans PM, Craig AD, Barbetti MJ, Si P, You MP. (2007). Coolamon subterranean clover (*Trifolium subterraneum* L. var. subterraneum (Katz. et Morley) Zohary and Heller). *Australian Journal of Experimental Agriculture* 47: 223-225.
19. Nichols PGH, Sandral GA, Dear BS, De Koning CT, Lloyd DL, Evans PM, Craig AD, Nutt BJ, Barbetti MJ, Si P, You MP. (2007). Izmir subterranean clover (*Trifolium subterraneum* L. var. *subterraneum*). *Australian Journal of Experimental Agriculture* 47: 226-229.
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21. Nichols PGH, Barbetti MJ, You MP. (2008). Inheritance of resistance to race 1 of *Kabatiella caulivora* in subterranean clover (*Trifolium subterraneum* L.) cultivars and breeding lines. *Australian Journal of Agricultural Research* 59: 561-566.
22. Riley IT, Barbetti MJ. (2008) Australian anguinids and their agricultural impact and control. *Australasian Plant Pathology* 37: 289-297.
23. You MP, Lancaster B, Sivasithamparam K, Barbetti MJ. (2008). Pathogenicity of different zymogram groups of the root rot pathogen *Rhizoctonia solani* on pasture legumes in Western Australia and implications for rotational crops. *Plant & Soil* 302: 203-11.
24. Ma K, Kong P, You MP, Hua Li, Sivasithamparam K, Barbetti MJ. (2009). Molecular characterization of isolates belonging to eight races of *Phytophthora clandestina*. *Australasian Plant Pathology* 38: 608-616.
25. Nichols PGH, Dear BS, Hackney BF, Craig AD, Evans PN, de Koning CT, Foster KJ, Barbetti MJ, You MP, Micic S. (2009). New subterranean clovers with reduced cotyledon susceptibility to redlegged earth mites. In: *Proceedings of the 14th Australasian Plant Breeding Conference and 11th Society for the Advancement of Plant Breeding in Asia and Oceania (SABRAO) Congress 2009,* Cairns, 10-14 August 2009.
26. Nichols PGH, Snowball R, D'Antuono MF, Barbetti MJ. (2009). Resistance to clover scorch (*Kabatiella caulivora*) among accessions of purple clover (*Trifolium purpureum*) and its relationship to eco-geography of collection site. *Crop & Pasture Science* 61: 40-4
27. O'Rourke TA, Scanlon TT, Ryan MH, Wade LJ, McKay AC, Riley IT, Hua Li, Sivasithamparam K, Barbetti MJ. (2009). Severity of root rot in mature subterranean clover and associated fungal pathogens in the wheatbelt of Western Australia. *Crop and Pasture Science* 60: 43-50.
28. You MP, Yang HA, Sivasithamparam K, Barbetti MJ. (2009). A new leaf blight disease of *Trifolium dasyurum* caused by *Botrytis fabae*. *European Journal of Plant Pathology* 123: 99-103.
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33. O'Rourke, T.A., Ryan, M.H., Hua Li, Ma, X., Sivasithamparam, K. Fatehi, J. and Barbetti, M.J. (2010). Taxonomic and pathogenic characteristics of a new species *Aphanomyces trifolii* causing root rot of subterranean clover (*Trifolium subterraneum*) in Western Australia. *Crop and Pasture Science* 61: 708–720.
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38. Li, Y.P., Wright, D.G., Lanoiselet, V., Wang, C.P., Eyres, N., Real, D., You, M.P. and Barbetti, M.J. (2012). First Report of *Phoma herbarum* on tedera (*Bituminaria bituminosa* (L.) C.H. Stirton var. *albomarginata*) in Australia. *Plant Disease* (accepted 22 January 2012).
39. Tan, D., Flematti, G.R., Ghisalberti, E.L., Sivasithamparam, K. and Barbetti, M.J. (2012). Mycotoxins produced by *Fusarium* spp. associated with Fusarium head blight of wheat in Western Australia. *Mycotoxin Research* (in press; DOI 10.1007/s12550-011-0122-7)

Review Articles and Book Chapters in the Last 7 Years in Relation to Pasture Research

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7.4 Draft Contract Schedule - Project for Scenario 1

SCHEDULE

Research Organisation

Name	The University of Western Australia
ABN	37-882-817-280
Street Address	35 Stirling Highway, Crawley, WA 6009
Postal Address	M459, Research Grants and Finance
	Research Grants Office (Research Services)
	The University of Western Australia
	35 Stirling Highway, Crawley, WA 6009
	(Attn Mrs Christine Casey)
Project Leader:	
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MLA

Meat & Livestock Australia Limited	**ABN**	39 081 678 364
Street Address	Level 1, 165 Walker Street North Sydney NSW 2060	
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Project

Project No.	**PSP.0005**		
Project Title	**Managing Soil-borne Root Disease in Sub-clover Pastures**		
Start date	1 July 2012	**Completion date**	30 June 2017

Purpose and description

A shortage of pasture feed during the autumn-winter period in southern Australian pasture systems is a major constraint to livestock production and profitability. Any extra carrying capacity during this period results in better pasture utilisation in spring and contributes directly to producer profit.

