Elucidation of the potential of biocontrol and biofertilizer inoculants to low-input crop production using genomic, metabolomic and conventional research methodologies

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End date: 30/04/2010

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Dr. Carl Ng, UCD
Dr. Ashley Franks, UCC
Dr. Mojibur Kahn, UCD

Please tick below the appropriate area on the research continuum where you feel this project fits

<table>
<thead>
<tr>
<th>BASIC/FUNDAMENTAL</th>
<th>APPLIED/PRE COMMERCIAL</th>
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<tbody>
<tr>
<td></td>
<td>X</td>
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</tbody>
</table>

Key words: (max 4)
Biocontrol, biofertiliser, cereals, disease
1. **Rationale for Undertaking the Research**

Biocontrol and plant growth-/soil health-supporting bacteria (biofertilisers) offer significant potential as pesticide and fertiliser alternatives and can be an eco-friendly, cost effective component of an integrated crop production system. We have identified a range of bacteria that (i) inhibit the economically important *Fusarium* diseases of barley and wheat and associated (and hazardous) mycotoxin contamination of grain and/or (2) promote plant growth. The ability of biocontrol bacteria to prevent mycotoxin contamination is particularly significant: new EU legislation dictates maximum permissible levels of mycotoxin in grain products (Commission Regulation No 856/2005), however, current strategies available for reducing mycotoxin contamination of grain are, at best, inconsistent in their efficacy. Plant growth-/soil health-supporting bacteria offer great potential as alternatives to chemical fertilisers (which are frequently associated with environmental pollution).

The aims of the project were to (i) assess the ability of selected bacteria to inhibit disease and promote the growth of barley and other plants under field and glasshouse conditions, (ii) characterise the plant growth-promoting and biocontrol bacteria with respect to their exact identity and (iii) determine their modes of action. Such studies are prerequisites for the optimization and commercialisation of enhanced biocontrol and biofertiliser formulations for Irish and international crop production systems.

2. **Research Approach**

- **Phylogenetic and morphological studies**: to identify the biocontrol and biofertiliser bacteria to the exact species/strain level.
- **Functional genomics (microarray and gene expression studies; bacterial mutagenesis)**: to identify/study bacterium-responsive plant genes and bacterial antifungal genes; such an approach helped elucidate the mode of action of biocontrol and biofertiliser bacteria. Future **plant transformation studies** will be used to study the function of bacterium-responsive plant genes (time and resources permitting, a limited number of such studies will conducted in this project).
- **Metabolomics**: specific metabolites were studied in order to determine their role in bacterium-induced disease resistance or bacterium-induced promotion of plant growth.
- **Glasshouse and field trial studies** (both *Fusarium* disease studies and plant growth studies); these studies provided material for the aforementioned studies and enabled the assessment of biocontrol/biofertiliser efficacy.

3. **Research Achievements**

- The results have shown diversity among the biocontrol strains studied. Based on morphological and phenotypic characteristics distinct growth variations were observed. The sequence information has confirmed that all of the biocontrol strains are fluorescent *Pseudomonads* and has also indicated a similar degree of diversity among the biocontrol isolates. The strain identity of these bacteria has been determined.
- The biocontrol strains were shown to have a broad-range of antifungal abilities against a range of fungi and yeasts. The strain MKB 156 was shown to be the only producer of the potent antifungal compound diacetyl phloroglucinol among the strains tested. Although
the bacteria produced low quantities of the compound, the antifungal capabilities of the strain were still extremely effective in vitro.

- This work elucidated new mechanisms whereby bacteria induce resistance to disease.
- This work has highlighted new functionality in plant biochemical pathways.
- We have shown that biocontrol involves a complexity of local and system responses.
- We have shown that select genes are responsive to particular plant hormones.
- This work highlighted the variability in the efficacy of biofertiliser bacteria, but also their potential for promoting plant growth under stress conditions.

4. **Impact of the Research**
This work identified bacteria of use as either biocontrol or biofertilisers and elucidated the mechanisms by which they work. New information and insights have been obtained into the traits that are necessary for effective plant growth promoting inoculants during their interaction with plants.

These results benefit the scientific community by enhancing our understanding of how plant and microbes communicate. The benefit industry in that they provide organisms that show promise as biocontrol and biofertilisers and also provide tools which can be used to screen for other biocontrol and biofertiliser agents.

