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Synthesis and Biological Characteristics of Low-calorie Omija Juice

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Synthesis and Biological Characteristics of Low-calorie Omija Juice

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Abstract

Omija whose binomial name is *Schisandra chinensis Turcz. (Baill.)* is a medicinal plant that contains many bioactive chemicals called dibenzocyclooctadiene liganans. Schisandrin, Gomisin N, Schizandrin C, Schisandrin A, Gomisins A and G are its major dibenzocyclooctadiene lignans. Using theses rare valuable compounds from omija with sucrose, omija juice was made by two glucansucrases from *Leuconostoc mesenteroides* B-512FMCM and B-1355CF10. Acceptor reaction occurred by dextransucrase and alternansucrase from respective *Leuconostoc mesenteroides* until all of sucrose was consumed. More than nine kinds of oligosaccharides which have α-(1,6) linkages as backbones and α-(1,2), α-(1,3), α-(1,4) linkages as branches were synthesized in Omija juice.

Omija oligosaccharides have lowered calories by 61.13% comparing to regular omija juice which have 50% sucrose after treatment with intestinal enzymes.

Newly synthesized omija oligosaccharides reduced formation of the insoluble glucan, mutan which is one of products that causes dental plaque by 96%.

Omija oligosaccharides was shown to have melanogenesis inhibition activity.

This paper revealed omija oligosaccharides have potential for medicinal and cosmetic properties in biological system.

Keywords: Omija; Dibenzocyclooctadiene liganan; *Leuconostoc mesenteroides*; Less-digestible oligosaccharides; glucansucrase; Mutansucrase inhibition; Bioavailability.

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Introduction

1. Omija Juice

Traditionally Korean people used to make fruit juice extract using sucrose at the same weight ratio of fruit (이혜정, 오재복, & 배숙희, 2005). So much sucrose consumption makes people gain weight and earn high cholesterol concentration in blood (Rippe & Angelopoulos, 2016). Also, it could lead to form dental plaque and caries (Cury, Rebelo, Del Bel Cury, Derbyshire, & Tabchoury, 2000). Therefore, there is need to make omija juice in more healthful way to lower calorie and prevent from forming plaque.

1.1. Omija

Omija whose binomial name is *Schisandra chinensis Turcz. (Baill.)* is a well-known medicinal plant not only in Korea and China but also in Russia for a long time (Bensky and Gamble 1993). However, only since 2007, WHO recognized this fruit to be pharmaceutical plant officially in monograph (World Health Organization, 2007). The *S. chinensis* contains many bioactive chemicals

called dibenzocyclooctadiene liganans. Schisandrin, Gomisin N, Schiznadrin C, Schisandrin A, Gomisin A, and Gomisin G are the major constituents of these lignans (Szopa, Ekiert, & Ekiert, 2017). Their biological activities are reported in scientific studies. Hepatoprotective activity, anti-inflammatory, antioxidative, detoxification activities, anticancer activity, immunostimulant activity, influence on the central nervous and respiratory system, adaptogenic and ergogenic activities, anti-obesity activities, and antiviral and antibacterial activity are their known beneficial effects to apply to pharmacology (Szopa et al., 2017).

2. *Leuconostoc mesenteroides* B-512FMCM & B-1355CF10

Leuconostoc mesenteroides produced dextran which is composed of D-glucose units united by α-(1,6) linkages as a backbone and branched by α-(1,2), α-(1,3), α-(1,4) linkages synthesized from sucrose (Robyt, 1996).

Leuconostoc and *Streptococcus* are the main genera of bacteria that make enzymes to synthesize dextrans. The *Leuconostoc* species require sucrose as an inducer in the culture to express the enzyme called glucansucrase whereas the *Streptococcus* elaborate constitutive

glucansucrase. However, *L. mesenteroides* B-512FMCM and 1355CF10, the mutants from strains, B512F(M) and B1355 are constitutive for glucansucrase production (D. Kim & Robyt, 1994).

L. mesenteroides B-512 elaborates a single dextransucrase that is widely used in food industry and medical field and makes various kinds of oligosaccharides (Demuth, Jördening, & Buchholz, 2002).

