

# Investigation of F-box protein-encoding genes in *Puccinia triticina* genome and their roles in pathogenicity during leaf-rust infection in wheat Anupama Gidhi, Manish Kumar, Kunal Mukhopadhyay

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Introduction: Wheat is an important cereal crop and staple food worldwide. It contains high nutrient value, therefore wheat consumption is expected to increase by 10% by 2050 in developing countries and prerequisite to increasing its production. However, *Puccinia triticina*, a biotrophic basidiomycetes fungal pathogen, causes leaf rust disease in wheat, which has critically threatened wheat productivity and reduced the yield by approximately 10% annually (Dean et al., 2012). F-box proteins are components of the SCF complex and have an essential role in substrate recognition and degradation accompanied by 26S proteasome (Bai et al., 1996). Very little studies are available for fungal F-box proteins belonging to basidiomycetes in contrast to ascomycetes fungi and plants.

## Aims:

- To identify F-box encoding genes in *Puccinia triticina* genome.
- Characterization of identified genes using bioinformatics approaches.
- Expression analysis of identified genes during the leaf-rust infection in wheat.



## **Methodology:**

# Identification

**Puccinia** genome sequences downloaded from Ensembl Fungi

**Translation of genome sequences using Transeq Emboss** 

Pfam analysis of translated sequences

**Gene prediction using FGENESH** 



Gene Ontology Gene structure analysis Phylogenetic analysis (WEGO) (GSDS; http://gsds.gao-lab.org/) (MEGA7)

## Fig. 2: Different domains of F-box proteins



# Fig. 3: Distribution of Intron and Exon in **F-box** genes



Fig. 4: Phylogenetic analysis of F-box proteins identified in *Puccinia triticina* with other plant pathogenic fungal species (A) Fusarium oxysporum, Magnaporthe oryzae, Puccinia graminis, Botrytis cinerea (B) Auxin signaling F-box (TaAFB) proteins of

#### **Expression** analysis

**QRT-pRCR** based expression of genes at different time points (0, 24, 48, 72, 120, 168 hpi) during leaf rust infection

## **Results:**



#### Triticum aestivum.



Fig. 5: Synteny analysis among the *F-box* genes of *Puccinia triticina* with *Puccinia* striiformis and Puccinia graminis.





Fig. 6: Gene Ontology (GO) analysis of *F*box genes using WEGO.

> Fig. 7: Expression of F-box genes during pathogen progression in susceptible isolines (HD2329)

Fig. 1: Pipeline for prediction of F-box protein encoding genes in *Puccinia triticina* 



## **Conclusions:**

In *Puccinia triticina*, 10 *F-box* genes were predicted having F-box motifs along with leucine-rich repeat (LRR) and WD40 at C-terminus. These genes were interrupted by introns. GG705655 showed proximity with MoMet30, the pathogenic F-box gene in Magnaporthe oryzae. GG705409 clustered with TaAFB4/TaAFB5 suggesting the possibilities of horizontal gene transfer during the coevolution. The *F-box* genes also showed orthologous relationship with Puccinia graminis, and Puccinia striiformis and annotated to be components of SCF complex that participate in protein ubiquitination degradation process. The expression analysis revealed positive response of GG705409 in pathogenicity at early stages of infection while GG711975 and GG705482 at later stages of infection. This study provides information on structure function of F-box proteins of a plant pathogenic fungi and their role during pathogenesis.

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