5. **Exploitation of the Research**
One interesting outcome of the biocontrol research is that select bacterium influence the formation of glucans in cereal grain; this has an impact re the effect of grain on greenhouse gas emissions from cattle. We will apply for money in collaboration with animal scientists and animal feed companies, to investigate the potential of testing the ‘proof of concept’ that grain treated with these bacteria can reduce GHG emissions from cattle.

The potential of a bacterium to significantly promote grass growth is particularly exciting but the variability of results means its needs further validation; we will approach companies (grass breeding/seed production) in order to determine their interest in this work.

This grant formed the basis of an application to the BILL and MELINDA GATES FOUNDATION – a major finding from this research was that bacteria that live as endophytes are more likely to be successful as biofertilisers rather than rhizosphere dwellers. Future applied research will focus on these, in collaboration with Industry.

6. **Summary of Research Outputs**

(a) Intellectual Property applications/licences/patents
None to date; the IP potential of a biofertiliser will be decided based on the results of an ongoing trial conducted as part of a continuing study.

(b) Innovations adopted by industry
None as yet
(c) Number of companies in receipt of information
None as yet

(d) Outcomes with economic potential
Bacteria that can reduce chemical inputs into cereal production.
Bacteria that may potentially influence GHG emissions.

(e) Outcomes with national/policy/social/environmental potential
Bacteria that can reduce chemical inputs into cereal production.
Bacteria that may potentially influence GHG emissions.

(f) Peer-reviewed publications, International Journal/Book chapters.

(g) Scientific abstracts or articles including those presented at conferences


(h) National Report

(i) Popular non-scientific publications

(j) Workshops/seminars/ open days at which results were presented (excluding those in (g))
7. **Permanent Researchers**

<table>
<thead>
<tr>
<th>Institution Name</th>
<th>Number of Permanent staff contributing to project</th>
<th>Total Time contribution (months)</th>
<th>Average time contribution per permanent staff member</th>
</tr>
</thead>
<tbody>
<tr>
<td>UCD</td>
<td>3</td>
<td>15.72</td>
<td>5.24</td>
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<tr>
<td>UCC</td>
<td>3</td>
<td>14.52</td>
<td>4.84</td>
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<td><strong>Total</strong></td>
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<td><strong>30.24</strong></td>
<td><strong>10.08</strong></td>
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8. **Researchers Funded by RSF**

<table>
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<tr>
<th>Type of Researcher</th>
<th>Number</th>
<th>Total Time contribution (months)</th>
<th>Average time</th>
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<tr>
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<tr>
<td>Contract Researchers</td>
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<tr>
<td>PhD postgraduates</td>
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<td>17.04</td>
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<td>Masters postgraduates</td>
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<tr>
<td>Temporary researcher</td>
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<td>3.96</td>
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<td><strong>150.48</strong></td>
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9. **Postgraduate Research**

Total Number of PhD theses: ___1___

Smyth, Eoghan, UCD (submitted in August 2011 and passed his viva voce in November 2011). Selection and analysis of bacteria from Irish soils on the basis of their ability to promote plant development and growth.

Jennifer McCarty completed her project work and is currently engaged in conducting a PhD in the faculty of Medicine.

Total Number of Masters theses: ___0___
10. **Project Expenditure**

Total expenditure of the project: €767,886.44

Total Award by RSF €775,997

Other sources of funding (specify) €

1. 
2. 

**Breakdown of Total Expenditure**

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<th>Category</th>
<th>UCD Institution 1</th>
<th>UCC Institution 2</th>
<th>Total</th>
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<tr>
<td>Contract staff</td>
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<tr>
<td>Temporary staff</td>
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<td>Post doctorates</td>
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<td>Post graduates</td>
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<td>Consumables</td>
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<td>Travel and subsistence</td>
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<td>Durable equipment</td>
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<td>Other</td>
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<td>Overheads</td>
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<td><strong>369856.12</strong></td>
<td><strong>767886.44</strong></td>
</tr>
</tbody>
</table>

11. **Future Strategies**

Based on the results of this research there are two important outputs we wish to pursue

1. Genes that may enhance disease resistance. – Based on the results of this and other research, we have identified a range of genes that potentially enhance disease resistance. Through SFI funding (awarded July 2011) we will determine the IP potential of three such genes.

Additionally, we will seek FP funding to continue biofertiliser work.

12. **Industry Collaboration**

There have been no direct industry collaborations within this project. However, see above re future strategies.