This period of critical feed shortage coincides with the dominant prevalence and intensity of soil borne root pathogens inducing significant production loss of pasture plant species. Documented losses of up to 45% are common, comprising 30-90% of seedlings failing to emerge, 10-40% of surviving seedlings succumbing post-emergence and 35-75% reduction in growth of surviving plants (e.g. Barbetti 2005). These losses strongly reduce the persistence of sown pasture species, adding further to the risks for producers to invest in pasture improvement.

Of most significance is the rapid deterioration and lack of persistence of sub-clover pastures due to soil borne disease (e.g. Barbetti et al 2006). Also, recent evidence from Simpson and Richardson (2009) has estimated that soil borne root pathogen induced seedling loss and root damage in clovers and grasses in southern Australian pastures during this autumn-winter pasture are causing between 25% and 58% loss of potential net farm income. Much of this pasture plant productivity loss is “sub-clinical” in that visual symptoms are not obvious so that the opportunity cost is not recognised.

The primary purpose of this project is to better understand the factors that influence the expression of disease in southern Australian sub-clover pastures, broaden the quantification of pasture productivity losses and develop practical measures that producers can adopt to reliably and significantly reduce the impact of soil borne pathogens on the productivity and persistence of their pastures.

Glasshouse and field experiments will be conducted to evaluate and select the most promising biocide, pasture cultural practices and nutrition treatments that producers can adopt to better manage soil-borne pasture diseases. Regional, environmental and edaphic factors that influence the dominance of particular pathogens and pathogen complexes in relation to the expression of disease (symptoms and sub-clover productivity) will be evaluated to help better formulate management strategies.

Further, molecular assays which are useful to identify and quantify pathogens present in soils and roots will be expanded and validated as research tools.

The project will also focus on training a pasture pathologist – soil biologist as an investment in maintaining and enhancing the knowledge on soil borne disease management which will be available to producers and future research.

The project will seek to interact and co-operate with complementary research projects on pasture root diseases (e.g. B.PUE.0103 - which is focussed on the interaction between phosphorus nutrition and root disease of sub-clover pastures).

The anticipated benefit from this project is a c. 25% improvement in productivity in autumn-winter sub-clover pastures affected by soil borne disease in southern Australian pasture systems

Objectives

The Research Organisation will achieve the following objective(s) to MLA's reasonable satisfaction:

***Objectives*:**

This project seeks to enable producers to reliably enhance the productivity from their pastures by c. 25% by:

13. Determining the farm management factors that influence the expression of soil-borne root diseases in sub-clover pastures.
14. Identifying effective and practical management techniques that reliably reduce the pasture productivity loss during autumn-winter induced by soil-borne root disease.
15. Characterising the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots.
16. Broadening the quantification of pasture productivity constraints induced by pasture soil-borne root pathogens across agro-ecological zones.
17. Developing molecular assays to fill gaps in experimental tools needed to adequately conduct research into, and monitor, soil borne diseases in pastures.
18. Securing pasture pathology skills and expertise for the future

These objectives also, in part, meet the objectives of the "Plant Health" research recommendations viz:

- Evaluation of non-breeding approaches to fungal and viral disease management, including cultural practices
- Identification of the impact of changes in farm practices on rhizobial function and nitrogen fixation.

Additional details

Background

Following the preparation of a discussion paper suggesting priority topics for pasture soil biology research, a soil biology workshop involving producers, advisors and researchers agreed that the objectives described above aimed at better managing soil borne diseases of sub-clover pastures were the most effective way to help producers enhance pasture productivity across southern Australia. The goal is to improve pasture plant health and production during critical feed shortage periods (autumn-winter) by reducing soil biological constraints imposed by root pathogens.

Planned outputs from this project

1. Provision of the knowledge and tools for meat producers to substantially increase sub-clover productivity in permanent pastures by at least 25% through better understanding and practical management of soil-borne root diseases.
2. Definition of extent that practical on-farm management practices can mitigate the expression of soil-borne root diseases of sub-clover and consequent plant performance.
3. Delivery and demonstration of the most effective practical management options identified to producers in field sites across southern Australia for their capacity, reliability and impact in improving sub-clover productivity in the face of soil-borne disease challenges.
4. Definition of relationships between the expression of disease on plants with the associations of particular soil-borne pathogens and how environmental influences determine the outcomes of these interactions and relationships. These are essential to define both the different symptom expressions on roots and the relative impacts of different individual pathogens and pathogen complexes in different regions and across different seasons, and also provide a basis for interpreting molecular DNA tests for producers
5. Quantification of the pasture productivity constraints induced by soil-borne disease of sub-clover across agro-ecological zones, also providing a focus for where and what type of future funding decisions are justified.
6. Training of a new post-doc scientist with skills in soil-borne plant pathogens to ensure availability of these skills for producers and research into the future.
7. Provision of the knowledge, pathogen isolates, soil and diseased plant samples, needed for SARDI to develop, refine and validate a package of molecular DNA assays of soil-borne pathogens useful as research and monitoring tools for better management of soil-borne diseases in pastures.

Planned outputs consistent with the recommended research priorities of the “Plant Health” program in the Feedbase investment plan include:

- Practical farm management practices to reduce fungal disease impacts in sub-clover pastures

- Quantification of the impact of changes in farm management practices on nitrogen fixation in sub-clover. This output will be enhanced by integration with improved root health through soil borne disease management.

