

Title: Application of Milk-Clotting Protease from *Aspergillus oryzae* DRDFS13 MN726447 and *Bacillus subtilis* SMDFS 2B MN715837 for Danbo Cheese Production.

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Abstract

This study aimed to investigate the efficiency, biochemical composition, and sensory quality of Danbo cheese produced using proteases derived from the fungus and bacterium compared to the commercial product. A fungal enzyme from *Aspergillus oryzae* DRDFS13MN726447 and a bacterial enzyme from *Bacillus subtilis* SMDFS 2B MN715837 were produced by solid-state and submerged fermentation, respectively. The crude enzyme from *A. oryzae* DRDFS13 and *B. subtilis* SMDFS 2B was partially purified by dialysis and used for Danbo cheese production using commercial rennet (CHY-MAX® Powder Extract NB, Christian Hansen, 2235 IMCU/g) as a control. The Danbo cheese produced using dialyzed fungal enzyme (E1) (267 U/mL), dialyzed bacterial enzyme (E2) (522 U/mL), and commercial rennet (C) were analyzed for body property, organoleptic characteristics, and proximate and mineral composition when fresh and after 2 months of ripening. There was no significant difference in the cheese yield (C = 9 kg, E1 = 8.6 kg, and E2 = 8.9 kg) among the three treatments. The body properties of Danbo cheese produced with the fungal enzyme (E1) were firm and acceptable as the control (C), whereas the Danbo cheese produced by bacterial enzymes has shown a watery body. The overall organoleptic characteristics of Danbo cheese produced by the fungal enzyme (5.3) were similar to control cheese produced by commercial rennet (5.5). Both cheese types were significantly different in organoleptic properties from Danbo cheese produced by the bacterial enzyme (4.9). There was no significant difference () in the proximate composition between the ripened Danbo cheese produced by fungal enzyme and the control cheese except for crude protein content. However, the ripened cheese products showed a significant difference in their mineral composition except for sodium. In conclusion, this study demonstrated that the fungal enzyme from *Aspergillus oryzae* DRDFS 13 is more appropriate for Danbo cheese production than the bacterial enzyme from *Bacillus subtilis* SMDFS 2B. However, it requires further application of the enzymes for the production of other cheese varieties.

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