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Whole-Genome Shotgun Sequencing of *Lactobacillus rhamnosus* MTCC 5462, a Strain with Probiotic Potential

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Lactobacillus rhamnosus MTCC 5462 was isolated from infant gastrointestinal flora. The strain exhibited an ability to reduce cholesterol and stimulate immunity. The strain has exhibited positive results in alleviating gastrointestinal discomfort and good potential as a probiotic. We sequenced the whole genome of the strain and compared it to the published genome sequence of *Lactobacillus rhamnosus* GG (ATCC 53103).

actobacillus rhamnosus is a bacterium that was originally considered to be a subspecies of Lactobacillus casei, but later genetic research found it to be a species of its own. The L. rhamnosus MTCC 5462 (earlier Lactobacillus acidophilus) LBKI4 strain was originally a gastrointestinal isolate from an infant fecal sample collected at Anand Agricultural University in India (3). The L. rhamnosus MTCC 5462 strain was able to tolerate bile, phenol, and salt concentrations up to 2.4%, 0.4%, and 4%, respectively. Inhibition of growth of pathogenic microorganisms during production and storage of cultured milk (4) and antibacterial activity against milk spoilage bacteria (5) are suggestive of its potential application in the dairy industry. The strain was reported to have bile tolerance and bile deconjugation and cholesterol-reducing properties (1). The positive implantation of a strain with potential to decrease coliform count in human subjects further validates the probiotic potential of the strain (6, 9, 10).

The genome sequence of *L. rhamnosus* GG (ATCC 53103) has been decoded (2, 8). As a probiotic, *L. rhamnosus* ATCC 53103 is claimed to colonize the digestive tract and balance intestinal microflora; however, *L. rhamnosus* is more likely a transient inhabitant, not autochthonous (11).

The whole-genome sequencing of strain *L. rhamnosus* MTCC 5462 was performed using Roche GS 454 technology (7). The reads obtained with 3.67-fold genomic coverage were assembled using GS Reference Mapper software V.2.3 using the *L. rhamnosus* Lc 705 genome as a reference to generate a 2,031,259-bp-long single chromosome. The genome annotation was carried out, and comparative analysis of the genome was done with the published complete genome of strain ATCC 53103 (8).

The genomic sequence of *L. rhamnosus* MTCC 5462 (2.03 Mb) was smaller than those of *L. rhamnosus* ATCC 53103 (3.01 Mb) and *L. rhamnosus* Lc 705 (2.96 Mb).

In strain *L. rhamnosus* MTCC 5462, 4,000 coding sequence (CDS) regions were identified, which is larger than those of *L. rhamnosus* ATCC 53103 (2,913 CDSs) and *L. rhamnosus* Lc 705 (2,878 CDSs). A total of 49 RNA coding regions were identified, a number which is lower than those of *L. rhamnosus* ATCC 53103 (72) and *L. rhamnosus* Lc 705 (76).

Respectively, 278 and 308 subsystems were reported in a metabolic reconstruction study for *L. rhamnosus* MTCC 5462 and *L. rhamnosus* ATCC 53103. The subsystem analysis revealed the common absence of subsystem structures for photosynthesis, iron acquisition and metabolism, motility and chemotaxis, and secondary metabolism in both organisms. However, comparative analysis in other subcategories was indicative of close genetic and biochemical similarity between *Lactobacillus helveticus* MTCC 5463 and *L. rhamnosus* ATCC 53103.

L. rhamnosus MTCC 5462 showed an absence of 103 genes distributed in the categories of amino acids and derivatives; carbohydrates; cell division and cell cycle; cell wall and capsule; cofactors; vitamins; prosthetic groups; pigments; DNA metabolism; fatty acids, lipids, and isoprenoids; phages; prophages; transposable elements; plasmids; potassium metabolism; protein metabolism; RNA metabolism; regulation and cell signaling; respiration; stress response; virulence; disease; and defense.

The similarity in the absence of these genes may be due to results of the adaptation process in response to different microenvironments in the gastrointestinal tracts of human populations due to geographical and physiological differences.

Nucleotide sequence accession numbers. The complete sequence of the *L rhamnosus* MTCC 5462 genome can be accessed under the GenBank accession numbers AEYM01000001 to AEYM01002543.

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Received 5 December 2011 Accepted 19 December 2011 Address correspondence to S. J. Jakhesara, drsubhash81@gmail.com. Copyright © 2012, American Society for Microbiology. All Rights Reserved. doi:10.1128/JB.06644-11 *acidophilus* in human vignal and gastero-intestinal tracts. Indian J. Comp. Microbiol. Immunol. Infect. Dis. **12**:107–109.

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