Methods

Objectives 1 and 2:

1: Determine the farm management factors that influence the expression of soil-borne root diseases in sub-clover pastures

2: Identify practical management techniques aimed at reducing the pasture productivity loss during autumn-winter induced by soil-borne root disease

i) *Years 1 and 2* – In this first phase, field (and complementary glasshouse) trials will be conducted to test:

- a range of pathogen biocide and resistance inducing chemicals (seed and foliar),
- cultural management practices (e.g. variety, rhizobia, grazing, cultivation, pH, nutrition, altered pasture composition etc – 10-15 practices will be screened)

for control of sub-clover seed infections that affect germination and emergence as well as seedling root disease during the winter-autumn period.

Best prospect treatments will be identified from exiting literature (Australian and international) and the local researcher knowledgebase.

The initial field screenings of prospective treatments will be undertaken in small field plots at Shenton Park Field Station in Western Australia, on land that has a history of several decades of sub-clover pastures.

Reductions in root disease from the treatments will be assessed on the plants as will responses in plant growth. Symptoms will be noted and root samples retained to extract root and pathogen DNA.

From these initial field testings, the most promising and cost-effective chemical treatments and cultural practices shown to have effective control of pre- and post-emergence damping-off and seedling root disease in sub-clover pastures will be identified.

ii) *Years 3 and 4* – In this phase, the most promising root disease management treatments will be demonstrated nationally in at least eight field trials (two each per WA, SA, Vic, NSW)

Both chemical and cultural practice treatments will be demonstrated in separate replicated trials at each site. Each experiment will trial at least 3 chemical or cultural practice treatments compared to a nil-treatment control.

Reductions in root disease from each treatment will be assessed on plants as will responses in plant productivity.

Composite soil and root samples will be taken from each plot at 3 and 9 weeks after the commencement of the growing season and sent to SARDI to molecularly confirm and quantify the differences in pathogen population and density present.

These field demonstration trials will be conducted in conjunction with Agronomists and consultants and in association with the pasture agronomy and extension personnel across Australia. Producer engagement with these trials will be encouraged to demonstrate the effectiveness and reliability of the treatments for better [pasture performance during autumn-winter.

The outputs from these trials will be packaged into MLA extension materials including "Tips & Tools" publications for dissemination among producers with advice on how best to manage soil borne diseases in sub-clover pastures.

Objective 3: Characterise the relationships between the expression of disease and the associations of soil-borne pathogens with affected plant roots

Relationships between the expression of disease and the associations of soil-borne pathogens infecting sub-clover roots will be determined in three elements:

Glasshouse trials – will be established to:

- define the relationships between expression of disease on plant roots with associations of individual and combinations of the major root pathogens known to affect sub-clovers
- explain the differences in symptoms, disease severity and expression as affected by varieties of different levels of host resistance and different temperature, moisture, soil type and nutritional regimes representative of regional and environmental conditions across southern Australian sub-clover pastures .

Expression of root disease will be assessed for all plants in each treatment as will correspondingly the effects on plant foliage and root productivity.

Calibration and validation of pathogen DNA assays:

Once these relationships have been defined, representative samples from the experiments described above will be sent to SARDI to enable calibration and validation of molecular DNA tests in terms of disease expression and quantification of the different pathogen populations present.

Field expression of sub-clover root diseases

In years 4 and 5, field studies will be undertaken to define relationships between field expressions of root disease with the main environmental factors consistent with the agro-ecological zones for southern Australia. This will also involve collection and collation of relevant environmental data across southern Australia. Then available data will be incorporated into an integrated modelling package to simulate potential risk(s) associated with deployment of different host cultivars, for different seasonal conditions and for different geographic scenarios occurring across the southern Australian pasture belt. This modelling will be utilised to not only define the field influence of environment on disease expression across the agro-ecological zones of southern Australia, but, more importantly enable ability to define the relative risks for producers in different regions and across different seasons.

Objective 4: Broaden the quantification of pasture productivity constraints induced by pasture soil-borne root pathogens across agro-ecological zones

In year 3, in conjunction with the field trials outlines for Objectives 1 and 2, field bioassay experiments, based the Wong et al. (1984) – Simpson et al. (2009) bioassay technique, will be established at 8 sites (2 in each of NSW, Vic, SA and WA) to quantify the losses occurring at pre- and post-emergence and on seedling and mature plants throughout the growing season. These trials will be maintained over two years.

Treatments and replication will allow for sampling and root assessment for disease and rhizobial colonization to be monitored throughout the growing season to allow quantification for any time or period of the season.

At each site:

- trials will be established on both natural and biocide fumigated soils to assess the potential production capacity in the absence of soil borne root pathogens
- sub-clover varieties will consist of one known susceptible variety and one with an effective level of field resistance to root disease
- assessments will be undertaken at 3, 9 and 20 weeks after sowing to determine the impact of disease from germination to plant maturity
- isolations and identification of pathogens from diseased roots of plants will be made to define the exact cause(s) of the disease expressed and the associated quantified losses occurring at critical times of the season
- at each assessment event, soil and plant samples will be sent to SARDI where DNA methods will be used to quantify pathogen(s) and their combinations occurring in sub-clover roots and to calibrate predicted levels from DNA tests to actual levels of root disease and root damage. This data will be linked to estimates of pasture production, as expressed across treatments at each field location. At SARDI, DNA will be extracted from composite soil/root samples and, after analysis, archived for future reference.
- In years 4,5, data from these field trials will be used in appropriate modelling to provide both the dollar losses incurred from varying levels of soil-borne disease and associated yield and productivity losses.

