Analysis of fungi community during fermentation of Thai fermented pork (nham) using Denaturing Gradient Gel Electrophoresis (DGGE) technique

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Thai fermented pork or nham is well known for lactic acid bacteria fermented product. However, fungi especially yeast has been suspected to play some roles in flavour and colour development at some point during nham fermentation. In this work, fungi community and their changes throughout the nham fermentation period, up to 15 days, were studied. Two formula of nham, with and without starter culture (Lactobacillus plantarum), were compared. DGGE technique, which was popularly used to analyse microbial community in an ecosystem including food products, was used. A primer set, EUK1427F-1616R, targeted at 18S ribosomal RNA gene of Eukaryotes was used to amplify the total DNA extracted from the nham samples at different time points of the fermentation (0, 2, 3, 4, 5, 7, 10 and 15 days). A total of 10 distinct band positions were found on the DGGE gel for both types of nham. An attempt to identify the band by direct sequencing was failed due to co-migration of different PCR products to the same location on the DGGE gel. The bands were hence identified by cloning into pDrive-cloning vector and several clones were sequenced. From sequencing data, seven band locations were identified as the DNA amplified from pig, garlic, and possible chilli plant which were the major component and ingredients in nham. Three bands were identified as DNA from fungi, two in the family Saccharomycetaceae and one as Candida tropicalis. Notably, all three fungi bands appeared only after 10 days of fermentation in naturally fermented nham and after 15 days in starter culture added nham. Therefore, it could be concluded that the fungi community did not play a significant role in nham fermentation since the fermentation usually stopped at 3-5 days when pH of nham reached 4.6 and was ready for consumption.

Keywords : DGGE, fungi diversity, fermented pork, nham