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PII: S0169-7439(17)30683-4

DOI: 10.1016/j.chemolab.2018.03.011

Reference: CHEMOM 3605

To appear in: Chemometrics and Intelligent Laboratory Systems

Received Date: 24 October 2017

Revised Date: 23 January 2018

Accepted Date: 20 March 2018

Please cite this article as: Q. Carboué, M. Claeys-Bruno, I. Bombarda, M. Sergent, Jéô. Jolain, S. Roussos, Experimental design and solid state fermentation: A holistic approach to improve cultural medium for the production of fungal secondary metabolites, *Chemometrics and Intelligent Laboratory Systems* (2018), doi: 10.1016/j.chemolab.2018.03.011.

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#### ACCEPTED MANUSCRIPT

# Experimental design and solid state fermentation: a holistic approach to improve cultural medium for the production of fungal secondary metabolites

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#### Abstract

Solid-state fermentation (SSF) is a cultural method that holds tremendous potentials for the production of numerous microbial value-added compounds in various industries. As for every other process, experimental designs can provide tools to improve the product yields, diminish the production time and thus eventually decreasing the cost of the whole process. However, SSF, because of its solid nature, implies some constraints which consequently require specific tools to efficiently overcome them. The aim of this study was the improvement of the production of antioxidant naphtho-gamma-pyrones produced by *Aspergillus niger* G131 cultivated using SSF. Two experimental designs were presented, a combined design, taking into account two different types of variables to determine a proper solid medium, and a screening design with mixed-level factors to find solutes with significant positive effects on the output.

Keywords: Solid-state fermentation; experimental design; Aspergillus niger; naphtho-gamma-pyrones

#### 1. Introduction

Solid-state fermentation (SSF) is defined as the fermentation involving solids in absence (or near absence) of free water; however, substrate must possess enough moisture to support growth and metabolism of microorganisms [1]. The SSF system offers several economical and practical advantages including high product concentration, improvement of product recovery, simple cultivation equipment, lower plant operational, potential direct utilization of the crude fermented products and also the possibility to use agro-industrial byproducts instead of synthetic ingredients as substrate, thereby to cost cutting the bulk of the production [2,3]. These solid byproducts could be either a source of carbon and other nutrients or an inert support material for absorption of nutrients and biomass anchorage. In this case, supplementation is needed in order to provide all necessary compounds for an optimum growth. Macro and micronutrients that are usually added to the medium include phosphorus, sulfur, potassium, magnesium, calcium, zinc, manganese, copper, iron, cobalt, and iodine [4,5]. Each microorganism has its own optimal cultivation conditions and requires specific substrates depending on the expected products. Optimization of the medium and cultural conditions is hence essential step to maximize the production, especially if the process heads towards industrial

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