All direct disease assessments and isolation of pathogens will be addressed and delivered through UWA. However, this field trial network will be conducted in association with pasture extension and agronomy programs across southern Australia for the establishment, maintenance and sampling of the trials outside of WA. These will be negotiated with research teams across various agencies with the capability to undertake the work to a high standard.

UWA will visit these sites regularly and assist with sampling and observations to the extent possible to ensure experimental and sampling protocols are consistently adhered to across all sites.

Objective 5: Develop molecular assays to fill gaps in experimental tools needed to adequately conduct research into, and monitor, soil borne diseases in pastures

SARDI will be contracted to develop a new DNA assay for *Aphanomyces trifolii*. This pathogen has been identified as a major soil borne disease of sub-clover, but for which no DNA assays currently exists. SARDI is nationally recognised as the best group to develop such tests.

This new assay will add to a suite of other tests for pastures root pathogens which were developed as part of MLA's previous pasture soil biology program. This new assay, once developed, can be applied retrospectively to archived DNA extracted from soils in the early parts of this project.

Objective 6: Secure pathology skills and expertise for pastures into the future

A Post Doctoral researcher will be appointed to this project to enhance and consolidate the technical and knowledge capacity on pasture plant pathology – soil biology available for future research and extension in southern Australian pasture systems.

The postdoc pathologist will collaborate and work closely with the new MLA-AWI project B.PUE.0103 that will examine interactions between root disease and plant P efficiency for subclover providing both learning for the postdoc pathologist and provision to the MLA-AWI project B.PUE.0103 project of the needed pathology support from both the postdoc pathologist and Barbetti.

In addition, the postdoc pathologist will have exposure by direct visits to other relevant projects that emerge from the MLA "Plant Health" theme of the Southern Feedbase Investment Plan; and the Feedbase Investment Plan on "Pasture legume performance and proportion", a project that will have direct relevance to legume productivity and seed bank persistence; and finally, from visits to related networks associated with these projects.

Also, in addition to Barbetti, the postdoc pathologist will be supported by Megan Ryan at UWA in relation to development of novel pasture species and associated mycorrhizal and rhizobial associations and by Phil Nichols the pasture group at DAFWA in relation to the breeding, selection and agronomy of pasture species with improved disease resistance, and through these, access to other relevant collaborative networks.

Also, the postdoc pathologist will have the opportunity to acquire skills in relation to development and interpretation of molecular DNA testing for root diseases.

References

Barbetti MJ (2006) Role and impact of diseases caused by soil-borne plant pathogens in reducing productivity in southern Australian pasture systems. Final project report to MLA (SHP.016)

Simpson RJ, Richardson AE (2009) Root damage & soil biology profiles of autumn-winter pastures. *Final project report to MLA* (SHP 0025).

Wong DH et al (1984) Effects of soil temperature and moisture on the pathogenicity of fungi associated with root rot of subterranean clover. AJAR 35; 675-684

Agents or subcontractors

Subject to the obligations relating to agents and subcontractors, MLA consents to the engagement of the following agents or subcontractors:

South Australian Research and Development Corporation (SARDI)

CSIRO

DPI NSW

DPI Vic

Private extension consultants, meat producers and producer groups as agreed by MLA

Interest

Company Name	Percentage
MLA	64%
UWA	36%

Background IP

Company Name	Description

Communications

Subject to the confidentiality obligations, the Project will be communicated by the Research Organisation:

Activity	**Key Message**
1.	The outputs from the project will feed directly into MLA's planned Feedbase Investment Plan communication strategies. They will also be communicated through MLA's developing "Producer Engagement" networks. Outputs will provide material for industry training modules and link in with relevant workshops arranged to extend technology emerging from the Feedbase Plan to producers and advisors.
2.	Clear techniques for producers to adopt in better managing soil borne diseases in sub-clover pastures. Definition of the factors that affect expression of disease in sub-clover pastures and risk assessment tools. Provision of calibrated molecular assays suitable for research and monitoring of soil borne diseases in pastures.
3.	Publication of scientific papers in the international peer reviewed literature. Articles will be prepared for dissemination in MLA (Feedback /Prograze or Frontier) publications, rural press articles and farm journals. Key information communicated at Farmer talks etc.
4.	Presentation of results to national science and advisory group meetings
5.	Milestone reports and a mid-term review that document progress towards the objectives of the research A Final Report submitted to MLA Scientific paper(s) for submission to peer-reviewed journals

