Review

jmb

Bioconversion Using Lactic Acid Bacteria: Ginsenosides, GABA, and Phenolic Compounds

Na-Kyoung Lee¹ and Hyun-Dong Paik^{1,2*}

¹Department of Food Science and Biotechnology of Animal Resources, ²Bio/Molecular Informatics Center, Konkuk University, Seoul 05029, Republic of Korea

Received: December 14, 2016 Revised: February 24, 2017 Accepted: March 15, 2017

First published online March 15, 2017

*Corresponding author Phone: +82-2-2049-6011; Fax: +82-2-455-3082; E-mail: hdpaik@konkuk.ac.kr

pISSN 1017-7825, eISSN 1738-8872

Copyright© 2017 by The Korean Society for Microbiology and Biotechnology Lactic acid bacteria (LAB) are used as fermentation starters in vegetable and dairy products and influence the pH and flavors of foods. For many centuries, LAB have been used to manufacture fermented foods; therefore, they are generally regarded as safe. LAB produce various substances, such as lactic acid, β -glucosidase, and β -galactosidase, making them useful as fermentation starters. Existing functional substances have been assessed as fermentation substrates for better component bioavailability or other functions. Representative materials that were bioconverted using LAB have been reported and include minor ginsenosides, γ -aminobutyric acid, equol, aglycones, bioactive isoflavones, genistein, and daidzein, among others. Fermentation mainly involves polyphenol and polysaccharide substrates and is conducted using bacterial strains such as *Streptococcus thermophilus*, *Lactobacillus plantarum*, and *Bifidobacterium* sp. In this review, we summarize recent studies of bioconversion using LAB and discuss future directions for this field.

Keywords: Lactic acid bacteria, fermentation, bioconversion, phenolic compound, β-glucosidase

Introduction

Consumers have become increasingly concerned about healthy and functional foods. The health-food industry has attempted to identify new materials or utilize existing materials in new ways. Intestinal bacteria are involved in metabolic pathways and can influence host health [1, 2]. Lactic acid bacteria (LAB) are present in fermented foods, such as vegetables (kimchi and sauerkraut) and dairy products (yogurt and cheese); therefore, they are generally regarded as safe. Some LAB have probiotic characteristics, including maintenance of intestinal microflora, and antioxidant, anti-allergy, and anticancer properties, and their use has increased with the goal of improving health or nutrition [1, 2]. Probiotic strains can be damaged by exposure to simulated gastric conditions; however, some LAB isolated from fermented foods have a high survival rate under harsh conditions [1].

Traditional starter cultures used in the food industry are selected for their ability to rapidly produce desirable organoleptic qualities; however, probiotic bacteria should be selected for their potential nutritional benefits. Traditional yogurt starter cultures include *Streptococcus thermophilus* and *Lactobacillus delbrueckii* sp. *bulgaricus*. Currently, a number of dairy products are marketed as containing probiotic bacteria such as *L. acidophilus* or *Bifidobacterium* species. Fermented tea containing LAB is thought to have enhanced flavor and phenolic compound bioavailability through decreased contents of epigallocatechin gallate, epigallocatechin, and epicatechin [3]. Tea fermented using microorganism-derived enzymes has been investigated for digestibility, anticancer effects, and prevention and treatment of cardiovascular disease [3–7].

The bioavailability of glucosides is increased by hydrolysis of the sugar moiety using β -glucosidase [8]. The water solubility and chemical stability of aglycone forms make them effective for detoxifying endogenous metabolites and xenobiotics, including defense against mycotoxins [4, 9]. β -Glucosidase removes glucopyranosyl residues from the non-reducing end of β -glucoside by catalyzing the hydrolysis of the glycosidic bond [10]. β -Glucosidases are known to be widely distributed among animals, plants, fungi, yeasts, and bacteria [11]. As producer strains, β -glucosidases have been reported in *L. acidophilus*, *L. delbrueckii* subsp. *bulgaricus*, *L. casei*, *L. plantarum* [12], *L. fermentum*, and several *Bifidobacterium* sp. [5]. Most β glucosidases can hydrolyze a broad range of substrates; however, their huge diversity makes it difficult to predict their aglycone specificity [13].

In this review, we summarize recent studies of bioconversion of representative substances using LAB. The roles of ginsenosides, γ -aminobutyric acid (GABA), hydroxytyrosol, isoflavones, and phenolic compounds in fermentation are considered. Additionally, the direction of future studies is discussed.

Bioconversion of Ginseng or Ginsenoside Using LAB

Definition and Classification of Ginseng

Ginseng (Panax ginseng C.A. Mayer) is a well-known medicinal plant in China, Korea, Japan, and other Asian countries [14, 15]. The major active ingredients of ginseng are ginsenosides, whose functions depend on the molecule type [16-18]. Major ginsenosides have multiple sugar moieties and are hardly absorbed by the human body [15]. The major ginsenosides are glycosides that contain an aglycone with tetracyclic triterpene, and include ginsenosides Rb1, Rb2, Rc, and Rd, and protopanaxatriol-types such as Re and Rg1, which comprise more than 90% of total ginsenosides. Major ginsenosides can be converted into deglycosylated ginsenosides and minor ginsenosides such as F2, compound K (C-K), compound Mc, compound Y, Rg3, Rg2, Rh2, Rh1, and F1. Minor ginsenosides have more effective pharmacological properties than major ginsenosides, and can be bioconverted using heat, enzymes, and microorganisms.

Biofunctional Effects of Ginseng

Ginseng has various biofunctional effects on liver, bone, and brain health in humans. Ginsenosides Rb1, Rb2, and Rb3 influence liver health [19], the immune system [20], and anti-inflammatory functions [18, 21], respectively. Rb1 attenuated plasma aminotransferase activities and liver inflammation to inhibit CCl₄-induced liver fibrosis in rats [19]. Rb2 prevented the lethal infection of the hemagglutinating virus of Japan (HCJ) in mice [20]. Ginsenosides Rg1, Rg2, and Rg3 are involved in bone health [22], neuroprotection [23], and vascular function modulation [24], respectively. In addition, mixtures of Rg1 and Rg2 improve cognitive function through their effects on brain metabolic pathways

J. Microbiol. Biotechnol.

[25]. Rh1 and Rh2 were reported to have anti-inflammatory effects [26, 27]. Rh2 produces anticancer [28, 29] and antidiabetic effects by affecting the secretion of insulin [30, 31].

