

## Effect of Edible Mushroom on The Development of Lactic Acid Bacteria

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### Abstract

Currently, many products for human health are available in the market as prebiotics and probiotics. Prebiotics are defined as indigestible dietary fibres and oligosaccharides that promote the growth of lactic acid bacteria in the gastrointestinal tract. With evidence showing that the microbiota is undoubtedly linked to overall health, the discovery of new prebiotics acting on functional foods and beneficial bacteria is important. Mushrooms are well known for their organoleptic qualities as well as for harbouring many different bioactive substances with beneficial health effects. Fungi are of increasing interest as a potential source of prebiotic substrates. Developing a new potential prebiotic from inexpensive and abundant materials such as mushrooms is one of the aspects to be considered. Prebiotic compounds obtained through fungal extraction have been used in numerous research as non-digestible dietary ingredients to show that they inhibit pathogenic organisms and promote probiotic growth. It will take more thorough research to ascertain the metabolic pathways that are triggered in the process of probiotic strains utilizing fungal polysaccharides. Future projections for the food industry indicate that mushrooms will become even more significant. This study examined mushrooms' potential as a prebiotic source and provided a detailed description of its theory and use.

**Key Words:** Lactic acid bacteria, edible mushroom, prebiotic, probiotic, functional foods

### Introduction

Although there are many members of the fungi kingdom, including yeasts, moulds and cap fungi, the word mushroom refers to large, fleshy, edible and medicinal fungi with a distinct sporocarp that are above or below ground, visible to the naked eye, that can be collected by hand. Macrofungi are defined as those sporocarps that have been utilized worldwide for centuries as both food and traditional medicine. Fungi, which do not have the ability to photosynthesise because they do not contain chlorophyll, feed symbiotically, parasitically and saprophytically. Many play important roles in the degradation and recycling of various agricultural and forest wastes and complex biopolymers (Miles and Chang, 2004). It is considered that there are around 1.5 million species of mushrooms on earth and although around 3000 are known to be edible, around 60 species can be cultivated. As mushroom cultivation has become more popular around the world, more than 700 mushroom species have been recognized as safe for human consumption and beneficial to human health. Common mushroom species produced under suitable ecological conditions include *Lentinula edodes*, *Volvariella volvacea*, *Agaricus* sp., *Pleurotus* sp., *Hericium* sp., *Ganoderma* sp., *Grifola frondosa* and *Coprinus* sp. (Cerletti et al., 2021). *Hypsizyugus tessellatus*, *Lentinula edodes*, *Agaricus bisporus*, *Pleurotus ostreatus* and *Flammulina velutipes* are some of the most widely cultivated mushroom species in the world (Niego et al., 2021). There has been a notable increase in interest among consumers in the potential health benefits of incorporating mushrooms into their diets. This is because mushrooms are increasingly recognized as a source of protein, minerals, essential vitamins, and fiber. In addition, mushrooms contain bioactive compounds with antioxidant, anti-inflammatory, anti-tumor, antimicrobial and antiviral properties that actively promote health and reduce the risk of disease in the human body (Cateni et al., 2022). Edible mushrooms have a carbohydrate and digestible protein composition, are a source of high-quality fiber and have a low fat content (Araújo-Rodrigues et al., 2022). In addition, mushrooms are a good source of vitamins, essential amino acids and minerals (Sousa et al., 2023).

Prebiotics are substances that promote the growth or action of microorganisms that contribute to the benefit of the host and are widely consumed by humans. Consumption of prebiotics increases immune function, reduces long chain fatty acids (LCFA) in the gut, improves colon integrity, reduces the duration of intestinal infections, and reduces allergic response for better digestion (Douglas & Sanders, 2008). Mushrooms are also regarded as a prospective source of prebiotics, given their content of diverse polysaccharides, including chitin, mannans, hemicellulose, glucans, galactans, and xylans (Singdevsachan et al., 2015). Some edible fungi are still an undiscovered treasure trove of prebiotic and bioactive compounds and their beneficial effects on the immune system. Numerous studies have demonstrated the positive effects of several fungal species in the prevention and treatment of chronic illnesses, including diabetes, cancer, obesity, hyperlipidemia, hypercholesterolemia, and neurodegenerative and cardiovascular diseases (Varshney et al., 2013; Roncero-Ramos & Delgado-Andrade, 2017; Ke et al., 2022). Various bioactive compounds derived from fungi have been shown to modify intestinal