Milestones

Milestone	Achievement Criteria	Due Date
1	Contract signed	1 July 2012
2	Milestone Report – reporting appointment of a post-doc pathologist, review of relevant literature, treatments and research plans for objectives 1 and 2 identified.	1 Dec 2012
3	Milestone Report – detailing putative best chemical and cultural measures to control sub-clover root disease (Objectives 1, 2); progress in characterizing associations between disease expression and pathogen complexes and environmental factors (Objective 3); progress in development of new pathogen DNA assays (objective 5); progress of past-doc pathologist in acquiring skills and knowledge as well as performance (objective 6).	30 Jun 2013
4	Milestone report – detailing progress in defining root disease control measures (Objectives 1 and 2); progress in characterizing associations between disease expression and pathogen complexes and environmental factors (objective 3); progress in development of new pathogen DNA assays (objective 5).	1 Dec 2013
5	Milestone Report – defining the most promising cultural methods to control soil borne disease for field demonstration (objectives 1, 2); conclusions of the glasshouse studies defining associations between pathogens, disease and environmental factors and progress in calibration and validation of DNA assays (objective 3); development of new DNA assay for *Aphanomyces* (objective 5); progress of past-doc pathologist in acquiring skills and knowledge as well as performance (objective 6)	30 June 2014

Milestone	Achievement Criteria	Due Date
6	**Mid-term Review** – mid-term report reporting achievements and progress against all objectives including progress in demonstrating cultural control practices in national field trials (Objectives 1,2); calibration and validation of pathogen DNA assays (objective 3); progress in quantification impacts of soil-borne disease on sub-clover productivity (objective 4); development and deployment of existing and new DNA assays in field experiments (objective 5); progress of past-doc pathologist in acquiring skills and knowledge as well as performance (objective 6). Failure to show progress in meeting the objectives of the research at this milestone will trigger a review of whether the project should proceed any further.	1 Dec 2014
7	Milestone Report – reporting the impact of chemical and cultural control measures for soil-borne disease in national field trials (objective 1,2); progress in field definition of the relationships between disease expression and association with soil-borne pathogens and environmental factors and development of modeling package (objective 3); progress in quantification of impacts of soil-borne disease on sub-clover productivity (objective 4); development and deployment of existing and new DNA assays in field experiments (objective 5); progress of past-doc pathologist in acquiring skills and knowledge as well as performance (objective 6); progress in producer and advisor engagement to disseminate information on emerging project outputs (communication obligations)	30 June 2015
8	Milestone Report – reporting the impact of chemical and cultural control measures for soil-borne disease in national field trials (objective 1,2); progress in field definition of the relationships between disease expression and association with soil-borne pathogens and environmental factors and development of modeling package (objective 3); progress in quantification of impacts of soil-borne disease on sub-clover productivity (objective 4); development and deployment of existing and new DNA assays in field experiments (objective 5); progress of past-doc pathologist in acquiring skills and knowledge as well as performance (objective 6); progress in producer and advisor engagement to disseminate information on emerging project outputs (communication obligations)	1 Dec 2015

Milestone	Achievement Criteria	Due Date
9	Milestone Report – reporting the impact of chemical and cultural control measures for soil-borne disease in national field trials (objective 1,2); progress in field definition of the relationships between disease expression and association with soil-borne pathogens and environmental factors and development of modeling package (objective 3); progress in quantification of impacts of soil-borne disease on sub-clover productivity (objective 4); development and deployment of existing and new DNA assays in field experiments (objective 5); progress of past-doc pathologist in acquiring skills and knowledge as well as performance (objective 6); progress in producer and advisor engagement to disseminate information on emerging project outputs (communication obligations)	30 June 2016
10	Milestone Report – confirming the best chemical and cultural control measures for soil-borne disease in sub-clover pastures (objective 1,2); confirming definition of the relationships between disease expression and association with soil-borne pathogens and environmental factors and development of modeling package (objective 3); quantification of impacts of soil-borne disease on sub-clover productivity completed (objective 4); development and deployment of existing and new DNA assays in field experiments (objective 5); achievements of past-doc pathologist in acquiring skills and knowledge as well as performance (objective 6); progress in producer and advisor engagement to disseminate information on project outputs (communication obligations)	1 Dec 2016
11	Final Report – detailing all progress in achieving the project milestones, including draft scientific publications and extension materials.	30 June 2017

A milestone is not achieved unless it is completed to MLA's reasonable satisfaction

Nominated Person(s)

Contact Name: Dr Martin Barbetti

Phone: 08-64883924

Fax: 08-64887077

Email: martin.barbetti@uwa.edu.au

Budget

Total Budget	Post Doc Salary	$641,920
	Operating expenses	$588,852
	Capital	0

Total Funds **AUD $1,230,772 (GST exclusive)**

Cash flow

Payment Date	Milestone	Fees	Expenses	Capital	Total
1 Jul 2012	1 *	50,000	50,000	0	100,000
30 Jun 2013	2 , 3**	60,000	40,000	0	100,000
30 Jun 2014	4, 5**	120,000	50,000	0	170,000
30 Jun 2015	6, 7 **	130,000	230,000	0	360,000
30 Jun 2016	8, 9 **	130,000	200,000	0	330,000
30 Jun 2017	10,11 ***	151,920	18,852	0	170,772
TOTAL					**AUD $1,230,772**

NOTE: Total overheads funding contribution from UWA = $689,985

*or on signing of this agreement

Payment Date	Milestone	Fees	Expenses	Capital	Total

**on acceptance and approval of corresponding milestone report, with tax invoice and copy of receipts attached

***on receipt and acceptance of final report by MLA, with tax invoice for payment attached

NB: any money uncommitted at the end of the Project must be returned to MLA

Additional Notes:

UWA guarantees to provide all the necessary infrastructure support for the postdoc pathologist, including full access to laboratories, controlled-environment rooms, glasshouses and field experimentation sites, and meet all other indirect costs (phone, printing, technical and professional support, etc).

