



## DIMINUTION OF TOXINS AND SIMULTANEOUS LIPASE PRODUCTION IN CASTOR BEAN CAKE BY SOLID STATE FERMENTATION

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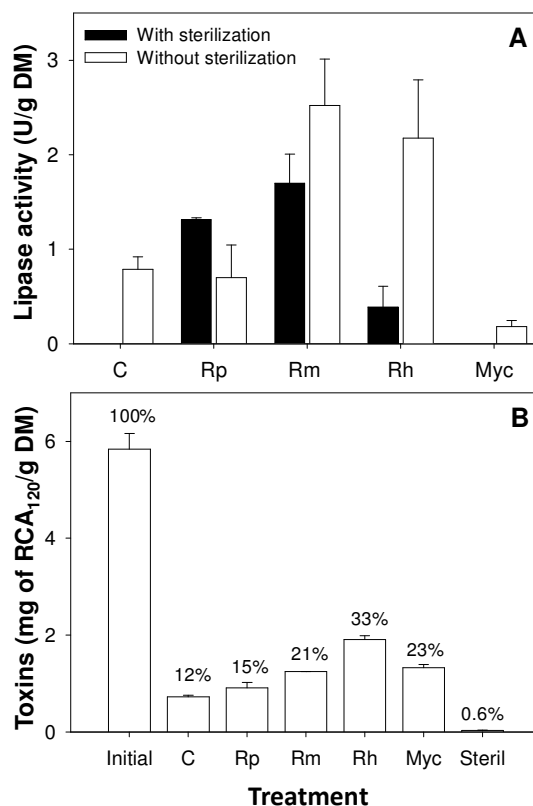
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**Introduction.** The castor bean consists of approximately 50% oil is a promising candidate for biodiesel and bio lubricant production. Along this process, there is a by-product called castor cake that is currently used in fertilizers (1). Although these castor cakes are rich in protein and fiber, the presence of toxic compounds limits their use as an animal feed. Biological detoxification, using solid-state fermentation (SSF) with filamentous fungi, has been used to detoxify other residues, showing good results (2). In addition to promoting residue-detoxification, SSF of castor bean waste represents an interesting and low-cost alternative for generating useful enzymes (3) and its residual oil can be used to produce lipases (4) that have found a great number of biotechnological they are able to catalyze and being considered as the most versatile biocatalyst (5).

The objective of this work was the detoxification of castor bean cake and the lipase production by using four filamentous fungi under SSF conditions.

**Methods.** *Rhizomucor pusillus* (Rp) and *Myceliophthora* sp. (Myc) were a generous gift from Dr. S. Roussos (IRD, France); *Rhizomucor miehei* (Rm) and *Rhizopus* sp. (Rh) are part of CIATEJ collection.  $1 \times 10^7$  spores per gram of dry solid of each filamentous fungi was added to individual vials with 1 g of castor bean seed cake and moisturized to 40% (w/w). The temperature of incubation was of 40°C for Rm and Rh and 37°C for Rp, Myc and the control without inoculums (C). Fermentation period was 4 days and measure of toxins and lipase activity was made at end of SSF. Toxins were measured by ELISA using as primary rabbit anti-RCA<sub>120</sub>, as secondary goat antirabbit IgG coupled to Horseradish peroxidase and revealed by 20 min incubation with TMB substrate, stopped with 2M H<sub>2</sub>SO<sub>4</sub> and the absorbance was measured at 450 nm in a microtiter plate-scanning spectrophotometer. Lipase activity was extracted from solid with a solution containing N-lauroylsarcosine at 0.5% (w/v). Lipase activity was measured with p-nitrophenyl palmitate (p-NPP) as substrate and the reaction was performed at pH 7.5 and 37°C. The hydrolysis rate of substrate was monitored at 410 nm. One unit (U) was defined as one μmol of p-nitrophenol produced by min in the assay conditions.

**Results.** The four fungi strains were able to growth and produce lipase activity in sterilized or not castor bean cake, being Rm and Rh the major producers in non sterile conditions followed by Rp and the endogenous microorganism growth in C (Fig 1A). A significant decrease of toxins was obtained at the end of SSF for the C, Rp, Rm and Myc, being the sterilization the best way to detoxify the castor bean cake (Fig. 1B).



**Fig.1 A)** Lipase activity quantification with p-NPP of SSF with filamentous fungi using castor bean seed cake with or without initial sterilization. **B)** Toxin quantification by ELISA in mg equivalents of RCA<sub>120</sub> for SSF with filamentous fungi using castor bean seed cake without initial sterilization. Steril: sterilized castor bean seed cake.

**Conclusions.** Castor bean seed cake constitutes an alternative source for lipase production with filamentous fungi (in particular for *Rhizomucor miehei* and *Rhizopus* sp.) by solid state fermentation in which the level of toxins was decreased significantly, making them an interesting alternative to obtain value-added products from the castor bean by-product.

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