FINAL PUBLISHABLE SUMMARY REPORT

The rice blast fungus *Magnaporthe oryzae* is one of the most damaging disease of cultivated rice worldwide and an emerging disease on wheat, impacting on global food security. In order to develop durable and environmentally friendly control methods, it is important to expand our knowledge on the molecular mechanisms underpinning *M. oryzae*-rice interaction.

RNA-binding proteins play a fundamental role in the control of gene expression at post-transcriptional level and are responsible for regulating essential biological activities. Here, we initiated studies in the post-transcriptional mechanisms that control *M. oryzae* infection-related processes. To this end, we characterised a RNA-binding protein required for full disease symptom production in the rice blast fungus. We found an insertional mutant M35 that showed reduced lesions on leaves and roots. The T-DNA was located in a gene encoding an RRM protein with six RGG tripeptides (RBP35). The RRM domain is widely spread in eukaryotes although only a small fraction has been studied. In humans, it is estimated that about 2% of the total proteome contain at least one RRM (497 gene products out of ~25 000 genes in the human genome). The *M. oryzae* genome encodes 76 RRM proteins and RBP35 represents the first *M. oryzae* RRM protein investigated to date. The combination of RRM and RGG modules is found in well characterised RNA-binding proteins with highly diverse functions in human and yeast. Orthologues of RBP35 are found only in filamentous fungi.

Using a combination of cell biology, biochemistry and transcriptomics, we investigated the involvement of RBP35 in *M. oryzae* full disease symptom production. Notably, we demonstrate that RBP35 interacts *in vivo* with a highly conserved protein component of the eukaryotic polyadenylation machinery. We show that RBP35 present different diffusional properties in nuclei of distinct fungal structures, and consequently different protein/nucleic acid interactions. Further, we find that RBP35 regulates the length of 3'UTRs of transcripts with developmental and virulence-associated functions. We prove that the Δ*rbp35* mutant is affected in the TOR (target of rapamycin) signaling pathway showing significant changes in nitrogen metabolism and protein secretion. We conclude that RBP35 is a novel component of the polyadenylation machinery of *M. oryzae* required for alternative 3'end processing of transcripts associated with signaling and metabolism. Results indicate that RBP35 acts as a gene-specific polyadenylation factor, ultimately regulating developmental and infection-related processes in the rice blast fungus. Nothing it is known about pre-mRNA 3' end processing in filamentous fungi and our study suggest that their polyadenylation machinery differs from yeast and higher organisms. This study can provide new insights into the evolution of the pre-mRNA maturation and the regulation of gene expression in eukaryotes (PLoS Pathogens 2011, accepted).

Socio-economic impacts of the project. It is estimated the existence of more than one million fungal species on the Earth. Despite their diversity in live styles (saprophytic, parasitic and mutualistic), fungi share common features distinctive from plants and animals and have been grouped taxonomically as an independent eukaryotic kingdom. Filamentous fungi are of great importance for industry, medicine and agriculture. They also represent invaluable tools for understanding fundamental biological processes since they are highly tractable organisms for experimental research and, unlike yeast, can be used to study important features of complex eukaryotes such as cellular differentiation, multicellular development, pathogenesis, natural product synthesis, DNA methylation, gene silencing, chromatin remodelling and programmed cell death.

Food security has become a serious global issue in the world. The number of people suffering from chronic hunger has increased slowly but steadily during the last years. It is expected that food production will have to increase ~40% in order to fulfil nutritional needs worldwide by 2050. It is urgent to increase investment in Agriculture in order to solve food security issues for the following decades. Undoubtedly, a better understanding of *M. oryzae* plant colonisation will have positive implications for the food security and economic stability of rice- and wheat-dependent populations worldwide and have important implications for the development of new strategies for plant breeding and disease control.

We have characterised a novel protein component of the *M. oryzae* polyadenylation machinery that is absent in yeasts, plants or metazoans required for alternative pre-mRNA 3' end processing. Presence of multiple potential 3' end cleavage sites is common in eukaryotic genes, and the selection of the right site is regulated during development and in response to cellular cues. The mechanism of ***alternative*** (or non canonical) polyadenylation regulates the presence of cis elements in the mRNA and generation of mRNA isoforms with different exon content or 3'UTR lengths. The cis elements present in the 3'UTRs such as microRNA target sites modulate gene expression by affecting cytoplasmic polyadenylation, subcellular localization, stability, translation and/or decay of the mRNA. Therefore, the selection of a proper 3´end cleavage site represents an important step of regulation of gene expression and can be regulated by multiple mechanisms. Our study demonstrated unique post-transcriptional mechanisms present in filamentous fungi. Consequently, our results may have impacts on medicine (parallelism between fungal pathogens of plants and animals exits), agriculture (identification of novel targets to control fungal diseases) and industry (improved expression of recombinant proteins).