Screening of potential endophytes for the biological control of *Crinipellis perniciosa*

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Endophytes

Endophytes (organisms which spend part or all of their life-cycle within the tissues of a host plant) have been shown to act as biocontrol agents within the *Graminaeceae* (Clay & Leuchtmann, 1989). They are considered to have potential to act as biocontrol agents of other plant species.

Endophytes were collected from the centre of diversity of *Theobroma* and related *Herrania* species (Amazon regions of Brazil and Ecuador). These endophytes may protect against pathogens of *T.cacao*, in particular witches' broom (*Crinipellis perniciosa*) and Frosty pod rot (*Moniliophthora roreri)*. This hypothesis is based on the premiss that when removed from its centre of origin *T.cacao* becomes depauperate in its natural endophytes, thus becoming more susceptible to disease.

A model system was developed in the UK to assess the ability of these endophytes to colonise *T.cacao* and reduce the incidence of witches' broom was assessed.

Screening Method

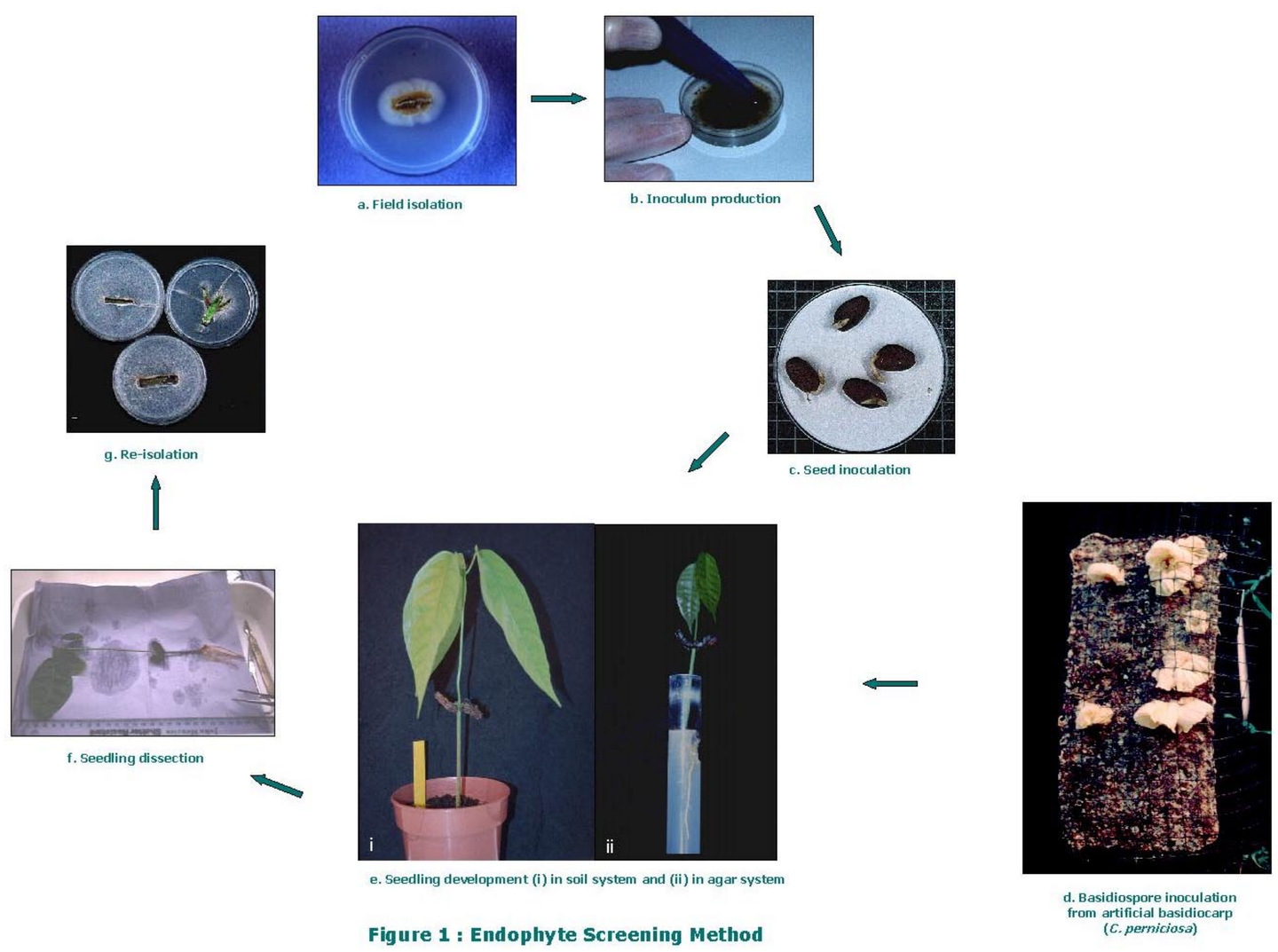

Figure 1 : Endophyte Screening Method

For Endophyte Screening:

1. endophytes are purified and identified
2. placed under near- UV to induce sporulation
3. inoculum prepared with 0.05% Tween
4. 10^6 spores/ml applied to the plumule of germinating seed
5. maintained in humid chamber for 3 days
6. seed planted in either sterile soil or agar
7. seedlings are dissected after 28 days and endophytes re-isolated.