L. mesenteroides B-1355 produces two kinds of glucansucrase: one is a dextran and the other is alternan. Alternan is a α-(1,6), α-(1,3) linked glucan alternatively as a main chain that has resistance to be hydrolyzed (D. Kim & Robyt, 1994).

One of major problems to purify *Leuconostoc* glucansucrase is due to the sucrose inducer that is required to make glucans in the culture supernatant. Consequently, the enzyme is in the culture supernatant tangled with glucan. As another problem, the product, glucan makes culture supernatant viscous. Dextranase treatment is indispensable purification step following these reactions.

By chemical mutation process using ethyl methane sulfonate (EMS), *Leuconostoc mesenteroides* B-512FMCM and B-1355CF10 are selected as constitutive mutants to solve these problems (D. Kim & Robyt, 1994).

3. Oligosaccharides

By using glucansucrase from *L. mesenteroides* B-512FMCM and 1355CF10, oligosaccharides are synthesized. Glucansucrase hydrolyzes sucrose to glucose and fructose, and then transfers the glucose to the acceptor to make oligosaccharides (Robyt & Eklund, 1983).

In general, oligosaccharides are called carbohydrates that have a degree of polymerization (DP) from 2 to 10 monosaccharide units (molecular weight of 300–2000). Oligosaccharides are used in foods, feeds, pharmaceuticals, or cosmetics as stabilizers, or prebiotic compounds. Commercially produced oligosaccharides are fructooligosaccharides, isomaltooligosaccharides, maltooligosaccharides and galactooligosaccharides. Among them, it is well known that maltooligosaccharides and isomaltooligosaccharides are acid- and heat-stable carbohydrates and less digestible ones by human gastric enzymes (Seo et al., 2007). They are less sweet than fructooligosaccharides, susceptible to acid and heat treatment. As a result, both types have restricted use as additives or sweeteners in foods that require heat treatment or acidic pH during process.

Many sweet foods contain mono- or disaccharides that are readily metabolized by cariogenic bacteria. To satisfy human craving for sweet substances without causing caries, the use of non-metabolizable dietary sweeteners has been proposed (M. Kim, Day, & Kim, 2010).

Oligosaccharides could be compounds that are safe, cheap and effective in blocking oral colonization of *Streptococcus* species.

3.1. Bioavailability of non-digestible Oligosaccharides

Oligosaccharides have been reported that they could promote the absorption of polyphenols (Lu, Lin, Li, & Yang, 2017; Shinoki et al., 2013). However, these mechanisms were not fully elucidated. Many phenomenon showed that oligosaccharides enhanced bioavailability of other compounds such as flavonoid or isoflavone (Matsukawa, Matsumoto, Chiji, & Hara, 2009). In previous studies, co-administration oligosaccharides with polyphenols reduced the expression of polyphenol efflux transporters in small intestine (Hollman, 2004). In this study, samples with increased bioavailability of dibezocyclooctadiene by using oligosaccharides were analyzed.

4. *Staphylococcus mutans,* its formation of plaque and dental caries

Three steps are processed to form dental plaque (Forssten, Björklund, & Ouwehand, 2010). First, saliva is absorbed to the enamel at the moment teeth have been cleaned. Therefore the enamel is coated with a complex of glycoproteins, proteins, and mucins. Secondly, bacteria interact with the formed pellicle by cell-to-surface. The biofilm forms mainly from *Streptococcus sanguis* and *Actinomyces viscosus*. In the third step, *Streptococcus mutans* adhere to the primary colonizers by cell-to-cell interactions. Subsequently growth of these bacteria results in biofilm formation onto the teeth, also referred to as dental plaque. Increasingly, more carbohydrates, especially sucrose, concentration makes acidic products in oral environment which leads to dental caries. *S. mutans* could make extracellular polsaccharides (EPS) using sucrose as a substrate and mutansucrase as an enzyme, releasing fructose and glucose. The EPS are long-chained and high molecular mass polymer that are important factor of *S. mutans* cariogenicity. The polymer, glucan is synthesized by glucosyltransferase (GTF) while fructans are produced by frutosyltransferase (FTF). Mutan is one of the glucan. They are composed of α-(1,6) linked glucose units as backbones and α-(1,3) linked glucose units as branches.