Please also note:

i. **That additional funding will also be sought from other sources (e.g. ARC) to further expand and build long-term capacity in pasture sciences, particularly pasture pathology;**
ii. **Prof Hans Lambers, Head of the School of Plant Biology at UWA, has indicated strong support for the postdoctoral fellow towards a tenure track at UWA after the initial 5 year period funded by MLA, subject to satisfactory performance. This aligns well with Plant Biology plans to further expand its research base towards underpinning the long-term feed-base requirements for the animal industries of Australia. Other plans include enhancing research capacity in pasture breeding, agronomy and nutrition.**

SIGNED AS AN AGREEMENT

Signed by **MEAT & LIVESTOCK AUSTRALIA LIMITED:**

………………………………………….. ……………………………………

Signature of witness

Peter Vaughan

General Manager

Livestock Production Innovation

………………………………………….

Name of witness (print)

Signed by **COMMONWEALTH SCIENTIFIC & INDUSTRIAL RESEARCH ORGANISATION a body corporate established by the Science & Industry Research Act 1949, through its DIVISION OF SUSTAINABLE AGRICULTURE FLAGSHIP:**

. . . . witness

. . . . …... Signature of Signature of authorised person

. . . .

Name of witness (print)

. . . .

Name of authorised person (print)

. . . . .Office held

7.5 Project Specification – Scenario 2

PART 2: FULL APPLICATION FORM

PARTIES

Research Organisation

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THE PROJECT

Using applied DNA methods to identify practices that improve sub-clover root health

Background of Research Work

Shortage of pasture feed during the autumn-winter period is a major constraint to livestock production and profitability in the southern region. Sub-clover dominates permanent pastures in this region and often suffers significant levels of root damage from soil borne pathogens. Even sub clover pastures that are perceived as being productive have been shown to produce less than 50% of their potential growth. Damage from soil borne disease and the production penalties they cause often go unrecognised in grazed pasture systems.

Simpson et al. (2009) concluded that the combined loss of seedlings and root disease penalties to surviving sub clover and grass species during autumn-winter reduced potential net farm income between 25% and 58%. These findings are consistent with studies of other pasture legumes. For lucerne in the SA Mallee, production losses due to soil borne disease have been estimated to be 50% (Ballard et al. 2009). For annual medics, average losses of 22% have been reported (Bellotti 2001).

Although many sub clover cultivars with combinations of disease resistance have been produced, their production benefits under different soil borne disease scenarios has not been adequately assessed, nor has their influence on paddock disease risk over time. To some extent this has been due to a lack of tools that enable the measurement of pathogen levels in soil. These tools are now available and will be used to highlight to producers the benefits of improved cultivars and how simple practices can be used to manage soil borne disease.

References: Simpson et al. 2011, Ballard et al. 2009, Bellotti 2001

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Project Description

This project will use applied DNA assays to simultaneously measure several pathogens in the soil and plant roots in grazed pastures to:

- Better understand how new cultivars and current management practices can be used to reduce the impact of soilborne diseases and increase profitability.
- Characterise relationships between pathogen levels in soil before the break and root disease levels during autumn/winter in key agro-ecological zones.

The project will link with:

- Project B.PUE.0103 "Root Disease Constraints in Pasture Productivity" which is focussed on the interaction between pasture phosphorus nutrition and root disease.
- Projects funded by the MLA "Plant Health" theme of the Southern Feedbase Investment Plan.
- Projects funded by the Feedbase Investment Plan on "Pasture legume performance and proportion".

The project will collect and archive soil extracted DNA for possible future studies on soil microbial community analysis including structure and function to assess impact of farm management on soilborne disease.

The project will seek to develop a pasture research capability to help address future RD&E needs of the livestock industry by mentoring a scientist in pasture soil biology who is equipped with a modern pathology skill set focussed around the use and interpretation of molecular assays.

Objectives and Outcomes of Project

Objective: To provide knowledge that will enable producers to increase autumn/winter subterranean clover productivity in permanent pastures by at least 15% through better use of current cultivars and management practices to reduce the impact of soilborne diseases.

Outcomes:

- Determine if new sub clover varieties and current farm practices can be used to manage soil pathogen levels and reduce autumn/winter production losses.
- Better understand the impact of environment and age of the pasture stands on the soil disease complex.
- Develop infrastructure, knowledge and experience to strategically use and interpret molecular assays to underpin industry needs for future research and address soil biology constraints to pasture production.

Method

This project will use high throughput quantitative DNA assays to study the pathogen, environment and host interactions that lead to poor root health and low autumn/winter production of sub-clover pastures.

i) Core field trials – managing soil borne disease

Four sub-clover cultivars, with different resistance to soilborne pathogens, will be sown into two field trials one each in south-east South Australia and western Victoria. Changes in soil pathogen levels and impact on root health in autumn/winter will be measured across five seasons to understand how the disease complexes are affected by seasonal conditions and change as the stands age.

