Jena School for Microbial Communication
Graduate School of Excellence

PhD project title: Filamentous microbial co-culture for biotechnological production of natural products

PhD Student: Ana Palacio-Barrera

Supervisors: Prof. Dr. Miriam Agler-Rosenbaum
Prof. Dr. Erika Kothe
Dr. Ivan Schlembach

Abstract:

So far, consolidated-bioprocessing has been studied mainly for its application in the production of bulk chemicals and biofuels; however the exploitation of such an idea for the production of secondary metabolites has been barely explored. The main aim of the research project is to develop a system that makes use of a synthetic microbial consortia for the development of a consolidated bioprocess in the production of secondary metabolites starting from cellulose, establishing a co-culture between model cellulolytic fungi as *Trichoderma reesei* or *Penicillium verruculosum* and the model filamentous bacteria *Streptomyces coelicolor* producing secondary metabolites such as actinorhodin and undecylprodigiosin.

In nature, it can be observed how microorganisms compete, antagonize or collaborate with each other, in different soil, aquatic, plant and animal niches; and as outcome of such interactions several things can occur, such as degradation of organic matter, carbon cycling, decrease/increase of a population from a specific microorganism, nutrient exchange between several partners in a community or transfer of nutrients to other higher organisms as plants.

Several types of interactions can occur in the inter-kingdom crosstalk in nature; such as predation and parasitism, ex. the soft rot disease caused by the bacteria *Janthinobacterium agaricidamnosum* to *Agaricus bisporus*; a symbiotic interaction with a final phytopathogenic purpose like the one observed between the bacterial strains *Burkholderia rhizoxina* sp. nov. or *Burkholderia endofungorum* sp. nov. and *Rhizopus* spp. both causing rice seedling blight; and symbiotic interactions with a final plant growth promoting purpose like, for instance, *Streptomyces* AcH05 with *Amanita muscaria* or *P. fluorescens* strain BBc6R8 with *Laccaria bicolor* S238N, both fungi involved in mycorrhiza formation.

In the present project, however, studies will be focused in establishment of synthetic microbial co-cultures for a biotechnological and in future further industrial application, making use as well of concepts and phenomena of the interactions that could actually take place in nature. The proposed system and key players involved are advantageous since both carbon limitation due to slow glucose release and microbial interaction can take place.

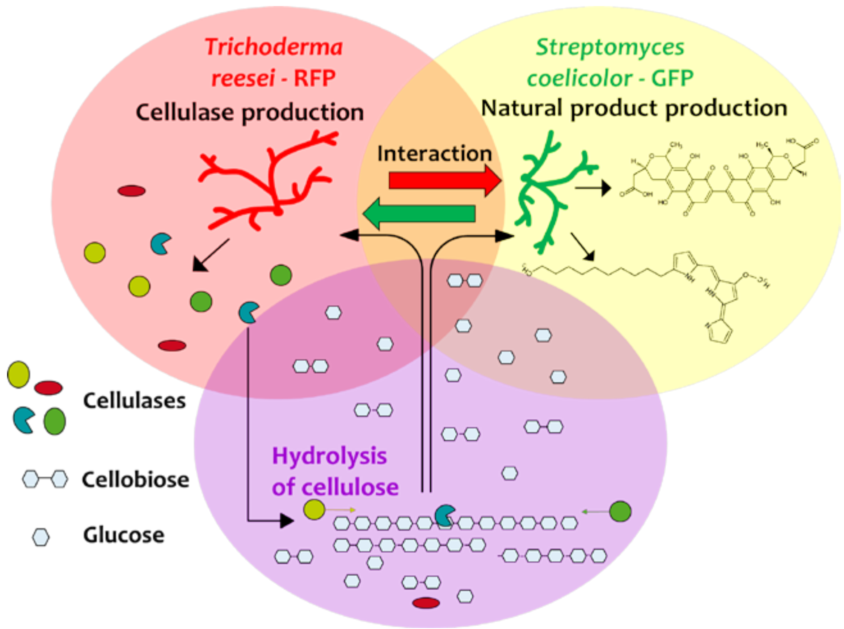

Fig 1. Proposed scheme of process in the microenvironment

These kind of features are among several that can resemble to those found in a natural environment, for example natural soil habitat supports interplay between microorganisms, as readily available carbon sources such as simple sugars are scarce and most of the available carbon is present in form of recalcitrant lignocellulosic plant material. Thereby, cellulolytic microorganisms play an important role in making this lignocellulosic material available to other microorganisms in the community.

As an alternative, if culture based strategies fail to establish stable co-culture conditions, the co-culture will be artificially stabilized by means of genetic engineering. The strategy would be then to establish an artificial metabolic co-dependency, by deleting complementary essential amino acids or essential vitamin production pathways, while overexpressing the corresponding counterpart in the respective organisms. Thereby, both organisms will supply essential nutrients to their partner while receiving another nutrient in exchange. Since both organisms are well studied, the respective amino acid synthesis genes can be simply knocked out using standard genetic tools such as split marker based homologous recombination.