Introduction: The use of Lactobacillus as probiotics requires the application of accurate and reliable methods for detection and identification of bacteria at the strain level. The aim of this study is evaluation and detection of genetic diversity of Lactobacilli species isolated from different sources in Iran based on REP-PCR.

Method: Twenty strains were isolated from Iranian traditional yoghurt, cheese, and Tarkhineh. PCR-mediated amplification was carried out by degenerate primers. Sequencing of 16s rDNA was performed after purification of the PCR product. The rep-PCR fingerprinting by REP-1 oligonucleotide primer was carried out for discrimination of isolates. Dice similarity was determined among the strains studied and used for grouping of the genotypes by UPGMA clustering methods and PCA analysis. Isolates were deposited as novel stains of Lactobacillus casei, brevis, plantarum, and Entrococcus facium in GenBank. The 20 isolates produced different banding patterns, with 13 visualized PCR products in the range of 200 to 2500 bp. Clustering methods performed on molecular data by two different software (NTSYS and Darwin), produced similar results which were also supported by PCA ordination plot. The REP-PCR fingerprinting grouped all studied isolates into a few clusters as four main clusters were observed in dendrograms. In all analysis, isolates of Lactobacillus casei, brevis, plantarum, Entrococcus facium form four separate clusters. Conclusions: The REP-PCR profiles showed that 20 type isolates produced different banding patterns. Thus it has been proved that REP-PCR appears to be a very practical method and highly sensitive in discrimination of the Lactobacillus species.