An undisturbed treatment will be included at each site to assess the effect of pasture renovation on pathogen levels and disease expression. The trials will be grazed and exclusion cages used to estimate early production.

Work to select and characterise the sites will need to be undertaken in late 2012 so that the sites can be sown in 2013.

Use of DNA assays underpins this research to understand the disease complex, how it changes over time and the impact of different management strategies. This technology has both the capacity to process large numbers of samples and monitor changes in levels of each pathogen involved in the disease complex in soil and plant roots.

Soil samples will be collected prior to the break and at the end of the season to quantify changes in inoculum levels. Root disease levels and herbage production will be assessed 4 and 10 weeks after pasture germination.

Current DNA assays for pathogens of interest include *Pythium* clades F & I, *Phytophthora clandestina*, *Rhizoctonia solani* AG2.1, AG8, root lesion nematodes (neglectus, thornei and penetrans). DNA from all samples will be stored and retested as new tests become available e.g. *Aphanomyces trifolii*

In year four, the best management practice identified by the project proposed by Prof. Barbetti, will be applied to half of each plot (split plot design) to assess the potential to further modify the different pathogen complexes that are expected to have developed under the four sub clover cultivars.

Effects of the management practice on inoculum level, disease expression and early pasture production will be measured for each cultivar.

ii) Satellite sites

Strategic sampling of selected pasture trials and farmer paddocks will be undertaken to:

- raise awareness of the impact of root diseases
- aid understanding of treatment responses in other MLA pasture trials
- provide relative importance of individual pathogens at different locations
- identify which pathogens are most relevant to different regions
- help identify key pathogens not covered by existing tests.
- better understand the effects of environment on development of root disease

The extended work program will comprise three areas:

1. Characterisation of MLA pasture sites
   Provision for 80 soil and 80 plant samples is provided within the budget to profile pathogen levels in samples from other MLA pasture trials. Each sample will be assessed for five pathogens and a root health rating is also included for the plant samples. Additional samples can be assessed on a Fee for Service basis.

2. Satellite cultivar sites
   Small row trials using the method described by Simpson et al. (2009) will be established at five sites. These will include the four cultivars in the core trials, plus a fumigant control (e.g. Basimid) to assess production losses in a broader range of locations. These trials will be established adjacent to sub clover evaluation trials (e.g. at NVT or benchmarking trial sites) where practical, to raise awareness of potential disease constraints at those sites. Measures of soil inoculum and root health will be made as per the core trials.

3. 100 farm paddocks to better link pathogen level and root health
   The relationship between pathogen levels before the break, environment and variation in root health during winter will be investigated in years 4 & 5 using soil and plant samples collected from up to 100 paddocks from permanent pastures distributed from central NSW to mid northern region of SA. Most of these samples will need to be collected by collaborators in other pasture projects, given the broad geographic spread needed to generate sufficient environmental variation.

   Similar work using focus sites in cereal paddocks has shown that including measures of environment with pathogen DNA levels greatly improves estimates of root damage.

iii) Development of new DNA tests

Development of a test for *Aphanomyces trifolii* has already been identified as critical. The test will be developed at SARDI with funding provided from a complementary project proposed by Prof. Martin Barbetti (UWA).

Satellite sites (described above) will be used to indicate pathogens not covered by the current suite of DNA tests. Where a critical pathogen is not covered there will be an obvious discrepancy between the DNA tests and root damage. Pathogens will be isolated from diseased roots for future identification and prioritisation of DNA test development. New test development would be subject to additional funding.

iv) Mechanistic studies at post graduate level

While this project will focus on practical outcomes for graziers, there will opportunities for more detailed studies that would be suited to PhD studies. Indicative areas include:

- Modelling pathogen, environment and host interactions for expression of disease.
- Investigate use of novel root DNA assays to establish relationships between root growth, pathogen levels and pasture production during winter, and the potential of to use the assays to assess impacts of disease management.

The field sites, samples and their extracted DNA, access to test results and input into project directions could be provided from this project to a suitable University based and funded PhD candidate.

Interest (IP proportions)

MLA	76 %
SARDI	24 %

Milestones

Milestone Number	Achievement criteria	Due date
1	Appointment of Research Officer	Feb 28 2013
2	Pathogen levels and root damage quantified in year of pasture renovation, at two core sites.	Dec 31 2013
3	Characterisation of pasture trials (NVT/benchmarking sites) completed.	Dec 31 2014
4	New DNA assay for Aphanomyces available (linked to Barbetti project).	June 30 2015
5	Five row trials completed. Estimates of production losses available. Priorities and isolates for new DNA tests available.	Dec 31 2015
6	Management practice applied to core trials	June 30 2016
7	100 focus paddocks assessed to determine relationships between pathogen levels, environment and root health.	Dec 31 2016
8	Assessment of core trial completed. Recommendations on strategy to improve root health developed.	Dec 31 2017
9	Final Report	March 31 2018

Nominated Person(s)

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Project Budget and Funding – Indicative Budget

