A standardized conjugation protocol to assess antibiotic resistance transfer between lactococcal species - DTU Orbit (23/01/2019)

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Optimal conditions and a standardized method for conjugation between two model lactococcal strains, Lactococcus lactis SH4174 (pAM beta 1-containing, erythromycin resistant donor) and L. lactis Bu2-60 (plasmid-free, erythromycin sensitive recipient), were developed and tested in a inter-laboratory experiments involving five laboratories from different countries. The ultimate goal of the study was to assess the microbial potential of antibiotic resistance transfer among Lactic Acid Bacteria (LAB). The influence of culture age (various OD values) and ratios of donor and recipient cultures as well as filter, solid and liquid mating techniques, were examined in order to optimize the conjugation protocol. In the result of these studies, we concluded that the donor-to-recipient ratio appear to be important: the most efficient technique for conjugation was filter mating and the optimal conditions for gene transfer were observed when late logarithmic Cultures of both donor and recipient were used. Comparison of conjugal transfer frequencies between five partner laboratories showed that results are sufficiently inter-laboratory repeatable and inter-laboratory comparable. This is the first study of this kind, in which a standardized protocol of conjugal mating for testing antibiotic resistance dissemination among LAB was established and validated. (C) 2008 Elsevier B.V. All rights reserved.

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