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Study of milk-clotting proteases produced by local fungal strains: Use in cheese-making

Souhila Bensmail and Fazouane Fethia
M'hamed Bougara University, Algeria

The coagulant traditionally used for cheese-making in most of the world is rennet extract from the fourth stomach of suckling calves. There have been temporary shortages of rennet in various countries, but a world chronic shortage occurred after. This has stimulated interest in the search for suitable substitutes for use as coagulants in cheese-making. Our work was carried out to study the production of extracellular milk-clotting enzymes by local fungal strains *Aspergillus niger* FFB1, *Rhizopus stolonifer* and *Mucor* sp. under SSF conditions using wheat bran as substrate, followed by the application of crud extracts in the manufacture of two kinds of cheese, compared to the commercial preparation of rennet used at "Laiterie et Fromagerie Boudouaou" (LFB). After fermentation using wheat bran (10g) at 30°C for 72h, the acid proteinases of *A. niger* FFB1 and *R. stolonifer* extracted with sterile distilled water (1:5w/v) present milk-clotting activities of 830SU/g and 504,6SU/g respectively. The maximum enzyme productivity of *Mucor* sp. (4800SU/g) was obtained under the optimum conditions of temperature (25°C), incubation time (96h), moisture content of solid substrate (43%) adjusted suitably with mineral solution (Czapek-Dox) of pH 3. The crude extract of *A. niger* FFB1 was applied in the production of the pressed and unripened cheese, where the others extracts (*R. stolonifer* and *Mucor* sp.) were used in the manufacture of cheese kind Edam using raw milk according to the conventional process of manufacture. The yields of cheese obtained by the commercial preparation and by the coagulating enzyme from *A. niger* FFB1 (a) differ only by 2.8%, where the organoleptic qualities of the two hard cheeses produced are very close. In the case of *R. stolonifer* (b) and *Mucor* sp. (c,c'), the yields cheese of 21.56g/l and 90.84g/l were obtained using the crud extract produced by this strain, where the commercial rennet gives a yield of 104.22g/l. The results obtained are encouraging and require more work to optimize the production procedure, but also pave the way to test the ability of the fungal crud extracts and the purified enzymes (acid proteases of *A. niger*, *R. stolonifer* and *Mucor* sp.: 47.7kDa, 43kDa and 36 kDa respectively) in the production of this kinds and other types of cheese.

Biography

Souhila Bensmail is a PhD student at University of M'Hamed Bougara (Boumerdès) and a teacher at the University of Akli Mohand Oulhadj (Bouira) Algeria. FETHIA FAZOUANE-Naimi Professor Researcher University of Boumerdes (ALGERIA), Engineer in food industry at university of Constantine (Algeria), Magister en nutrition, Doctorate biochemistry-endocrinology at the same university Researcher group head "Biotechnology and Environment" in laboratory of food technology, faculty of Engineer Science, University of Boumerdes, President of scientific committee of Department of Biology (2001-2003), (2006-2008) and (2008-2011), Responsible of Applied Biochemistry Master since October 2006, Responsible of magister post graduation "Biochemistry and Microbiology"; Teaching materials "Structural and Metabolic Biochemistry, Food Biochemistry", Department of Biology, Faculty of Science, University M'Hamed Bougara of Boumerdes. Expert at the Centre Regional Conference for validation of specifications LMD training. Head of Scientific research project 2004-2014 "Ability project manufacturing cheese substitutes from rennet. Case of traditional cheese and hard cheese "and "Production, purification and characterization of bacterial and fungal proteases (origin local strains) for use in cheese" and "Optimization of proteases production by local mold for use in food".

souhilabensmail@yahoo.fr