polysaccharides, act as prebiotics and reduce pathogen proliferation by promoting the growth of probiotic bacteria in the gut (Kumari, 2020). Prebiotic substrates are present in certain beneficial bacteria, including lactic acid bacteria (*Lactobacillus* sp., *Pediococcus* sp., *Lactococcus* sp., *Leuconostoc* sp. and *Streptococcus* sp.) and bifidogenic bacteria (*Bifidobacterium* sp.). By lowering intestinal pathogens and/or modifying the synthesis of bacterial compounds linked to health, these bacteria benefit the host (Ringo et al., 2010; Margalho et al., 2021).

### Properties of Probiotic Microorganisms

According to the International Scientific Association for Probiotics and Prebiotics (ISAPP), probiotics are defined as non-pathogenic microorganisms that, when administered in sufficient quantities, provide health benefits to the host individual (Paulino do Nascimento et al., 2022). In order for a product to be defined as a probiotic, it must possess a number of specific characteristics. The aforementioned characteristics can be enumerated as follows: the product must be of human origin, demonstrate resistance to bile and gastric acid, survive in the digestive tract, adhere to the intestinal wall, adapt to the natural flora, colonize the digestive tract, secrete antimicrobial products such as bacteriocin, be non-toxic and non-pathogenic, exert beneficial effects on host health, remain stable during production and storage processes, and retain its viability (Şahin, 2018). Concurrently, probiotics must also possess antimicrobial properties (Gülbandılar et al., 2017).

Most of the probiotic microorganisms are included in lactic acid bacteria, which are generally recognized as safe and have GRAS (Generally Recognised As Safe) status. The most commonly utilized bacteria are those belonging to the *Lactobacillus*, *Bifidobacterium*, *Streptococcus*, *Enterococcus*, *Leuconostoc* and *Pediococcus* species (Stavropoulou & Bezirtzoglou, 2020; Buran & Bütünöz, 2022). Especially *Lactobacillus acidophilus*, *L. casei*, *L. rhamnosus*, *L. reuteri*, and *Bifidobacterium lactis* Bb12 species are widely used in commercial preparations since their beneficial effects on health have been proven by clinical trials (Thamacharoensuk et al., 2017). Probiotics must survive in the gastrointestinal tract in order to exert their beneficial effects. *Lactobacillus* and *Bifidobacterium* species are particularly well adapted to this environment, frequently utilizing host-derived glycans for metabolism, which enhances their survival and activity on mucosal surfaces (Zuniga et al., 2018). The regular consumption of probiotics has been linked to a number of beneficial effects, including improvements in gut health, enhanced immunity and protection against certain infections (Aguirre Garcia et al., 2024). The ability of these probiotics to colonize the gut is of significant importance with regard to their efficacy. Several studies have demonstrated that the capacity for colonization of various *Lactobacillus* and *Bifidobacterium* species is dependent on their genetic characteristics and ability to adapt to the host environment (Xiao et al., 2021). In light of the considerable diversity of probiotic products currently available on the market, ensuring the quality and efficacy of these microorganisms is paramount. The specific strain, viability and antimicrobial effect against pathogenic organisms are crucial factors in determining the health benefits of probiotic products (Zawistowska Rojek et al., 2022).

### Lactic acid bacteria

In 1919, Orlo-Jensen first described lactic acid bacteria as rod- or cocci-shaped, Gram-positive microorganisms that are catalase- and nitrate-reductase negative and do not form spores. These bacteria typically produce lactic acid as an end product during carbohydrate fermentation. The inability to synthesize the requisite enzymes and cytochromes precludes the possibility of oxidative phosphorylation in the presence of oxygen. Lactic acid bacteria are capable of surviving in temperatures between 5-10 °C, with some species demonstrating tolerance to salt, acid, or alkyls (Mokoena et al., 2021). They need relatively more complex carbon sources to realize their bioactivities. Therefore, they use complex organic molecules as carbon sources. Lactic acid bacteria need many free amino acid molecules, organic acids and sugars for their growth (Fernandes & Jobby 2022; Sharma et al., 2022).