Bioconversion of Ginsenosides by Enzymes and Microbial Fermentation

The production of the minor ginsenosides Rh1, Rh2, C-K, Rg2, and F2 from major ginsenosides has been evaluated previously (Fig. 1). Ginsenosides Rb1, Rb2, and Rc can be transformed to C-K by human intestinal microorganisms [32], and C-K has been shown to have anticancer effects in tumor cell lines [33]. The efficacy of ginseng depends on the host's intestinal condition, and therefore bioconversion is necessary for specific ginsenoside compounds. L. rossiae DC05, isolated using esculin-MRS agar, was used for bioconversion of the major ginsenosides Rb1 and Re to C-K and Rg2 [34]. Leuconostoc mesenteroides LH1 isolated from kimchi was used to produce β-glucosidase, and to convert Rb1 to Rd, F2, and C-K [35]. The optimum conditions for conversion of Rb1 to C-K were pH 6.0, 30°C, and 72 h. Several recombinant technologies have applied Bifidobacterium strains to the production of ginsenoside aglycones [36, 37]. These bioconversion reactions are used to produce microbial crude enzymes whose mechanisms depend on the bacterial strain. Lee et al. [38] reported changes in ginsenoside composition during fermentation of mixed cultures including Saccharomyces cerevisiae, L. acidophilus, L. plantarum, L. brevis, and Bacillus subtilis. In addition, Jung et al. [39] conducted microbial fermentation of yogurt fortified with red ginseng extract.

Bioconversion of Sodium Glutamate to γ -Polyglutamic Acid (γ -PGA) and GABA

Characteristics, Function, and Synthesis of y-PGA and GABA

PGA has been used as an implantable biomaterial for hydrogel formation because of its biodegradable, nontoxic, and non-immunogenic properties [41, 42]. GABA is a nonprotein amino acid and the major inhibitory neurotransmitter of the central nervous system. GABA has antianxiety, antidepressant, and antihypertensive activities and regulates hormone secretion [43, 44]. GABA is widely distributed in mammals and plants, but its content under natural conditions is low. Therefore, γ -PGA and GABA are produced by chemical synthesis or bioconversion methods developed using microorganisms [45–47]. Various microorganisms have been reported for GABA and γ -PGA production, including *Bacillus* sp., *Escherichia coli, Aspergillus* sp., and LAB (Table 1) [43, 48, 49].

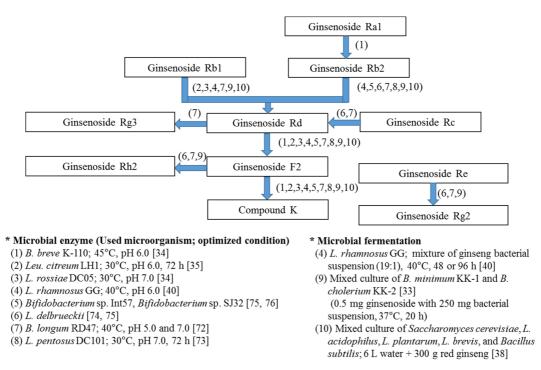


Fig. 1. Metabolic pathway of ginsenoside bioconversion using enzymes and microbial fermentation.

B., Bifidobacterium; Leu., Leuconostoc; L., Lactobacillus; Rb1, 3-O-[β-D-glucopyranosyl-(1-2)-β-D-glucopyranosyl]-20-O-[β-D-glucopyranosyl]-20-O-[β-D-glucopyranosyl]-20(S)-protopanaxadiol; Rb2, 3-O-[β-D-glucopyranosyl]-20-O-[α-L-arabinopyranosyl]-20-O-[α-L-arabinopyranosyl]-20(S)-protopanaxadiol; Rc, 3-O-[β-D-glucopyranosyl]-20-O-[α-L-arabinofuranosyl-(1-6)-β-D-glucopyranosyl]-20(S)-protopanaxadiol; Rd, 3-O-[β-D-glucopyranosyl]-20-O-[α-L-arabinofuranosyl-(1-6)-β-D-glucopyranosyl]-20(S)-protopanaxadiol; F2, 3-O-β-D-glucopyranosyl]-20-O-β-D-glucopyranosyl-20(S)-protopanaxadiol; F2, 3-O-β-D-glucopyranosyl-20-O-β-D-glucopyranosyl-20(S)-protopanaxadiol; F2, 3-O-β-D-glucopyranosyl-20(S)-protopanaxadiol; Rdz, 3-O-[α-L-rhamnopyranosyl-(1-2)-β-D-glucopyranosyl-20(S)-protopanaxadiol; Rdz, 3-O-β-D-glucopyranosyl-20(S)-protopanaxadiol; Rdz, 3-O-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20(S)-protopanaxadiol; Rdz, 3-O-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20(S)-protopanaxadiol; Rdz, 3-O-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20(S)-protopanaxadiol; Rdz, 3-O-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-glucopyranosyl-20-β-D-gl

Bioconversion of Sodium Glutamate to γ-PGA and GABA

Serial coculturing of B. subtilis HA and L. plantarum K154 produced 0.86% GABA from sodium glutamate [42]. Bacillus sp. SW1-2 and *B. licheniformis* produced 12.64 and 75 g/l of γ-PGA from L-glutamic acid, respectively. B. subtilis HA produced 22.5 g/l of γ -PGA from soybeans. The decrease in pH during LAB fermentation may be related to the microbial conversion of glutamate to GABA [50]. L. brevis, isolated from Korean kimchi using gas release and HPLC methods, reportedly produced 17.7–25.8 g/l of GABA with the addition of 70 g/l glutamic acid after a 72 h incubation time [51]. Skim milk fermented by L. helveticus ND01 showed angiotensin-converting enzyme (ACE) inhibitory effects and GABA production [47]. The production of GABA was increased with increasing incubation time, reaching 165.11 mg/l at 30 h. A correlation was observed between the ACE inhibitory activity and GABA content with free amino acid content in fermented milk and soymilk (r = 0.94, p < 0.05). Milk was fermented by L. plantarum PU11 and Lactococcus lactis DIBCA2 [48]; after

fermentation, the milk showed a cell density of 8.0 log CFU/ml and GABA production of 144.5 mg/kg.

