



Genetic Analysis of the Interaction Between the Host Perennial Ryegrass and the Crown Rust Pathogen (*Puccinia Coronata* F.SP. *Lolii*)

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Presenter Information

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Introduction Crown rust (*Puccinia coronata* f.sp. *lolii*) is the most important fungal pathogen of perennial ryegrass (*L.perenne* L.). The physiological effects associated with infection include reduction of water soluble carbohydrate (WSC) reserves, causing decreased dry matter yield, digestibility and palatability for herbivores reared for meat, milk and wool production. Phenotypic variability of rust-infection in perennial ryegrass is likely to be due to environmental effects, as well as the interaction of defence and resistance genes in the grass and virulence genes in the pathogen. Classical and molecular genetic marker-based studies have previously detected both qualitative and quantitative resistance, due respectively to major genes and quantitative trait loci (QTL). In addition, evidence for physiological race variation has been demonstrated for *P. coronata* f.sp. *avenae*, the causative organisms of crown rust in oat, and has been inferred for *P. coronata* f.sp. *lolii*. Evaluation of genotypic variation in both the host and pathogen is consequently important for the analysis of the interaction.

Materials and methods Candidate gene-based genetic markers related to the plant defence response (DR genes) were added to the genetic map of the F₁(NA₆xAU₆) mapping family as restriction fragment length polymorphism (RFLPs), and locus-specific single nucleotide polymorphism (SNP) markers were developed from both DR and resistance gene analogue (RGA) genes. The co-location of functionally-associated markers and QTLs for disease resistance was evaluated. Assessment of genetic variation and race structure within the pathogen is based on the use of gene-associated simple sequence repeat (SSR) markers and sampling at the single pustule level. A set of 55 unique SSR markers were developed from crown rust expressed sequence tags (ESTs) and characterised in terms of motif structure, sequence annotation, efficiency and specificity of amplification and polymorphism detection. A method based on multiple displacement amplification (MDA) was used to obtain templates from single pustules, in association with a detached leaf assay method.

Results and Conclusions In a preliminary analysis, genetic variability was detected within and between isolates from geographical locations in the UK, Japan, Australia and New Zealand (Fig 1). The sources of variation may include spore migration, mutation or sexual recombination within the pathogen. Further studies of Australian populations will provide evidence to compare between these hypotheses.

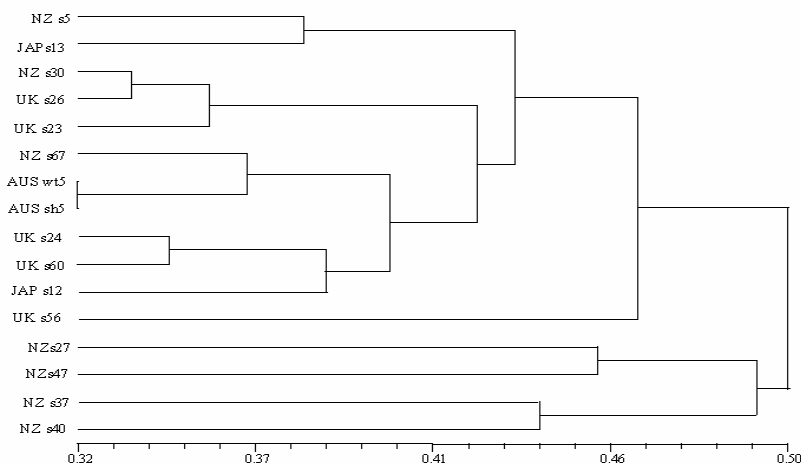


Figure 1 UPGMA dendrogram based on the analysis of variation of 12 EST-SSR markers on selected single pustule samples from UK, Japan, Australia and New Zealand