5. Melanogenesis inhibition assay using Zebrafish larvae

Melanin is a pigment existed in most animal organs such as skin, hair, eyes, and brain (Simon et al., 2002). Melanogenesis is a complicated process where diverse signaling pathways are necessary. Among them, the melanocortin 1 receptor (MC1R) is an important regulator in melanogenesis, signaling through its ligands including melanocyte-stimulating hormone (MSH) and adrenocorticotropic hormone (ACTH) (Slominski, Tobin, Shibahara, & Wortsman, 2004). Skin melanin is formed by melanocytes in the epidermis and then transferred to keratinocytes, where they play key roles in skin protection by absorbing UV radiation from sunlight and scavenging reactive free radicals. L-tyrosine and L-dihydroxyphenylalanine (L-DOPA), major substrates of melanogenic enzymes, also function as hormone-like regulators in melanogenesis (Andrzej, A., & John, 2012). Previously, Gomisin N of *Schizandrin* lignans was reported to inhibit melanin biosynthesis (Chae et al., 2017). Based on this previous research, experiment on the increase in bioavailability due to oligosaccharides needs to be carried out by Omija oligosaccharides comparing to only omija sample.

6. Research purpose of Omija oligosaccharides

To make healthful omija juice, low-calorie omija oligosaccharides were synthesized by *Leuconostoc mesenteroides* dextransucrase acceptor reaction. In previous study, oligosaccharides could increase bioavailabiliy of polyphenols such as flavonoid. Also, omija was reported to effect on melanogenesis inhibition and prevention of forming plaque, and anti-inflammatory effect. In this paper, we carried out these experiments to figure out synergy effect between omija extract and oligosaccharides as well as improved bioavailability of *Schizandra* lignan due to oligosaccharides.

Materials and Methods

1. Preparation of culture media

For cultivating *Leuconostoc mesenteroides*, LWG media were used. LWG media was composed of 0.5% (w/v) yeast extract, 0.5% (w/v) peptone, 2% (w/v) K_2HPO_4, 2% (w/v) glucose, 0.02% (w/v) $MgSO_4 \cdot 7H_2O$, 0.001% (w/v) NaCl, 0.001% (w/v) $FeSO_4 \cdot 7H_2O$, 0.0013% (w/v) $MnSO_4 \cdot H_2O$, and 0.0013% (w/v) $CaCl_2 \cdot H_2O$.

Three groups were sterilized respectively at 121℃ for 15 min. One group is yeast extract, peptone, and glucose. Another group is K_2HPO_4, and the other group is mineral solution. The components were mixed at 60℃ after sterilization. LWS media was the same as LWG except that They substituted 2% (w/v) glucose with 2% (w/v) sucrose.

2. Enzyme preparation from the B-512FMCM & B-1355CF10 strain

Dextransucrase (Enz_{512} & Enz_{1355}) was prepared by culturing the *Leuconostoc mesenteroides* B-512FMCM and 1355CF10 strain in

LWG media at 28°C and purified as described previously (Nguyen et al., 2014).

3. Fermentation of the B-512FMCM & B-1355CF10 strain

14L broth fermentation was carried out in 19L volumetric fermenter (NLF 19L, Bioengineering, Switzerland). 2% (v/v) of seed were added and agitated at 150 rpm, 0.5 bar/min aeration (measured at 15°C) at 28°C. To verify glucose consumption and enzyme activity, samples were collected every 2 h. Also, optical density (OD) was measured at 600 nm wave length to figure out the growth degree of microbes. Fermentation was suspended when glucose consumption was almost completed. The samples were centrifuged with SUPRA 25K centrifugal separator.

4. Concentration of enzyme from the B-512FMCM & B-1355CF10 strain

The concentration of broth supernatant was injected into polyethersulfone hollow fiber (Millipore, Bedford, USA) which has 100 kDa pore size at 4°C. The injection pump (LongerPump, BT300-2J, China) was used setting 167 mL/min (100 rpm) flow rate. Finally, supernatant was collected and lyophilized.