For Biocontrol studies:

7. seedlings are inoculated with artificially produced basidiospores (10^6/ml) of *C.perniciosa* after 1 month.
8. assessment of disease incidence (presence of witches' brooms) is made over 30 days.
9. after 30 days the plants are dissected to re-isolate the endophytes.

Preliminary Results

- Initial assessments of the model system demonstrated that it was possible to inoculate and re-isolate endophytes from *T.cacao*. To date approximately 70% have been successfully re-isolated after seedling inoculation.
- The continued development of the system to assess the ability of the introduced endophytes to control witches' broom was pursued in Brazil.
- Preliminary results (Table 1) show that introduced endophytes can reduce disease incidence. For example isolate 10 reduced disease incidence by 75%.

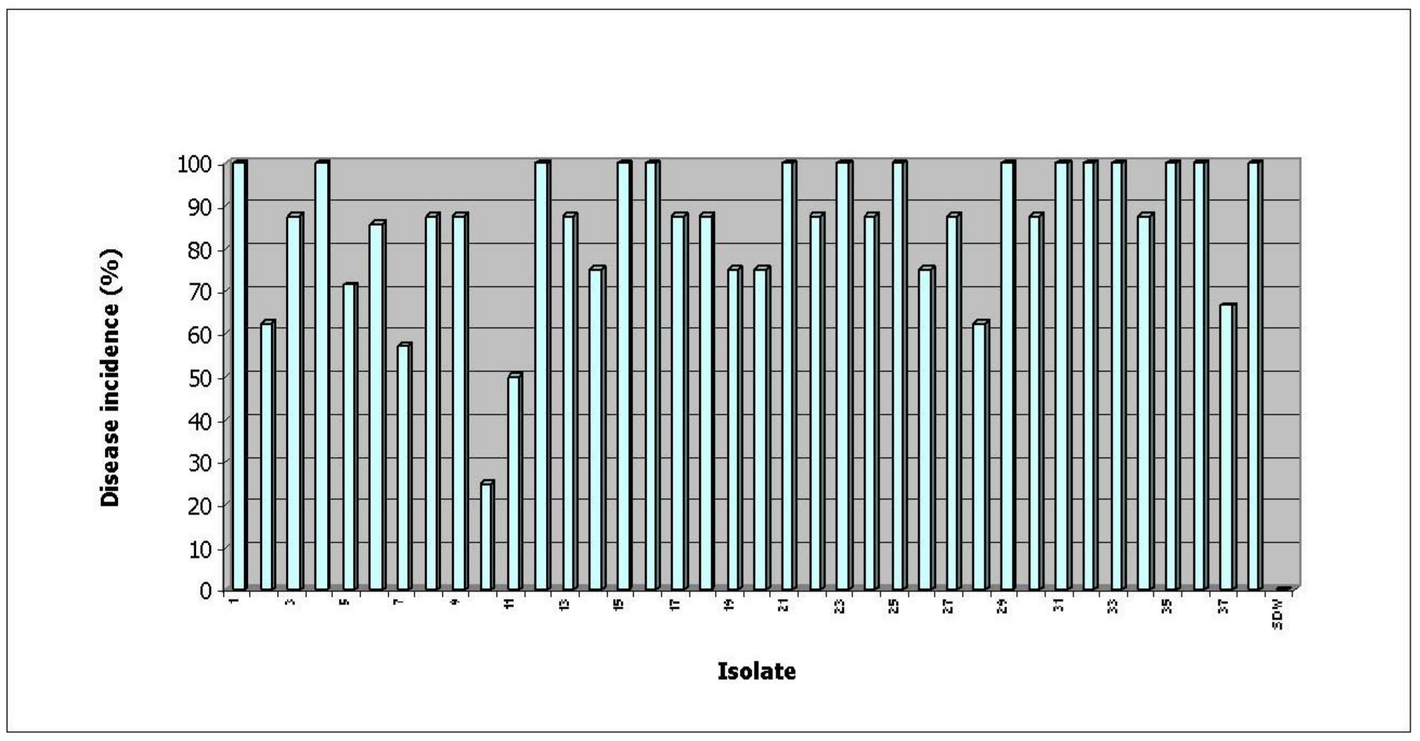

Table 1 : Witches' broom incidence in endophyte-inoculated *Theobroma cacao*

Conclusions and future prospects

Initial studies carried out in the UK and Brazil have shown that the endophytes collected in the Amazon regions of Ecuador and Brazil from *Theobroma* and *Herrania* species have the potential to control diseases of cocoa such as witches' broom (and hopefully *M. roreri*).

Assessment of the ability of endophytes to control witches' broom is continuing in glasshouse studies in the UK. Future trials are planned for Brazil and Ecuador to continue to assess the efficacy of the endophytes to control diseases of cocoa.

Reference

Clay, K. & Leuchtmann, A. (1989) Infection of woodland grasses by fungal endophytes. *Mycologia*, 81(5), pp 805-811.