Year 1	**MLA Direct project costs**	**RO cash**	**RO salary**	**RO overhead**	**TOTAL Year 1**
Senior Research Officer (100%, TBA).	100,626				100,626
Casual labour (50d)	10,931				10,931
Ballard 20%, McKay 10%			40,127	46,725	86,852
Travel	13,500				13,500,
Operating	150,796				150,796
Equipment					
TOTAL BUDGET (excl. GST)	275,853		40,127	46,725	362,705

Year 2	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 2
Senior Research Officer (100%, TBA).	103,616				103,616
Casual labour (50d)	10,391				10,391
Ballard 20%, McKay 10%			41,524	46,725	88,249
Travel	13,500				13,500
Operating	153,724				153,724
Equipment					
TOTAL BUDGET (excl. GST)	281,231		41,524	46,725	369,480

Year 3

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 3
Senior Research Officer (100%, TBA).	106,696				106,696
Casual labour (50d)	10,931				10,931
Ballard 20%, McKay 10%			42,968	46,725	89,693
Travel	13,500				13,500
Operating	153,612				153,612
Equipment	0				0
TOTAL BUDGET (excl. GST)	284,739		42,968	46,725	374,432

Year 4

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 4
Senior Research Officer (100%, TBA).	109,868				109,868
Casual labour (50d)	10,931				10,931
Ballard 20%, McKay 10%			44,461	46,725	91,186
Travel	13,500				13,500
Operating	168,780				168,780
Equipment	0				
TOTAL BUDGET (excl. GST)	303,079		44,461	46,725	394,265

Year 5

	MLA Direct project costs	RO cash	RO salary	RO overhead	TOTAL Year 5
Senior Research Officer (100%, TBA).	113,136				113,136
Casual labour (50d)	10,931				10,931
Ballard 20%, McKay 10%			46,006	46,725	92,731
Travel	13,500				13,500
Operating	168,954				168,954
Equipment	0				
TOTAL BUDGET (excl. GST)	306,521		46,006	46,725	399,252

Budget Summary

	Year 1	Year 2	Year 3	Year 4	Year 5	TOTAL
MLA	275,853	281,231	284,739	303,079	306,521	1,451,423
Research Organisation contributions	86,852	88,249	89,693	91,186	92,731	448,711

Salaries are to support a Senior Research Officer at SARDI. A small provision for casual labour (40 days per year) is requested to assist with field sampling, root health assessments and preparation of samples for DNA analysis.

Operating costs include DNA analyses (approx. 550 samples per year @ $70 per sample), soil chemistry analyses ($5000), establishment, maintenance and sampling of the two core field sites is ($40,000 per year) and laboratory consumables. Collection of samples by collaborators has been costed at $150 per sample in years 4 & 5. Business support costs ($43,000 per year) are included in the operating budget.

Contributors/Other Funds

Options to fund this project will need to be negotiated. MLA, AWI or a combination of both are possible options.

Agent(s)/Subcontractor(s)

Name of Company **ABN** **Contact Person Title/First Name/Surname** **Mailing Address** **Phone Number** **Facsimile Number** **Email Address**	Note: The collection of samples from the 100 pasture sites is regarded as the minimum number needed. These samples will need to be collected by collaborators (still to be arranged) and provision has been made in the budget for this. If additional samples are needed to improve statistical rigour, both the funding and agreement of collaborators will need to be negotiated.

Communications and Delivery

Target groups

Raising awareness of the impacts of root diseases and their management for the Groups specified below is what can reasonably be delivered with the timeline and budget of the proposed research project. Broader industry adoption will be dependent upon the identification of practices that deliver significant benefit. This will begin in Years 4 and 5, but will mostly fall outside the timeframe of the current project.

Group	Total Potential Number of People
Consultants	40
Leading high rainfall pasture growers	150
Pasture researchers	20
RDC's	10
Pasture seed industry	10

Awareness/Participation/Adoption Objectives

Year following launch:	**Awareness (specified targets above)**	**Participation (all consultants)**	**Grower Adoption**
Year 1 - 2013	10%	0 %	0 %
Year 2 - 2014	20%	0 %	0 %
Year 3 - 2015	50%	0 %	0 %
Year 4 – 2016	75%	5 %	1 %
Year 5 - 2017	100%	10 %	3 %

Communication Products/Tools/Services

Production and distribution of simple diagnostic aids (e.g. similar to 'Back Pocket Guides' produced for GRDC.

Cost estimate included in budget? [ ] Yes [X] No

Communication/Delivery Channels

The principle channel of delivery will be though consultant networks and directly to leading growers. A consultant workshop will be run in the fifth year of the project. This will be supplemented with trial walks, diagnostic sessions and support, talks at pasture forums, production of media articles and direct interaction with growers and the seed industry.

A focus on raising consultant awareness of soil borne disease is seen as an important first step based on generally poor recognition of the problem by consultants in a recent survey completed for MLA by Mike Stephens and Associates.

Cost estimate included in budget? [X] Yes [ ] No

Other Capacity Building

This will largely be through developing the pathology skill set of the Research Officer employed in this project and development of industry linkages to improve the skill level of consultants.

Cost estimate included in budget? [x] Yes [ ] No

Timelines

This is a five year research project. Grower adoption will mostly take place and need to be resourced outside of this project.