led by Revistas UPO (Universidad Pablo de Olivade

nº9 (March, 2020)

Poster

Study of PMT target specificity in Ustilago maydis

Biosaia (revista de los másteres de Biotecnología Sanitaria y Biotecnología Ambiental, Industrial y Alimentaria de la UPO)



Rodríguez Rivas, Álvaro (1); Ramos Barrales, Ramón (1), Pejenaute Ochoa, María Dolores (1)

(1)Área de Genética, CABD (Sevilla)

Tutor académico: Ramos Barrales, Ramón

Keywords: PMT; Ustilago maydis; infection; adhesion; mannosylation; protein domain

ABSTRACT

Ustilago maydis is a pathogenic fungi responsible for the corn smut fungus disease, which causes a significant loss in mayze production every year. The PMT is a family of well-conserved O-mannosylating proteins. In Ustilago maydis, the deletion of Pmt2 has been shown to be deleterious and the deletion of Pmt4 disrupts completely the infectious process; on the other hand, the deletion of Pmt1 doesn't manifest any phenotype. All PMT proteins have three domains, named PMT, MIR and 4TMC, and several transmembrane regions. We hipothesized that one of these domains of Pmt4 is responsable for the substrate specificity that confers the virulence phenotype in Ustilago maydis Pmt4 and, if this would be the case, we could develop an antifungal drug specific to this domain. To check this, we built three chimerical protein strains, chanching one domain in Pmt4 for the same domain of Pmt1 at a time, and measure the tumor formation in 3 independent experiments. We also built a full Pmt4 length strain as a positive control. The results indicate that either the protein requires all three Pmt4 domains to infect or the chimerical protein is not working properly.

Furthermore, we also wanted to check what region gives Pmt2 its specificity for growth. In Schizosaccharomyces pombe, a ortholog of Pmt2, named Ogm2, present the same essencial-for-growth phenotype than in U. maydis. Since U. maydis doesn't have any repressable promoter, we built a S. pombe strain with the shut-off promoter nmt81 in the endougenous Ogm2 gene and will complement the phenotype with mutagenized plasmids containing the U. maydis Pmt2 and measure its viability. Finally, we also plan to build other three strains interchanching Pmt4 domains in the Pmt1 protein.

REFERENCES

Fernández-Álvarez, A., Marín-Menguiano, M., Lanver, D., Jiménez-Martín, A., Elías-Villalobos, A., Pérez-Pulido, A.J., Kahmann, R., Ibeas, J.I., 2012. Identification of O-mannosylated virulence factors in Ustilago maydis. PLoS Pathog. 8. https://doi.org/10.1371/journal.ppat.1002563

Fernández-Álvarez, A., Elias-Villalobos, A., Ibeas, J.I., 2009. The O-mannosyltransferase PMT4 is essential for normal appressorium formation and penetration in Ustilago maydis. Plant Cell 21, 3397–3412. https://doi.org/10.1105/tpc.109.065839

Blanquero, A., Ramon, R., Biológico, C., Universidad, D., Olavide, P. De, 2019. Development of an antifungal drug discovery system against the plant pathogen Ustilago maydis 21, 2019.

Maundrell, K., 1990. nmtl of Fission Yeast. J. Biol. Chem. 265, 10857-10864.

Tanaka, N., Fujita, Y., Suzuki, S., Morishita, M., Giga-Hama, Y., Shimoda, C., Takegawa, K., 2005. Characterization of O-mannosyltransferase family in Schizosaccharomyces pombe. Biochem. Biophys. Res. Commun.