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Isolation, identification and characterization of probiotic Lactobacilli spp. from Tarkhineh

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C Tarkhineh" is a traditional fermented food produced from a mixture of spontaneously fermented butter milk and wheat flour in Iran. Nine samples of Tarkhineh were collected from different rural areas in Kermanshah province, Iran. The isolates were grouped and identified using a combination of phenotypic and genotypic methods including repetitive extra-genic palindrome polymerase chain reaction (REP-PCR) fingerprinting, biochemical methods and carbohydrate profiling and then evaluation of the probiotic properties of them. According to the results these 54 isolates belonged to *Lactobacillus plantarum* (19), *Lactobacillus fermentum* (17), *Lactobacillus pentosus* (9), *Lactobacillus brevis* (8) and *Lactobacillus diolivorans* (1) that profile bonding from rep-PCR showed *Lactobacillus plantarum* has high intra-species diversity. Media of pH=2.0–7.0 and bile salt concentrations of 0.0-0.5% were used as stress conditions. Antibacterial activity of the probiotic *Lactobacillus* was determined by means of the spot-on-lawn method. In conclusion, our results showed that 3 strains have potential probiotic value that two of them were *Lactobacillus fermentum* and one of them was *Lactobacillus plantarum*.

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Catabolism of cholesterol rings C and D by Mycobacterium tuberculosis

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Cholesterol catabolism is critical for *Mycobacterium tuberculosis* (*Mtb*) virulence. However, many aspects of this catabolism remain unknown. In *Mtb* and related *Actinobacteria*, the cholesterol side chain and rings A and B are degraded to yield $3a\alpha$ -H-4 α (3'-propanoate)- $7a\alpha$ -methylhexahydro-1,5-indanedione (HIP), a metabolite that contains steroid rings C and D. HIP catabolism is initiated by thioesterification to HIP-CoA and appears to be specified by the enzymes encoded by the KstR2 regulon. The KstR2 regulon of *Mtb* is up-regulated during growth on HIP and contains several genes encoding putative β -oxidation-type enzymes, ipdAB, and *Rv3553*. Both *ipdAB* and *Rv3553* are essential for cholesterol catabolism by *Mtb*, but only ipdA appears to be essential for its survival in infection models. A $\Delta ipdAB$ mutant of *Mtb* was slowly killed when incubated with cholesterol, and did not grow on glycerol in presence of cholesterol, suggesting it accumulated bacterio-toxic metabolites. Furthermore, deletion of *ipdAB* and *ro04649*, the ortholog of *Rv3553*, in Rhodococcus jostii RHA1 (RHA1) abrogated growth on cholesterol and HIP, and significantly delayed growth when these strains were incubated in presence of HIP and a second carbon source, suggesting generation of toxic metabolites from HIP. Similar results were obtained by deleting other KstR2-regulated genes in Mycobacterium smegmatis. Using LCMS-based metabolite profiling, we identified a number of previously undescribed cholesterol-derived CoA thioesters in deletion mutants. Some of these were characterized as ring C-opened metabolites. Altogether, these results allow us to suggest specific roles for several KstR2-encoded enzymes and to propose a model for HIP catabolism in *Mtb*.

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