Microorganisms known as lactic acid bacteria (LAB) are able to ferment carbohydrates like lactose and glucose and turn them into lactic acid (Wang et al., 2021; Abedin et al., 2023). LAB have positive effects on the human immune system in addition to preserving the harmony of the gut flora (Rastogi & Singh, 2022). Recently, LAB is widely used in the production and processing of fermented foods due to their generally accepted probiotic effects, safety and acid-producing properties (Alan & Yıldız, 2022). Regular consumption of foods fermented by LAB has been shown to have a beneficial effect on immune system strength and disease protection (Abdul Hakim et al., 2023). Moreover, LAB is employed in the food industry as a natural antibacterial agent, which plays a role in the preservation of food products through the application of antiseptic and freshness-preserving techniques (Kousha et al., 2022).

LAB produces a wide range of metabolites during fermentation, including exopolysaccharides (EPS), amino acids, organic acids, bioactive peptides, and bacteriocins. Antioxidant properties, cholesterol-lowering effects, and improved mineral absorption are just a few of the host health advantages associated with these metabolites (Juraskova et al., 2022; Zapašnik et al., 2022; Sionek et al., 2023, Tang et al., 2023). In addition to assisting in the preservation of foodstuffs, the metabolic processes of LAB contribute to the enhancement of flavour and texture in fermented foods, thereby increasing their appeal to consumers (Aguirre Garcia et al., 2024). Moreover, antinutrients in food that may prevent the absorption of essential nutrients can be decreased by LAB. This decrement enhances the nutritional profile of fermented foods, increasing their safety and benefits (Zapasnik et al., 2022).



**Table 1.** The most common probiotic-producing lactic acid bacterium (Tamang et al 2016; Onyeaka et al., 2022)

Microorganisms	Strain names
<i>Lactobacillus</i> sp.	<i>Lactobacillus brevis</i> , <i>Lactobacillus fermentum</i> , <i>Lactobacillus delbrueckii</i> , <i>Lactobacillus plantarum</i> , <i>Lactobacillus lactis</i> , <i>Lactobacillus rhamnosus</i> , <i>Lactobacillus acidophilus</i> , <i>Lactobacillus reuteri</i> , <i>Lactobacillus casei</i> , <i>Lactobacillus curvatus</i> , <i>Lactobacillus paraplantarum</i> , <i>Lactobacillus johnsonii</i> , <i>Lactobacillus salivarius</i> , <i>Lactobacillus helveticus</i> , <i>Lactobacillus gasseri</i>
<i>Bifidobacterium</i> sp.	<i>Bifidobacterium longum</i> , <i>Bifidobacterium adolescentis</i> , <i>Bifidobacterium infantis</i> , <i>Bifidobacterium bifidum</i> , <i>Bifidobacterium thermophilum</i> , <i>Bifidobacterium breve</i> ,
<i>Pediococcus</i> sp.	<i>Pediococcus pentoseceus</i> , <i>Pediococcus acidilactici</i> <i>Pediococcus cerevisiae</i>
<i>Streptococcus</i> sp.	<i>Streptococcus intermedius</i> , <i>Streptococcus cremoris</i> , <i>Streptococcus diacetylactis</i> , <i>Streptococcus bovis</i> , <i>Streptococcus suis</i> , <i>Streptococcus lactis</i>
<i>Leuconostoc</i> sp.	<i>Leuconostoc mesenteroides ssp. mesenteroides</i> , <i>Leuconostoc. inhae</i> , <i>Leuconostoc kimchii</i>
<i>Enterococcus</i> sp.	<i>Enterococcus faecium</i> , <i>Enterococcus faecalis</i> , <i>Enterococcus durans</i>
<i>Propionibacterium</i> sp.	<i>Propionibacterium shermanii</i> , <i>Propionibacterium acidipropionici</i> , <i>Propionibacterium freudenreichii</i>
<i>Weissella</i> sp.	<i>Weissella confusa</i> , <i>Weissella cibaria</i> , <i>Weissella paramesenteroides</i> , <i>Weissella hellenica</i> , <i>Weissella kimchi</i>