Bioconversion of Oleuropein to Hydroxytyrosol Using LAB

Characteristics, Function, and Synthesis of Oleuropein and Hydroxytyrosol

Oleuropein is the most important phenolic compound in olives [52]. However, this phenolic glucoside is responsible for the bitterness of unprocessed olives, and an *L. plantarum* strain has been used to reduce this bitterness [4]. Oleuropein has been shown to have antioxidant, anti-inflammatory, anticancer, anti-atherogenic, antiviral, antimicrobial, hypolipidemic, and hypoglycemic effects [52]. Oleuropein can be converted to hydroxytyrosol by β -glucosidase and esterase [53]. *L. pentosus*, *L. brevis*, and *Pediococcus pentosaceus* were isolated from fermented olives and were found to produce β -glucosidase [54]. Hydroxytyrosol has shown interesting biological properties, such as antioxidant,

Bioconversion ^a (substrate \rightarrow product)	LAB strains	Characteristics
Sodium glutamate $\rightarrow \gamma$ -PGA and GABA	Lactobacillus plantarum K154 and Bacillus subtilis HA	Serial fermentation: first fermentation using <i>Bacillus subtilis</i> HA/ mixed fermentation using two strains/2 nd fermentation with skim milk using <i>Lactobacillus plantarum</i> K154 [42]
Sodium glutamate \rightarrow GABA	Lactobacillus brevis NCL912	Screening in MRS medium with 1% sodium glutamate, pH 5.0 [77]
$MSG \rightarrow GABA$	Lactobacillus brevis NPS-QW-145	Prescreening method using MSG and pH control [51]
	Lactobacillus brevis TCCC 13007	Two-step biotransformation; growing and resting cells [49]
	Lactobacillus brevis K203	Optimization and purification [78]
	Lactobacillus brevis DPC6108	Potential bioconversion in intestinal condition [79]
$Glutamate \rightarrow GABA$	Lactobacillus plantarum DSM19463	Application in grape must; skin protection [50]
	Lactobacillus plantarum PU11	ACE inhibitory effect and GABA production of fermented milk [48]
Skim milk \rightarrow GABA	Lactobacillus helveticus ND01	ACE inhibitory effect and GABA production of fermented milk [47]
White wheat, wholemeal wheat, and rye flours → GABA	Lactobacillus plantarum C48 Lactococcus lactis subsp. lactis PU1	Application in sour dough [46]

Table 1. Bioconversion of sodium glutamate using LAB.

^aMSG, monosodium glutamate; γ-PGA, γ-polyglutamic acid; GABA, γ-aminobutyric acid.

antibacterial, and free-radical scavenging activities, that were higher than those of oleuropein [53].

Bioconversion of Oleuropein to Hydroxytyrosol

Table 2 shows the bioconversion of oleuropein using LAB. Ciafardini *et al.* [55] and Marsilio *et al.* [56] reported that *L. plantarum* strains degraded oleuropein in olive fruit. *L. plantarum* initially hydrolyzes oleuropein through its β -glucosidase action to produce its aglycone form. In the second step, the esterase activity of *L. plantarum* gives rise to hydroxytyrosol and elenolic acid. β -Glucosidase activity is partially inhibited by glucose; however, esterase activity in the second step of biodegradation is not influenced by glucose [57]. *L. plantarum* 6907, *L. paracasei* 9192, *L. casei, Bif. lactis* BO, *Enterococcus faecium* 32, and *L. acidophilus* strain LAFTI®L10 were tested for their ability to convert oleuropein byproducts in olives and olive oil into hydroxytyrosol [53]. Of these strains, *L. plantarum* 6907, due to its auxotrophic characteristics, was the most

effective with a hydroxytyrosol yield of 30%.

Bioconversion of Isoflavones Using LAB

Characteristics, Function, and Synthesis of Isoflavones and Aglycone Isoflavones

Isoflavones are estrogen-like compounds found in legumes. Isoflavones have shown biological activities for the prevention and therapy of hormone-related diseases, cardiovascular diseases, breast, prostate, and colon cancers, and menopausal symptoms [58, 59]. Daidzein and genistein are the most widely studied isoflavones. Daidzein can be metabolized to produce equol ((3S)-3-(4-hydroxyphenyl)-7-chromanol) by intestinal bacteria in only 30–40% of the human population [58]. Equol has stronger estrogenic activity than daidzein and *O*-desmethylangolensin [59]. The beneficial effects of isoflavones depend on host health; therefore, bioconversion of isoflavones has been in steady demand (Table 3).

Bioconversion (substrate \rightarrow product)	LAB strains	Characteristics
Oleuropein \rightarrow hydroxytyrosol	Lactobacillus plantarum 6907 and Lactobacillus paracasei 9192	Hydrolysis reaction; comparison of aerobic and anaerobic conditions [53]
	Lactobacillus plantarum (B17, B20, and B21)	β-Glucosidase production; stable in pH 3.5 and 8% NaCl [55]
Oleuropein/5-bromo-4-chloro-3-indo- lyl β-D-glucuronide → hydroxytyrosol	Lactobacillus plantarum, Lactobacillus pentosus, Lactobacillus brevis, and Pediococcus pentosaceus	β-Glucosidase production [54]
Oleuropein → oleuropein-aglycone → hydroxytyrosol + elenolic acid	Lactobacillus plantarum B21	Table olive debittering; β -glucosidase and esterase production [56, 57]

Table 2. Bioconversion of oleuropein using LAB.

