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CHOLESTEROL-LOWERING PROBIOTICS: A NEW FRONTIER IN CARDIOVASCULAR DISEASE PREVENTION

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Cardiovascular diseases (CVDs), particularly acute myocardial infarction, remain the leading cause of mortality worldwide. One of the main risk factors is hypercholesterolemia. Although pharmacological therapy (e.g., statins) is effective, it is associated with serious side effects, which have stimulated interest in alternatives such as probiotics, postbiotics (metabolic products of probiotic microorganisms), and functional foods enriched with probiotic biomass.

Probiotics show potential in reducing the risk of CVDs through the following mechanisms of action: reducing cholesterol absorption (influencing the expression of the Niemann-Pick C1-like 1 gene (NPC1L1)); deconjugation of bile acids, reducing their reabsorption; production of short-chain fatty acids (SCFAs), which inhibit 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase; lowering levels of trimethylamine N-oxide (TMAO), a compound associated with atherosclerosis; exerting antioxidant and anti-inflammatory effects (via inhibition of nuclear factor kappa-light-chain-enhancer of activated B cells (NF- κ B)).

Randomized studies have shown that low doses of probiotics ($<10^{10}$ CFU/day) are more effective in lowering low-density lipoprotein cholesterol (LDL-C), that an intake duration of ≥ 12 weeks is associated with improved lipid profiles and that multi-strain compositions are more effective than single strains.

All tested lactic acid bacteria strains met essential probiotic criteria, including gastrointestinal tolerance; antagonistic activity (all strains inhibited pathogenic and opportunistic microorganisms, including *Candida*); biocompatibility; adhesion ability (all strains were highly adhesive, ranked: *L. casei* IMV B-7280 $>$ *B. bifidum* VK-1 $>$ *B. longum* VK-2 $>$ *L. bulgaricus* IMV B-7281 $>$ *L. acidophilus* IMV B-7279); antibiotic resistance profile; *in vitro* cholesterol-lowering activity (*L. casei* IMV B-7280 $>$ *L. bulgaricus* IMV B-7281 $>$ *L. acidophilus* IMV B-7279 $>$ *B. longum* VK-2 $>$ *B. Bifidum* VK-1); and synergistic effects in strain combinations.

The study revealed that probiotic strains exhibited increasing cholesterase activity over time in a mouse model of hypercholesterolemia which we developed. *L. acidophilus* IMV B-7279 showed the highest activity across multiple conditions, reaching up to $78.04 \pm 3.0\%$ by day 7 in female mice under prophylactic administration. The combination *B. bifidum* VK-1 + *B. longum* VK-2 (1:1) was also highly effective (up to $74.08 \pm 3.0\%$).

Prophylactic schemes consistently resulted in higher cholesterase activity compared to therapeutic ones. Younger mice (16–18 g) responded better to probiotics than older ones (18–20 g), with greater increases observed over the 7-day period.

Overall, the most effective strains were *L. acidophilus* IMV B-7279 and *B. bifidum* VK-1, as well as the composition *B. bifidum* VK-1 + *B. longum* VK-2 (1:1), with cholesterol-lowering activity ranging from 40–78%.

Conclusions.

The studied probiotic strains meet the key criteria for effective probiotics and demonstrate significant potential for the development of functional products with hypocholesterolemic effects aimed at the prevention and adjunctive treatment of CVDs.

Future research will involve the use of small animal models of CVD to further evaluate the *in vivo* therapeutic effects of these cholesterol-lowering probiotic strains. Employing animal models that closely mimic human pathophysiology is essential for elucidating the mechanisms underlying cardiovascular diseases and for validating the clinical relevance of probiotic interventions.