### Prebiotic Potential of Edible Mushrooms

The development of prebiotics is directed towards fungi, which contain carbohydrates, act as potential prebiotics and are also associated with various health-promoting effects (Abd Rahman et al., 2012). Because of its immunosuppressive properties,  $\beta$ -glucan from *Pleurotus* sp. (Pleuran) is utilized as a dietary supplement. Polysaccharides from oyster mushrooms can promote the growth of colonic microorganisms (probiotics), i.e. act as prebiotics. In the study of the structure and potential prebiotic activity of glucans obtained from *Pleurotus ostreatus* and *Pleurotus eryngii* species, oyster mushroom, a biologically active source of glucans, and *Lactobacillus*, *Bifidobacterium* and *Enterococcus* species were used. Specific glucans from the oyster mushroom were used as a carbon source. The polysaccharide composition of these mushroom extracts and the probiotic species showed different growth characteristics. It was reported that *Lactobacillus* species can be used symbiotically with glucan and proteoglucan obtained from *Pleurotus ostreatus* and *Bifidobacterium* species can be used symbiotically with *Pleurotus eryngii* extracts (Synytsya et al. 2009).

$\beta$ -glucan is derived from a variety of sources, including the cell walls of bread yeast (*Saccharomyces cerevisiae*), cereals (oats and barley), and an array of mushroom species. A study of eight fungi revealed that *Schizophyllum commune* and *Auricularia auricula* exhibited the highest  $\beta$ -glucan content and the lowest cost per  $\beta$ -glucan content, respectively. Despite the established role of  $\beta$ -glucan in immune modulation, there is currently no evidence regarding its interaction with the human gut microbiota. It is hypothesized that the gut microbiota has healing effects through its interaction with indigestible components, particularly fermentable dietary fibre. It is crucial to establish a correlation between the specific microbial communities associated with  $\beta$ -glucan fermentation and the resulting profiles of short-chain fatty acids (SCFAs). Mushroom-derived  $\beta$ -glucan has been demonstrated to possess a prebiotic function that is comparable to that of  $\beta$ -glucan derived from commercial yeast (*S. cerevisiae*) (Chaikliang et al., 2015).

In a different investigation, the impact of polysaccharides from *Ganoderma lucidum* on *Bifidobacterium* species proliferation was ascertained. The prebiotic effect of *G. lucidum* extracts was determined using batch fermentation. It was found that mushroom extracts increased the number of *Bifidobacterium* species by 0.3 and 0.7 log<sub>10</sub> cell/mL units, and the number of *Lactobacillus* species by 0.7 and 1 log<sub>10</sub> cell/mL units, and inhibited the growth of *Salmonella*. The presence of *Bifidobacterium* species was found to increase the rate of organic acid formation in the environment (Yamin et al., 2012).



In the study in which the prebiotic effect of five different edible mushrooms (*Auricularia auricula-judae*, *Pleurotus ostreatus*, *Pleurotus sajorcaju*, *Pleurotus abalonus* and *Volvariella volvacea*) was investigated, the soluble and insoluble polysaccharides of the mushrooms were extracted. The bifidogenic effect was determined using the *Bifidobacterium bifidum* TISTR 2129, *B. breve* TISTR 2130, *B. longum* TISTR 2194 and *B. animalis* TISTR 2195 strains, and the prebiotic index values were subsequently measured. The highest prebiotic index number was observed in the *Pleurotus sajorcaju* mushroom, followed by the *Pleurotus abalonus* mushroom. It was reported that both mushroom species stimulated the growth of *Bifidobacterium* and *Lactobacillus* species in a human intestinal model and inhibited the growth of harmful bacteria (Saman et al., 2016).