Bioconversion (substrate \rightarrow product)	LAB strains	Characteristics
Black soymilk (isoflavone glycosides daidzein and genistein)	Streptococcus thermophilus S10	Health functional fermented black soymilk having antioxidant activity [63]
Brain heart infusion medium with daidzein (daidzein → equol)	Bifidobacterium breve ATCC 15700 and Bifidobacterium longum BB536	Potential probiotic strains with function of equol [62]
Soymilk-tea (isoflavone glycoside → aglycone)	Streptococcus thermophilus ASCC 1275; Lactobacillus bulgaricus ASCC 859; Lactobacillus acidophilus CSCC 279	Probiotic characteristics and bioconversion rate [45]
Soya bean (isoflavone glycoside → aglycone)	Lactobacillus acidophilus B4496; Lactobacillus bulgaricus CFR2028; Lactobacillus casei B1922; Lactobacillus plantarum B4495; Lactobacillus fermentum B4655 with Saccharomyces boulardii	Fermentation of probiotic LAB with yeast; improvement of the bioavailability of isoflavone, mineral, and vitamin B complex [66]
Biotin-supplemented soymilk (isoflavone \rightarrow isoflavone aglycone)	Lactobacillus fermentum BT 8633	Ultrasound treatment; increment of permeabilization in the membrane of treated cells [65]
Soy milks (isoflavone → isoflavone aglycone (equol))	Mixed culture of <i>Lactobacillus plantarum</i> DPPMASL33; DPPMA 24W; <i>Lactobacillus rhamnosus</i> DPPMAAZ1; <i>Lactobacillus fermentum</i> DPPMA114	Manufacture of soy milk using commercial soy flour; immunomodulatory effect on intestinal human Caco-2/TC7 cells [64]

Table 3. Bioconversion of isoflavone glycosides using LAB.

Bioconversion of Isoflavones to Aglycone Forms

Equol producers among LAB include Lactococcus garvieae strain 20-92 [59, 60], Bif. animalis, Bif. longum-a, Bif. pseudolongum [61], Bif. breve 15700, Bif. longum BB536 [62], and Lactobacillus sp. Niu-O16 [60]. Soymilk fermented with L. plantarum TWK10 was found to improve nutritional and bioactive properties of foods [8]. β-Glucosidase increases the aglycone isoflavone content and suppresses tyrosinase activity and melanin production. Black soymilk fermented with S. thermophilus S10 converted daidzin and genistin to daidzein and genistein [63]. Total phenol contents, DPPH radical scavenging activity, and reducing power were increased during fermentation. A mixed culture of L. plantarum (DPPMA24W and DPPMASL33), L. fermentum DPPMA114, and L. rhamnosus DPPMAAZ1 was selected for its βglucosidase activity, and these strains were incubated in soymilk for 96 h. After fermentation, daidzein, genistein, glycitein, and equol were produced; anti-inflammatory effects of these compounds were observed on nitric oxide and IL-8 production in intestinal human Caco-2/TC7 cells [64]. Ultrasound of L. fermentum BT8633 affected β-glucosidase production and increased the bioconversion of isoflavones to aglycone forms in biotin-supplemented soymilk [65]; ultrasound may temporarily permeabilize the membrane of treated cells. L. acidophilus B4496, L. bulgaricus CFR2028, L. casei B1922, L. plantarum B4495, and L. fermentum B4655 were used with Saccharomyces boulardii to ferment soymilk to obtain the bioactive isoflavones genistein and daidzein [66].

Bioconversion of Other Phenolic Compounds

Characterization of Phenolic Compounds

Phenolic compounds are important constituents of food products of plant origin, and these compounds are directly related to the sensory characteristics of foods, such as flavor, stringency, and color. Phenolic compounds are beneficial to health because of their activities against carcinogenesis and mutagenesis, mainly through antioxidant effects [8, 67, 68]. Medicinal and tea plants reported to have functional components include *Magnolia*, *Cudrania tricuspidata* (Carr.) Bureau [69], and *Inula britannica* [70]. Various phenolic compounds from these plants can be converted to aglycone forms, which are more bioactive and have more potent antioxidant and anticancer activities, among other effects (Table 4).

Bioconversion of Phenolic Compounds to Aglycone Forms

A representative anthocyanin (malvidin-3-O-glucoside) was extracted from Pinot Noir grape skins and fermented with *L. plantarum* WCFS1 [68]. The main metabolites were gallic acid and protocatechuic acid, and increased antioxidant activity was observed. *Magnolia* flower petal extract was fermented by *Pediococcus acidilactici* KCCM 11614; fermentation increased the total phenolic contents, total flavonoids, and antioxidative and anticancer activities [67]. *C. tricuspidata* (Carr.) Bureau leaves were fermented with *Lactobacillus* derived from Korean soybean paste [68]; flavonoid glycosides were converted to flavonols, quercetin,

$Mechanisms^{a}$ (substrate \rightarrow product)	LAB strains	Characteristics
Green tea (EGCG, EGC, and EC \rightarrow GCG and GC)	Lactobacillus plantarum 62901 and Leuconostoc pseudomesenteroides K200132	Decrement of grass flavor and bitter taste [3]
Magnolia flower petal extract	Pediococcus acidilactici KCCM 11614	Increment of antioxidative and anticancer activities [67]
Pinot Noir (anthocyanin glycoside \rightarrow phenolic acid such as gallic acid and protocatechuic acid)	Lactobacillus plantarum WCFS1	Application in the human digestive system; antioxidant activity [68]
<i>Cudrania tricuspidata</i> leaves (flavonoid glycoside flavonols, quercetin, and kaempferol)	Lactobacillus plantarum SDL1413	Fermentation in 10% plant powder; antioxidant effect and identification through LC-MS [69]
Inula britannica extract	Lactobacillus acidophilus, Bifidobacterium longum, and Streptococcus thermophilus (ABTL)	Application in Cheddar-type cheese; antioxidant activity [70]

Table 4. Bioconversion of phenolic compounds using LAB.

