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Editorial

Use of food grade lactic acid bacteria for the prevention of undesired fungal growth

Loss of crops due to diseases and food materials including fruits, vegetables and also mushroom fruit bodies by pathogenic or spoilage microorganisms especially by fungi is of great concerns of todays agricultural scientists. A number of techniques as well as agents are being employed to prevent such losses.

Applications of fungicides and various preservatives, and not providing the ideal conditions for fungal growth viz., cold storage during post-harvest period are the commonest ways we adopt to prevent fungal contamination and infection. The major drawbacks of such practices are multiple. Even the best chemical fungicide whether it is a systemic fungicide like Bavistin or a bio-preservative like sodium- benzoate or -propionate it has certain negative impact on human health upon consumption. This thing got worsen when such or other chemical agents are applied directly onto the surfaces of food crops and vegetables. In such applications we have minimum scope to wash the chemical agent out before its consumption.

There are certain biocontrol agents viz., species of *Trichoderma* and others who can very effectively kill the undesired fungal organisms by their various metabolic potentials. In most of the occasions they have strong chitinolytic potential due to their different classes of chitinase producing capabilities. Alongwith that they produce other cell wall degrading enzymes â-glucanases and proteases, and also produce an array of antifungal metabolites like HCN, iturin, fengycin, surfactin etc. and kill the contaminating fungal organisms. But for human consumption acceptability of them become a big question in terms of toxicity on mammalian cell and systems.

Thus, searching a suitable bio-control agent would become very imperative which itself is food-grade in nature or are obtained from ready-to-eat good quality naturally fermented products. Probiotic lactic acid bacteria (LAB) fulfil these requirements and come into scientific research as boon.

The idea that LAB are a class of organisms emerged in the late 1800s. Scientists celebrated Pasteur’s important discovery of lactic acid fermentation in 1857 and Lister’s first isolation of a pure bacterial culture in food in 1873. Several bacteria in this category are classified as GRAS (generally recognised as safe) by the US Food and Drug Administration (US FDA). The term “lactic acid bacteria” mentions to a broad, diverse categories of microbes which produce lactic acid from lactose. They belong to approximately 20 genera and include *Lactobacillus, Pediococcus*, *Lactococcus* and others. These LAB, produce an array of antimicrobial ingredients such as organic acids, reuterin, reutericyclin, diacetyl, hydrogen peroxide, antifungal peptides, acetoin, and bacteriocins. These LAB are Gram positive, usually non-motile, non-spore forming rods and cocci. However, no report was there on this technique being used in mushroom cultivation and preservation. Furthermore, certain strains of LAB possess interesting health-promoting properties; and act as probiotics, have the potential to combat gastrointestinal pathogenic bacteria (*Escherichia coli, Helicobacter pylori, and Salmonella*). Numerous antagonist activities were already reported for LAB that they have potentialities to prevent the growth of food pathogens of Traditional Iranian Cheese.

In order to evaluate their antifungal properties, various LAB strains are examined. *Lactobacillus acidophilus* produced an active substance against *Candida albicans* that was initially identified by Marth and Hussong. LAB are quite safer and cause almost no side-effects on mammalian system. So, concerns about bio-preservatives are becoming most popular now a days where LABs are most useful. Several species of *Lactobacillus* (*L. plantarum, L. pentosus* and *L. brevis*) and their cell free supernatant (CFS) had exhibited antifungal activity towards *Aspergillus niger, Penicillium* sp*, Fusarium oxysporum, Rhizopus* sp. as well as prevent the growth of some yeasts, viz., *Candida pelliculosa* and *Rhodotorula* sp. Utilizing the *L. plantarum* S61 strain as a food biopreservative and a preventive measure against yeast and mold-related gastrointestinal disorders can be effective.

Chitin, a polymer present in the cell walls of fungi and the exoskeletons of insects, is broken down by *Lactobacillus* species’ chitinase enzymes. The probiotic qualities of *Lactobacillus* are enhanced by these enzymes, which also boost gut health and function as biocontrol agents in agricultural applications. These consequences are significant from an ecological and biotechnological standpoint. Through several processes of metabolism, *Lactobacillus* species make use of chitin, a polymer of N-acetyl glucosamine. The synthesis of chitinase enzymes, which disintegrate chitin into more easily absorbed parts that the bacteria can process, is a key factor of these systems. Chitinases, produced by *Lactobacillus*, hydrolyze the β-1,4 links in chitin, releasing N-acetyl glucosamine (GlcNAc) monomers and then absorb these monomers and use them for further metabolism. Therefore, degrading chitin and utilizing their chitinolytic activity, *Lactobacillus* strains can be employed in agricultural settings to combat fungal infections. Some investigations identified the presence of chitin-binding proteins (CBPs) and the chitinolytic activity of *Lactobacillus plantarum*. *chiA* gene of *L. plantarum* was amplified effectively by using the primers (chiFEMSF and chiFEMSR primers) already reported earlier. As a result of this enzyme activity produced by LAB fungal mycelia got degraded as beautifully evidenced through scanning electron microscopic studies as performed in author's laboratory and in some other laboratories abroad. Incorporating *L. plantarum* in food products can help to prevent fungal spoilage, extending the shelf life of perishable items. In the gastrointestinal tract, *L. plantarum* can prevent harmful fungi from growing, contributing to a balanced microbial community and supporting gut health.

