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IMMOBILIZATION OF A LIPASE ACTIVITY FROM *Aspergillus niger* MYA 135 AND ITS APPLICATION IN THE BIODIESEL SYNTHESIS

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Lipases have been widely used in the organic synthesis of industrially important chemicals such as emulsifiers, surfactants, wax esters, biopolymers, structured lipids, flavor-associated esters, and biodiesel. Concerning the biodiesel production, in order to get an efficient biodiesel production, the proper selection of the immobilization matrix and the subsequent reaction optimization have attracted the interest of several research in recent years. In this work, those steps were carried out by using the one factor at a time optimization method. Thus, a culture supernatant from *Aspergillus niger* MYA 135 showing a lipase activity was firstly immobilized by adsorption on different low-cost supports (sand, PET and PP plastic, rubber, silicone, glass beads, silica gel and bagasse) applying a vacuum drying procedure. All biocatalysts were evaluated at 40°C, at 800 rpm, and in the presence of different combinations of oil (soybean or waste frying oils) and alcohols (ethanol or butanol). After a three-stepwise addition of the corresponding alcohol, the biodiesel synthesis was evaluated by thin layer chromatography (TLC). The most promising reaction mixture comprised a lipase activity immobilized in silica gel as biocatalyst, and soybean oil and butanol as substrates. Then, the following parameters were analyzed: a) the enzyme concentration (1, 2, 3, and 4 mL of culture supernatant), b) the molar ratio oil:alcohol (1:3, 1:4, 1:5, 1:6, 1:7), and c) the reaction time (the addition of alcohol carried out in three equal parts every 24, 12, 6 or 3 h). In addition, the crosslinking immobilization technique was also studied. Taking into account the qualitative analysis by TLC, the best conditions for biodiesel production were: 2 mL of culture supernatant immobilized in silica, 1:4 soybean oil to butanol molar ratio, and a reaction time of 18 h. Under these optimal reaction conditions, a biodiesel yield of 93.36% (w/w) was achieved in a solvent free system. The composition of fatty acid butyl esters was 12.97% palmitic acid, 6.57% estearic acid, 25.15% oleic acid, 45.24% linoleic acid, 4.72% linolenic acid, 0.67% araquidic acid, 0.34% eicosenoic acid, and 3.83% others. Finally, it is interesting to mention that the cloud point of butyl esters is around 10°C lower than that of methyl esters, meaning that they have better performance under cold conditions.

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IMMUNOMODULATORY PROPERTIES OF A GABA-ENRICHED STRAWBERRY JUICE PRODUCED BY *Levilactobacillus brevis* CRL2013

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Gamma-aminobutyric acid (GABA) plays a key role in mammals as the major inhibitory neurotransmitter of the central nervous system. Although GABA may not be able to cross the human blood-brain barrier, it was approved as a food ingredient because of its benefits to the host after oral administration including anti-hypertensive, anti-depressant, and anti-inflammatory activities. Considering the current trend towards the development of new functional and natural products and that microbial fermentation is one of the most promising methods to produce this non-protein amino acid, the *in-situ* production of GABA through fermentation of strawberry and blueberry juices by the efficient GABA producer strain, *Levilactobacillus* (L.) *brevis* CRL 2013, was evaluated. A high GABA production (262 mM GABA) was obtained after fermenting strawberry juice supplemented with yeast extract for 168 h. *In vitro* functional analysis of the GABA-enriched fermented strawberry juice (FSJ) demonstrated its ability to significantly decrease the expression of *cox-2* gene in LPS stimulated RAW 264.7 macrophages. In addition, *in vivo* studies in mice demonstrated that both, *L. brevis* CRL 2013 and the GABA-enriched FSJ were capable of reducing the levels of peritoneal, intestinal and serum TNF-α, IL-6, and CXCL1, and increasing IL-10 and IFN-γ in mice exposed to an intraperitoneal challenge of LPS. Of note, the GABA-enriched FSJ was more efficient than the CRL 2013 strain to reduce the pro-inflammatory factors and enhance IL-10 production. These results indicated that the CRL 2013 strain exerts anti-inflammatory effects in the context of Toll-like receptor (TLR)-4 activation and that this effect is potentiated by fermentation. Our results support the potential use of *L. brevis* CRL 2013 as an immunomodulatory starter culture and strawberry juice as a remarkable vegetable matrix for the manufacture of GABA-enriched fermented functional foods capable of differentially modulating the inflammatory response triggered by TLR4 activation.

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OBTENTION OF RECOMBINANT BACTERIOCINS WITH ANTIMICROBIAL ACTIVITY AGAINST *S. aureus* ISOLATED FROM BOVINE MASTITIS

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Bovine mastitis is the disease of dairy cattle that causes the greatest economic losses to dairy farmers and industry over the world. *Staphylococcus aureus* is the most frequently isolated pathogen from bovine intramammary infections in Argentina and worldwide. Lack of effectiveness of traditional control measures based on milking hygiene and antibiotic therapy against this organism has led to the development of alternatives to complement classical measures. Among them, the use of natural