^aEGCG, epigallocatechin gallate; EGC, epigallocatechin; EC, epicatechin; GCG, gallocatechin gallate; GC, gallocatechin.

and kaempferol. *S. thermophilus* ASCC 1275, *L. acidophilus* CSCC 2400, *L. paracasei* CSCC 279, and *L. delbrueckii* ssp. *bulgaricus* ATCC 859 converted isoflavone glycosides to aglycones in soymilk containing tea extract [45]. *L. acidophilus* CSCC 2400 showed the highest activity with a bioconversion ratio of 67–78%, resulting from the production of β-glucosidase and β-galactosidase. Extracts from green, oolong, and black teas influenced the cell population and production of bacterial enzymes in soymilk supplemented with them. After addition of *I. britannica* extract containing quercetin to yogurt strains (*S. thermophilus, L. acidophilus,* and *Bif. longum*), Cheddar-type cheese showed increment of total phenolic contents and antioxidant activity [70].

Microbial Fermentation of LAB for Bioconversion

Fermentation of LAB for bioconversion should involve an appropriate bacterial strain and fermentation medium. First, a bacterial strain for enzyme production using *p*-nitrophenyl-β-D-glucopyranoside (*p*NPG) substrate should be selected. These bacterial strains have been screened, mainly in fermented foods of plant origin, for safety and survival. L. plantarum is the most frequently used commercial starter in food fermentation [53]. Fermentation media should (i) include natural substances such as soymilk [8, 45, 63, 64, 66], olive [55], or Magnolia flower petal extract [67], (ii) contain added natural substrates in MRS medium [49, 51, 56], and (iii) contain added galactose [71], cellobiose [40], or ascorbic acid [72] in MRS medium for enzyme production. The strains Bif. longum, L. plantarum, L. pentosus, L. brevis, L. fermentum, and L. mesenteroides [5, 39, 50, 62, 71] have been used as fermentation starters to increase flavor and bioavailability; however, bacterial cell growth was

inhibited by hydroxytyrosol, oleuropein, and tyrosol, and vanillic, p-hydroxybenzoic, sinapic, syringic, protocatechuic, and cinnamic acids at high concentrations [72]. In addition, the concentration of glucose must be controlled in fermentation media to increase the production of glycoside hydrolase from probiotic bacteria. High concentrations of glucose induce increased bacterial cell growth but reduce enzyme induction. To avoid glucose catabolite repression, L. rhamnosus GG was cultured in MRS medium containing cellobiose and showed 25-fold higher β-glucosidase productivity [40]. Activities of α -L-arabinofuranosidase and α-L-arabinopyranosidase from Bif. longum RD47 were increased with the addition of 2% ascorbic acid [72]. Therefore, the concentration of these materials in the fermentation medium should be controlled to achieve high enzyme induction and avoid inhibition.

Conclusions and Perspectives

The use of LAB to enhance probiotic functions in industrial applications has increased. From a commercial perspective, fermented tea and yogurt are representative uses of LAB. In particular, plant-derived LAB are used as fermentation starters, as these strains show resistance to components such as flavonoids. Various natural substances have different bioactive properties because of their structures. In this regard, bioconversion of natural substances has gained industrial and scientific attention to reveal new functions or enhance nutraceutical properties. However, further improvement of bioconversion should be investigated to identify fermentation starters applicable to various substances and to optimize conditions, such as the ratio of substance to fermentation starter and incubation conditions. In addition, it may be possible to use these strains or enzymes to obtain high value-added compounds with antioxidant, estrogenic, and other effects.

Acknowledgments

This research was supported by the High Value-added Food Technology Development Program, Ministry of Agriculture, Food, and Rural Affairs (#314073-03) and the Priority Research Centers Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education, Science, and Technology (2009-0093824).

References

- Fortina MG, Ricci G, Foschino R, Picozzi C, Dolci P, Zeppa G, et al. 2007. Phenotyping, technological properties and safety aspects of *Lactococcus garvieae* strains from dairy environments. J. Appl. Microbiol. 103: 445-453.
- Lee NK, Han KJ, Son SH, Eom SJ, Lee SK, Paik HD. 2015. Multifunctional effect of probiotic *Lactococcus lactis* KC24 isolated from kimchi. *LWT Food Sci. Technol.* 64: 1036-1041.
- Park SB, Han BK, Oh HJ, Lee SJ, Cha SK, Park YS, Choi HJ. 2012. Bioconversion of green tea extract using lactic acid bacteria. *Food Eng. Prog.* 16: 26-32.
- 4. Gachon CMM, Langlois-Meurinne M, Saindrenan P. 2005. Plant secondary metabolism glycosyltransferases: the emerging functional analysis. *Trends Plant Sci.* **10**: 542-549.
- 5. Michlmayr H, Kneifel W. 2014. β-Glucosidase activities of lactic acid bacteria: mechanisms, impact on fermented food and human health. *FEMS Microbiol. Lett.* **352**: 1-10.
- 6. Rodríguez H, Curiel JA, Landete JM, de las Rivas B, de Felipe FL, Gómez-Cordovés C, *et al.* 2009. Food phenolics and lactic acid bacteria. *Int. J. Food Microbiol.* **132**: 79-90.
- Park MJ, General T, Lee SP. 2012. Physicochemical properties of roasted soybean flour bioconverted by solidstate fermentation using *Bacillus subtilis* and *Lactobacillus plantarum. Prev. Nutr. Food Sci.* 17: 36-45.
- Chen YM, Shin TW, Chiu CP, Pan TM, Tsai TY. 2013. Effects of lactic acid bacteria-fermented soy milk on melanogenesis in B16F0 melanocytes. *J. Funct. Foods* 5: 395-405.
- Berthiller F, Krska R, Domig KJ, Kneifel W, Juge N, Schuhmacher R, Adam G. 2011. Hydrolytic fate of deoxynivalenol-3-glucoside during digestion. *Toxicol. Lett.* 206: 264-267.
- Cairns JRK, Esen A. 2010. β-Glucosidases. *Cell. Mol. Life Sci.* 67: 3389-3405.
- Veena V, Poornima P, Parvatham R, Sivapriyadharsini Kalaiselvi K. 2011. Isolation and characterization of βglucosidase producing bacteria from different sources. *Afr. J. Biotechnol.* 10: 14907-14912.
- 12. Spano G, Rinaldi A, Ugliano M, Beneduce L, Massa S. 2005.