LAB particularly different subspecies of *Lactococcus lactis* have enormous potential to produce antimicrobial peptides which can prevent contaminating bacterial growth during preservation in one hand, and on the other hand they also produce antifungal principles which prevent the growth of human pathogenic fungi *Candida albicans* and *Trichophyton gypseum.*

Management of mycotoxin production in stored seed grains of food crops such as chickpea, groundnut, maize etc. is also very important. Various species of LAB have already shown their potential in managing this mycotoxin production. In a proper formulation when LAB are grown after admixing with the crop seeds, (i) growth of the invading spoilage fungal pathogens are strongly prevented, (ii) mycotoxin production is reduced significantly by the surviving spoilage pathogens and (iii) degradation of mycotoxins produced into their inactive forms by the involvement of LAB or their metabolites. The author's group confirmed that *Limosilactobacillus fermentum* effectively degrade the mycelia of several mycotoxin producing molds. Also, the LAB species could detoxify the aflatoxin contamination as observed in a bioassay experiment with mouse macrophage cell line (RAW 264.7) and western blot analyses.

Making a suitable formulation is very crucial in getting effectiveness of potent lactic acid bacteria. Cellulosic carrier materials are good but survival index is not very promising in such materials. This problem is overcomed when natural prebiotic compounds viz., inulin, maltodextrin and other plant derived compounds like *Sechium edule* (Jacq) and *Dioscorea alata* L. are amended in the formulation. Recently scientists have surprised to see that many potent lactic bacteria those are growing on plant surfaces do not cause any tissue damage, rather they enjoy the released oligosaccharides as their food and add many benefits to the host plant analogically similar to those of mammals.

Probably same things happen when presence of LAB alone or in combination with other PGP organisms control a number of plant diseases. Fruit rot of jackfruit caused by *Rhizopus* spp. and wilt of *Ocimum sanctum* by *Alternaria* sp. are worth to mention.

Culturing mushrooms, both its spawn and cultivation as well as post-harvest preservation is also a challenge from microbial attack, particularly mould spoilage. Mushrooms undergo a variety of physical and chemical changes throughout the postharvest phase, including microbial deterioration, moisture loss, discoloration, texture loss, and nutritional loss. Many techniques of preservation, involving thermal, physical, and chemical treatments, are employed to enhance the shelf-life and maintain the quality of mushrooms. Unfortunately, these methods are harmful to humans or to the nutritional makeup of mushrooms. The application of chemicals to manage infections is comparatively costly, has environmental risks, and is deemed hazardous due to its high degree of toxicity to humans. Food grade LAB can also contribute a lot here too. Nevertheless, there were no reports on the use of LAB during the cultivation and preservation of mushrooms. Therefore, any one's endeavour, in this line will definitely contribute something meaningful to act against mushroom spoilage organisms.

Multiple factors are operating in maintaining the selective toxicity of LAB or their metabolites against mushroom fungi in one hand and pathogenic/spoilage fungi in the other hand. Now-a-days it is well accepted that mushrooms produce remarkably good prebiotic compounds which can support the growth of lactic acid bacteria, thus there might be some kind of symbiosis or cross talk during the interaction of mushroom mycelia with LAB. This need to be worked out in a focused way. Another important factor is the specificity of LAB chitinase towards the nature of chitin of both the fungal organisms. Bioinformatic predictive analyses indicate more affinity of LAB chitinases towards α-chitinases which are normally found in *Rhizopus* and other spoilage moulds, whereas, mushroom mycelia contain γ-form of chitins. Whole genome data of the respective LAB also endorse the presence of similar nature of chitinases.

Judicious application of food grade LAB will have enormous prospects and researchers should explore the possibilities of exploiting them with more and more emphasis. Application of such beautiful microbial wealth may have opened up the avenues of entrepreneurship in the food and agriculture sector and boost up the health of human beings.

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