*Talk*


# FUNCTIONAL METAGENOMIC FOR THE IDENTIFICATION OF NEW LIGNOCELLULOSE-DEGRADING ENZYMES

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## ABSTRACT

**Motivation:** Due to the necessity of mitigation of climate change we are facing nowadays, new economic and industrial approaches are needed to be developed. Enzymatic biotechnology has demonstrated to be an efficient tool to reach a more sustainable economic system, allowing to discover enzymatic activities with industrial interest. Nevertheless, there are some limitations on the already discovered enzymes when applied industrially due to the extreme conditions they have to bear, such as high temperatures or pH. Functional metagenomic permits accessing to the genomic information of the vast microbial diversity of a specific ecosystem without the need of prior cultivation of microorganisms and to reveal new and more efficient enzymatic activities without knowing its gene sequence (1). In this project, we focus on the identification of new enzymes for developing a biocatalyst for lignocellulosic revalorization and plastic degradation. For this purpose, we are using specialised vectors and bacterial strains and several metagenomic libraries previously constructed (2, 3). Moreover, we are constructing a new metagenomic library from a compost pile of agro-industrial waste, that must be enriched in lignocellulose and plastic degrading microorganisms.

**Methods:** The methodology applied screening of enzymatic activities consists of conjugating the metagenomic library to E. coli MPO554Nal or MPO554Nal/pMPO1077 strains. Plasmid pMPO1077 contains an inducible autolysis system to facilitate phenotype detection of extracellular enzymes (2). For exoglycanase activity screening, the metagenomic library was transferred to E. coli MPO554Nal and positive clones were selected in M9 agar plates with Avicel as carbon source. For screening of endoglucanase activity, the metagenomic library was transferred to E. coli MPO554Nal/pMPO1077, transconjugants were plated on LB agar plates with 0.7% CMC, and after colonies grew, lysis was induced with anhydrotetracycline (2). After confirmation of positive clones phenotypes, we analyse enzymatic restriction patterns in order to select only different clones for posterior sequencing analysis.

**Results:** To date we have 248 candidates for endoglucanase and 223 for exoglucanase activity from screening of a metagenomic library obtained from the soil of Los Alcornocales natural park (Cádiz). Identification of the genes responsible for each activity will be done by the metagenomic plasmid sequencing and comparation with previuos reported data.

## REFERENCES

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