A β-glucosidase producing gene isolated from wine *Lactobacillus plantarum* is regulated by abiotic stresses. *J. Appl. Microbiol.* **98:** 855-861.

- Grandits M, Michlmayr H, Sygmund C, Oostenbrink C. 2013. Calculation of substrate binding affinities for a bacterial GH78 rhamnosidase through molecular dynamics simulations. J. Mol. Catal. B Enzym. 92: 34-43.
- 14. Jee HS, Chang KH, Park SH, Kim KT, Paik HD. 2014. Morphological characterization, chemical components, and biofunctional activities of *Panax ginseng*, *Panax quinquefolium*, and *Panax notoginseng* roots: a comparative study. *Food Rev. Int.* **30**: 91-111.
- Ligor T, Ludwiczuk A, Wolski T, Buszewski B. 2005. Isolation and determination of ginsenosides in American ginseng leaves and root extracts by LC-MS. *Anal. Bioanal. Chem.* 383: 1098-1105.
- Attele AS, Wu JA, Yuan CS. 1999. Ginseng pharmacology: multiple constituents and multiple actions. *Biochem. Pharmacol.* 58: 1685-1693.
- Chang KH, Jee HS, Lee NK, Parik SH, Lee NW, Paik HD. 2009. Optimization of the enzymatic production of 20(S)ginsenoside Rg3 from white ginseng extract using response surface methodology. *New Biotechnol.* 26: 181-186.
- Yu T, Yang Y, Kwak YS, Song GG, Kim MY, Rhee MH, Cho JY. 2017. Ginsenoside Rc from *Panax ginseng* exerts antiinflammatory activity by targeting TANK-binding kinase 1/ interferon regulatory factor-3 and p38/ATF-2. *J. Ginseng Res.* [In Press].
- 19. Hou YL, Tsai YH, Lin YH, Chao JCJ. 2014. Ginseng extract and ginsenoside Rb1 attenuate carbon tetrachloride-induced liver fibrosis in rats. *BMC Complement. Altern. Med.* **14:** 415.
- Yoo YC, Lee J, Park SR, Nam KY, Cho YH, Choi JE. 2013. Protective effect of ginsenoside-Rb2 from Korean red ginseng on the lethal infection of haemagglutinating virus of Japan in mice. J. Ginseng Res. 37: 80-86.
- Yang JW, Kim SS. 2015. Ginsenoside Rc promotes antiadipogenic activity on 3T3-L1 adipocytes by down-regulating C/EBPα and PPARAγ. *Molecules* 20: 1293-1303.
- 22. Wang L, Liu QM, Sung BH, An DS, Lee HG, Kim SG, *et al.* 2011. Bioconversion of ginsenosides Rb(1), Rb(2), Rc and Rd by novel β-glucosidase hydrolyzing outer 3-O glycoside from *Sphingomonas* sp. 2F2: cloning, expression, and enzyme characterization. *J. Biotechnol.* **156**: 125-133.
- 23. Kim HJ, Kim P, Shin CY. 2013. A comprehensive review of the therapeutic and pharmacological effects of ginseng and ginsenosides in central nervous system. J. Ginseng Res. 37: 8-29.
- 24. Jovanovski E, Bateman EA, Bhardwaj J, Fairgrieve C, Mucalo I, Jenkins AL, et al. 2014. Effect of Rg3-enriched Korean red ginseng (*Panax ginseng*) on arterial stiffness and blood pressure in healthy individuals: a randomized controlled trial. J. Am. Soc. Hypertens. 8: 537-541.
- 25. Li N, Liu Y, Li W, Zhou L, Li Q, Wang X, *et al.* 2015. A UPLC/MS-based metabolomics investigation of the protective

effect of ginsenosides Rg1 and Rg2 in mice with Alzheimer's disease. *J. Ginseng Res.* **40:** 9-17.

- 26. Jia P, Chen G, Li R, Rong X, Zhou G, Zhong Y. 2013. Ginsenoside retinoblastoma 1 (Rb1) suppresses NO production and inducible nitric oxide synthase (iNOS) expression by inhibiting nuclear factor κB (NF-κB) activation in SW1353 chondrosarcoma cells. *Afr. J. Pharm. Pharmacol.* 7: 2584-2590.
- Park EK, Choo MK, Han MJ, Kim DH. 2004. Ginsenoside Rh1 possesses antiallergic and anti-inflammatory activities. *Int. Arch. Allergy Immunol.* 133: 113-120.
- Kim HS, Lee EH, Ko SR, Choi KJ, Park JH, Im DS. 2004. Effects of ginsenosides Rg3 and Rh2 on the proliferation of prostate cancer cells. *Arch. Pharm. Res.* 27: 429-435.
- Park YC, Lee CH, Kang HS, Kim KW, Chung HT, Kim HD. 1996. Ginsenoside-Rh1 and Rh2 inhibit the induction of nitric oxide synthesis in murine peritoneal macrophages. *Biochem. Mol. Biol. Int.* 40: 751-757.
- Lai DM, Tu YK, Liu IM, Chen PF, Cheng JT. 2006. Mediation of beta-endorphin by ginsenoside Rh2 to lower plasma glucose in streptozotocin-induced diabetic rats. *Planta Med.* 72: 9-13.
- Lee BH, You HJ, Park MS, Kwon B, Ji GE. 2006. Transformation of glycosides from food materials by probiotics and food microorganisms. J. Microbiol. Biotechnol. 16: 497-504.
- Bae EA, Park SY, Kim DH. 2000. Constitutive β-glucosidases hydrolyzing ginsenoside Rb1 and Rb2 from human intestinal bacteria. *Biol. Pharm. Bull.* 23: 1481-1485.
- 33. Bae EA, Kim NY, Han MJ, Choo MK, Kim DH. 2003. Transformation of ginsenosides to compound K (IH-901) by lactic acid bacteria. *J. Microbiol. Biotechnol.* **13**: 9-14.
- Huq MA, Kim YJ, Min JW, Bae KS, Yang DC. 2014. Use of Lactobacillus rossiae DC05 for bioconversion of the major ginsenosides Rb1 and Re into the pharmacologically active ginsenosides C-K and Rg2. Food Sci. Biotechnol. 23: 1561-1567.
- Quan LH, Plao JY, Min JW, Yang DU, Lee HN, Yang DC. 2011. Bioconversion of ginsenoside Rb1 into compound by *Leuconostoc citreum* LH1 isolated from kimchi. *Braz. J. Microbiol.* 42: 1227-1237.
- Hyun YJ. 2012. Cloning and characterization of ginsenoside Ra1-hydrolyszing β-D-xylosidase from *Bifidobacterium breve* K-110. J. Microbiol. Biotechnol. 22: 535-540.
- Lee J, Hyun YJ, Kim DH. 2011. Cloning and characterization of α-L-arabinofuranosidase and bifunctional α-L-arabinopyranosidase/β-D-galactopyranosidase from *Bifidobacterium longum* H-1. J. Appl. Microbiol. 111: 1097-1107.
- Lee SJ, Kim Y, Kim MG. 2015. Changes in the ginsenoside content during the fermentation process using microbial strains. J. Ginseng Res. 39: 392-397.
- Jung J, Paik HD, Yoon HJ, Jang HJ, Jeewanthi CRK, Jee HS, et al. 2016. Physicochemical characteristics and antioxidant capacity in yogurt fortified with red ginseng extract. Korean J. Food Sci. Anim. Resour. 36: 412-420.
- 40. Ku S, You HJ, Park MS, Ji GE. 2016. Whole-cell biocatalysis for producing ginsenoside Rd from Rb1 using *Lactobacillus*

