Pre-selection of microbial isolates for pollutants degradation in coffee processing wastewater

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The interest for quality improving coffee production has led to an increased use of processing forms with increased water use (Bruno and Oliveira, 2008) and consequent increase of wastewater with high organic load (Dias et al., 2014) that requires proper treatment prior to their disposal or re-use (Matos, 2006). The pollutants degradation can be achieved by microorganisms' action (Sant'anna, 2013). However, biological treatment effectiveness is achieved only from the knowledge of existing microorganisms in the waste, which once adapted, can be used after the bioaugmentation to the treatment.

In this sense, the objective of this study was to isolate and characterize the microbiota present in wastewater from coffee beans processing (ARC) in order to investigate the presence of microorganisms capable of promoting the reduction of pollution load of livestock manure since literature lacks information about this residue.

Methodology: Microorganisms were isolated from ARC samples collected from a sewage treatment plant in a coffee producing unit. Yeasts and bacteria were characterized in morphotypes. The biochemical characteristics, were used for characterization and screening of microorganisms. Different sources of carbon and nitrogen were evaluated, and growth in the presence of pectin and caffeine was considered a crucial feature for strains pre-selection.

The pre-selected microorganisms were inoculated (10⁷ cells/mL) in 10 mL of ARC with three different pH: 4.2 (pH of the residue), 6 and 7, adjusted with CaO. The cell growth was assessed after 6 days from plating to bacteria, and direct counting in a Neubauer chamber, to yeasts. Microorganisms with final population greater than or equal to 10⁷ cells/ml were selected for analysis of pollutants degradation in the ARC and identified by ITS and 16S rRNA sequencing regions.

Results and discussion: Were purified 116 yeast and 127 bacteria strains and analyzed their biochemical features. Between the microorganisms groups, the population and morphotypes number was higher for bacteria (16 morphotypes), which was expected since this group is the most abundant and active in biological treatment systems (Sant'anna, 2013).

Nineteen bacteria and twelve yeasts grown in the caffeine and pectin presence as the single source of carbon and nitrogen. Of these isolates, only five bacteria strains grew at pH 4.2 ARC, therefore, just the count data in pHs 6 and 7 were considered in the analysis of growth. Seven isolates were selected with the greatest growth, whose populations ranged from 3.7×10^8 cell/mL (isolated called UFLA ARC 6) to 2.3×10^6 cell/mL (ARC UFLA 94). This last isolate showed smaller population, however, it produces larger cell sizes, which can result in increased production of microbial biomass, and possibly greater degradation of pollutants.

The yeasts grow in the three pH values. Eight strains showed higher growth, ranging from 8.4x 10⁷ cell/mL (UFLA ARC 231) to 2.1 x 10⁷ cell/mL (UFLA ARC 213). After sequencing, it was found that microorganisms selected among the three species of yeast (*Meyerozyma caribbica, Pichia anomala* and *Trichosporon montevideense*) and three species of bacteria (*Serratia marcescens, Corynebacterium flavescens* and *Bacillus* sp). Due to the greatest growth at pH 6 and 7, these values were used in the following tests to check for influence of pH values in the clearance of ARC.

Conclusion: microorganisms are able to use complex substrates such as pectin and caffeine, as well as developing the ARC and must, therefore, be evaluated for application in biological treatment of wastewater, aimed at organic matter degradation and consequent reduction of biochemical oxygen demand and chemical oxygen demand known as two pollution indicators.

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