Polysaccharides from *Lentinula edodes*, *Flammulina velutipes* and *Pleurotus eryngii* mushrooms were found to increase the viability of *Bifidobacterium longum*, *Lactobacillus casei* and *Lactobacillus acidophilus* at different concentrations in MRS medium. Mushroom polysaccharides were found to maintain the number of probiotics above  $10^7$  cfu/mL during storage in yoghurt cultures. The researchers noted that fungal polysaccharides are a cheaper source of raw materials for obtaining new prebiotic components. Liu et al. (2015) investigated the prebiotic properties and immune system supporting effects of *Ganoderma lucidum* residues during tea preparation. They used *G. lucidum* extracts in the diet of chickens. *G. lucidum* was found to stimulate the growth of *Lactobacillus rhamnosus* and *Bifidobacterium longum*. The probiotic activity of bifidobacteria in the feces of chickens was found to be significantly different from the control group. They concluded that *G. lucidum* waste has beneficial effects and can potentially be used in waste recycling.

It was established that *G. lucidum* extracts altered the composition of the intestinal microbiota and resulted in an increase in *Lactobacillus* species in comparison to the control group (Meneses et al., 2016). In a study in which *A. bisporus* mushrooms were fermented with *Lactobacillus plantarum* strains, a sensory analysis, colour analysis and total phenolic compound analysis were conducted. It was reported that mushrooms fermented with probiotic bacteria exhibited elevated phenolic component and antioxidant activity values (Jabłońska-Ryś et al., 2016).

A study was conducted to examine the growth of *Lactobacillus acidophilus* and *L. plantarum* in MRS broth medium containing mushroom extracts. Seven different edible mushroom extracts, including *Auricularia auricula-judae*, *Lentinus edodes*, *Pleurotus citrinopileatus*, *Pleurotus djamor*, *Pleurotus pulmonarius* and *Pleurotus ostreatus*, were investigated for their prebiotic properties. It was reported that the *L. edodes* extract had the greatest stimulatory effect on the growth of the probiotic bacteria, with the *L. acidophilus* and *P. pulmonarius* extracts demonstrating the most significant impact on the growth of *L. plantarum*. In the analysis of gastrointestinal tolerance of probiotics, the highest viability rate was observed in the *P. djamor* extract containing *L. acidophilus* (Sawangwan et al., 2018).

In the study in which the effect of *P. ostreatus* mushroom powder as a prebiotic on yoghurt quality was investigated, mushroom powder was incorporated into the yoghurt production process at concentrations of 0%, 0.5%, 1% and 1.5%. It was reported that the addition of mushroom powder resulted in an increase in lactic acid concentration and the number of lactic acid-producing bacteria (LAB) present (Tupamahu & Budiarmo, 2017). The highest viability of LAB was observed in samples containing 1.5% mushroom powder. In a further study, the prebiotic effect of three different edible mushroom species, namely *Pleurotus sajorcaju*, *P. florida* and *L. edodes*, was compared with that of a range of commercial prebiotics. The optimal prebiotic effect was observed in the medium containing *P. sajorcaju* mushroom, in which *L. acidophilus* was able to grow (Mallik & Bhawsar, 2018). In a study of 53 naturally occurring mushroom extracts, probiotic bacteria (*L. acidophilus* and *L. rhamnosus*) were used in comparison with commercial prebiotics, namely inulin and fructooligosaccharide. The results demonstrated that the tested fungi exhibited a greater capacity to stimulate the growth of *Lactobacillus* species than commercially used prebiotics. It has been demonstrated that mushroom polysaccharides are able to reach the intestine without undergoing alteration in the stomach, where they have been observed to stimulate the growth of beneficial bacteria (Nowak et al., 2018).

## Conclusion

Probiotics and prebiotics are the most scientifically studied active ingredients due to their biotherapeutic properties. Among probiotics, *Bifidobacterium* and *Lactobacillus* species are bacteria that have become increasingly important due to their use as starter cultures in foods and in tablet form. The definition of the metabolic activities of these microorganisms and the study of their ability to grow in environments containing different carbohydrate sources has led to the definition of "potential prebiotic components". Due to the increasing demand for industrial foods such as dairy products, bakery products, beverages, sauces, confectionery, baby foods, etc. containing commercial prebiotic components, researchers and manufacturers are turning to the cheaper extraction of prebiotic components from natural materials found in nature.



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