rhamnosus GG. J. Microbiol. Biotechnol. 26: 1206-1215.

- Liu C, Tung YT, Wu CL, Lee BH, Hsu WH, Pan TM. 2011. Antihypertensive effects of *Lactobacillus*-fermented milk orally administered to spontaneously hypertensive rats. *J. Agric. Food Chem.* 59: 4537-4543.
- 42. Kim JE, Kim JS, Song YC, Lee J, Lee SP. 2014. Novel bioconversion of sodium glutamate to γ-poly-glutamic acid and γ-amino-butyric acid in a mixed fermentation using *Bacillus subtilis* HA and *Lactobacillus plantarum* K154. *Food Sci. Biotechnol.* 23: 1551-1559.
- Ogunleye A, Bhat A, Irorere VU, Hill D, Williams C, Radecka I. 2015. Poly-γ-glutamic acid: production, properties and applications. *Microbiology* **161**: 1-17
- 44. Yoshimura M, Toyoshi T, Sano A, Izumi T, Fujii T, Konishi S, *et al.* 2010. Antihypertensive effect of a gamma-aminobutyric acid rich tomato cultivar 'DG03-9' in spontaneously hypertensive rats. *J. Agric. Food Chem.* **58**: 615-619.
- 45. Zhao D, Shah NP. 2014. Effect of tea extract on lactic acid bacterial growth, their cell surface characteristics and isoflavone bioconversion during soymilk fermentation. *Food Res. Int.* **62**: 877-885.
- 46. Rizzello CG, Cassone A, Di Cagno R, Gobbetti M. 2008. Synthesis of angiotensin I-converting enzyme (ACE)-inhibitory peptides and γ-aminobutyric acid (GABA) during sourdough fermentation by selected lactic acid bacteria. *J. Agric. Food Chem.* **56**: 6936-6943.
- 47. Sun TS, Zhao SP, Wang HK, Cai CK, Chen YF, Zhang HP. 2009. ACE-inhibitory activity and gamma-aminobutyric acid content of fermented skim milk by *Lactobacillus helveticus* isolated from Xingjiang koumiss in China. *Eur. Food Res. Technol.* 228: 607-612.
- 48. Nejati F, Rizzello CG, Di Cagno R, Sheikh-Zeinoddin M, Diviccaro A, Minervini G, *et al.* 2013. Manufacture of a functional fermented milk enriched of angiotensin-I converting enzyme (ACE)-inhibitory peptides and gamma-amino butyric acid (GABA). *LWT-Food Sci. Technol.* 51: 183-189.
- Zhang Y, Song L, Gao Q, Yu SM, Li L, Gao NF. 2012. The two-step biotransformation of monosodium glutamate to GABA by *Lactobacillus brevis* growing and resting cells. *Appl. Microbiol. Biotechnol.* 94: 1619-1627.
- 50. Di Cagno R, Mazzacane F, Rizzello GG, Angelis MD, Gluliani GT, Meloni M, et al. 2010. Synthesis of gammaaminobutyric acid by *Lactobacillus plantarum* DSM19463: functional grape must beverage and dermatological applications. *Appl. Microbiol. Biotechnol.* 86: 731-741.
- Wu Q, Shah NP. 2015. Gas release-based prescreening combined with reversed-phase HPLC quantitation for efficient selection of high γ-aminobutyric acid (GABA)-producing lactic acid bacteria. J. Dairy Sci. 98: 790-797.
- 52. Omar SH. 2010. Oleuropein in olive and its pharmacological effects. *Sci. Pharm.* **78:** 133-154.
- Santos MM, Piccirillo C, Castro PLM, Kalogerakis N, Pintado ME. 2012. Bioconversion of oleuropein to hydroxytyrosol by

lactic acid bacteria. World J. Microbiol. Biotechnol. 28: 2435-2440.

- 54. Ghabbour N, Lamzira Z, Thonart P, Cidalia P, Markaouid M, Asehraoua A. 2011. Selection of oleuropein-degrading lactic acid bacteria strains isolated from fermenting Moroccan green olives. *Grasas Aceites* **62**: 84-89.
- 55. Ciafardini G, Marsilio V, Lanza B, Pozzi N. 1994. Hydrolysis of oleuropein by *Lactobacillus plantarum* strains associated with olive fermentation. *Appl. Environ. Microbiol.* **60**: 4142-4147.
- Marsilio V, Lanza B, Pozzi N. 1996. Progress in table olive debittering: degradation in vitro of oleuropein and its derivatives by *Lactobacillus plantarum*. J. Am. Oil Chem. Soc. 93: 593-597.
- Marsilio V, Lanza B. 1998. Characterization of an oleuropein degrading strain of *Lactobacillus plantarum*. Combined effects of compounds present in olive fermenting brines (phenols, glucose and NaCl) on bacterial activity. *J. Sci. Food Agric*. 96: 520-524.
- 58. Atkinson C, Frankenfeld CL, Lampe JW. 2005. Gut bacterial metabolism of the soy isoflavone daidzein: exploring the relevance to human health. *Exp. Biol. Med.* **230**: 155-170.
- 59. Uchiyama S, Ueno T, Suzuki T. 2007. Identification of a newly isolated equol-producing lactic acid bacterium from the human feces. *J. Intest. Microbiol.* **21:** 217-220.
- 60. Shimada Y, Yasuda S, Takahashi M, Hayashi T, Miyazawa N, Sato I. 2010. Cloning and expression of a novel NADP (H)dependent daidzein reductase, an enzyme involved in the metabolism of daidzein, from equol-producing *Lactococcus* strain 20–92. *Appl. Environ. Microbiol.* **76**: 5892-5901.
- Tsangalis D, Ashton JF, McGill AEJ, Shah NP. 2002. Enzymic transformation of isoflavone phytoestrogens in soy milk by beta glucosidase producing bacteria. *J. Food Sci.* 67: 3104-3113.
- Elghali S, Mustafa S, Amid M, Manap MYA, Ismali A, Abas F. 2012. Bioconversion of daidzein to equol by *Bifidobacterium breve* 15700 and *Bifidobacterium longum* BB536. *J. Funct. Foods* 4: 736-745.
- 63. Lee M, Hong GE, Zhang H, Yang CY, Han KH, Mandal PK, *et al.* 2015. Production of the isoflavone aglycone and antioxidant activities in black soymilk using fermentation with *Streptococcus thermophilus* S10. *Food Sci. Biotechnol.* 24: 537-544.
- 64. Di Cagno R, Mazzacane F, Rizzello CG, Vincentini O, Silano M, Giuliani G, et al. 2010. Synthesis of isoflavone aglycones and equol in soy milks fermented by food-related lactic acid bacteria and their effect on human intestinal Caco-2 cells. J. Agric. Food Chem. 58: 10338-10346.
- 65. Ewe JA, Wan-Abdullah WN, Alias AK, Ling MZ. 2012. Effects of ultrasound on growth, bioconversion of isoflavones and probiotic properties of parent and subsequent passages of *Lactobacillus fermentum* BT 8633 in biotin-supplemented soymilk. *Ultrason. Sonochem.* 19: 890-900.
- 66. Rekha CR, Vijayalakshmi G. 2010. Bioconversion of

isoflavone glycosides to aglycones, mineral bioavailability and vitamin B complex in fermented soymilk by probiotic bacteria and yeast. J. Appl. Microbiol. **109**: 1198-1208.

- Park EH, Kim HS, Eom SJ, Kim KT, Paik HD. 2015. Antioxidative and anticanceric activities of *Magnolia (Magnolia denudata)* flower petal extract fermented by *Pediococcus acidilactici* KCCM 11614. *Molecules* 20: 12154-12165.
- Suthanthangjai W, Kilmartin PA, Phillips ARJ, Davies K, Ansell J. 2014. Bioconversion of Pinot Noir anthocyanins into bioactive phenolic compounds by lactic acid bacteria. *Nutr. Aging* 2: 145-149.
- Lee Y, Oh J, Jeong YS. 2015. Lactobacillus plantarum-mediated conversion of flavonoid glycosides into flavonols, quercetin, and kaempferol in Cudrania tricuspidata leaves. Food Sci. Biotechnol. 24: 1817-1821.
- Lee NK, Jeewanthi RK, Park EH, Paik HD. 2016. Physicochemical and antioxidant properties of Cheddar-type cheese fortified with *Inula britannica* extract. *J. Dairy Sci.* 99: 83-88.
- Landete JM, Curiel JA, Rodríguez H, de las Rivas B, Muňoz R. 2008. Study of the inhibitory activity of phenolic compounds found in olive products and their degradation by *Lactobacillus plantarum* strains. *Food Chem.* **107**: 320-326.
- 72. Ku S, You HJ, Park MS, Ji GE. 2015. Effects of ascorbic acid on α-L-arabinofuranosidase and α-L-arabinopyranosidase activities from *Bifidobacterium longum* RD47 and its application to whole cell bioconversion of ginsenoside. *J. Korean Soc. Appl. Biol. Chem.* 58: 857-865.
- 73. Quan LH, Cheng LQ, Kim HB, Kim JH, Son NR, Kim SY, et al. 2010. Bioconversion of ginsenoside Rd into compound K by Lactobacillus pentosus DC101 isolated from kimchi. J. Ginseng Res. 34: 288-295.
- 74. Chi H, Kim DH, Ji GE. 2005. Transformation of ginsenosides Rb2 and Rc from *Panax ginseng* by food microbial enzyme. *Biol. Pharm. Bull.* 28: 2102-2105.
- Chi H, Ji GE. 2005. Transformation of ginsenosides Rb1 and Re from *Panax ginseng* by food microorganisms. *Biotechnol. Lett.* 27: 765-771.
- Chi H, Lee BH, You HJ, Park MS, Ji GE. 2006. Different transformation of ginsenosides from *Panax ginseng* by lactic acid bacteria. J. Microbiol. Biotechnol. 16: 1629-1633.
- Li HZ, Gao DD, Cao YS, Xu HY. 2008. A high gammaaminobutyric acid-producing *Lactobacillus brevis* isolated from Chinese traditional paocai. *Ann. Microbiol.* 58: 649-653.
- Binh TTT, Ju WT, Jung WJ, Park RD. 2014. Optimization of gamma-amino butyric acid production in a newly isolated *Lactobacillus brevis*. *Biotechnol. Lett.* 36: 93-98.
- Barrett E, Ross RP, O'Toole PWO, Fitzgerald GF, Stanton C. 2012. Gamma-aminobutyric acid production by culturable bacteria from the human intestine. *J. Appl. Microbiol.* 113: 411-417.