5. Preparation of omija oligosaccharides sample

40 brix of Omija extract (Jin Seong FM food company) was used and 10% Calcium hydroxide in 25% sucrose solution was dropped to Omija extract to adjust to be above pH 3.8. The optimal pH of dextransucrase from *Leuconostoc mesenteroides*is is pH 5.2. However, browning effect of omija occurs when it is astray from acidic condition. Sucrose was added to omija extract to make it 50% (w/v) sucrose as omija juice. 512FMCM dextransucrase is 0.3 unit/mg. B-1355 CF10 dextransucrase is 0.03 unit/mg. In this reaction condition, final concentration of these enzymes was 10 unit/ml and 1 unit/ml respectively. The reaction time was determined at the time that sucrose was totally consumed. Omija oligosaccharides were synthesized in shaking incubator at 37°C in 100 rpm.

Negative controls are omija and less-digestible oligosaccharides. Omija was also adjusted to the same pH condition as omija oligosaccharides using Calcium hydroxide solution. Less-digestible oligosaccharides were

made in the same manner as described above except that sodium acetate was used to reach optimal pH 5.2.

6. Removement of monosaccharides using yeast beads and large-scale production of omija oligosaccharides, omija, and less-digestible oligosaccharides

To get large amount of omija oligosaccharides, omija, and less-digestible oligosaccharides, reaction volume of each sample was up to 100 mL. To get higher purity of the less-digestible oligosaccharides, yeast beads were treated in reaction sample. Methods was applied the described in *Yoon et al* (Yoon, Mukerjea, & Robyt, 2003). 5g of alginic acid sodium salt (from brown algae, low viscosity, Sigma Co.) was dissolved in 200 mL of hot water with vigorous stirring and kept in the ice for 30 min. Meanwhile, 2 L of 4% (w/v) calcium chloride dihydrate (Duksan, Korea) solution was prepared. 4g of yeast powder (*Saccharomyce scerevisiae* 98.5%, Saf Instant Yeast Red, Societe Industrielle Lesaffre, France) was mixed in 200 mL of 2.5% (w/v) sodium alginate solution. This yeast dispersion was degassed for 20 min. Using injection pump (Longer Pump, BT300-2J), alginate beads trapped by the yeast powder were dropped in calcium chloride solution with stirring in the ice. After washing beads with pure water, the wet

beads were kept in 4℃ for 3 h to be harden. These beads were added adequately to 100 mL of the each sample. After incubation at 37°C, 150 rpm for 4 days, consumption of monosaccharides was verified by TLC analysis. The TLC plate was twice developed with nitromethane:propanol:water (2:5:1.5, v:v:v) solvent. Finally, these samples were lyophilized in order to do further studies.

7. HPLC Analysis of the Less-digestible oligosaccharides

Omija oligosaccharides were prepared with dilution using 100% Ethanol by 4-fold times and filtered using a 0.2 μm membrane syringe filter (Santorious AG, Germany). It was injected into HPLC analysis system. Developing solvents were 100% water (solvent A) and 100% acetonitrile (solvent B). The following elution gradient was applied: 0-60 min, 40% A; 60-65 min, 40-25% A; 65-75 min, 25% A; 75-80 min, 25-40% A; 80-90 min, 40% A. YMC polyamine ii column was used. Waters 2424 ELSD was used as a detector.

8. Prevention of insoluble glucan synthesis by *Streptococcus mutans* mutansucrase

Mutansucrase was prepared by culturing *Streptococcus mutans* in brain heart infusion (BHI) broth containing 2% sucrose as seed culture and then subculturing them in the BHI broth containing 0.5% (w/v) glucose as main culture at 37°C in shaking incubator at 150 rpm for 8 h, as described by *Ryu et al* (Su-Jin et al., 2000). After fermentation in 7 L at three times, the cells were separated from the broth by centrifugation at 6,000×g for 20 min. The culture concentrated to 300 mL using hollow fiber membrane (30 K cut-off, Millipore, USA) with 20 mM sodium phosphate buffer (pH 6.8). The enzyme was pooled and stored at −20°C until further study. One unit of mutansucrase activity was defined as the amount of enzyme that liberates 1 μM of fructose per min at 37°C, pH 6.8. 6.25% to 50% (v/v) of the original or modified concentrated juice samples was then mixed with 0.1 U/mL mutansucrase in sodium phosphate buffer.

There were no inhibitor in the positive control. In negative control, only enzyme and any inhibitor (sample) was not added. In each positive sample, 100 mM of sucrose, 20 mM of sodium phosphate, enzyme, and each sample were mixed. In each negative sample, only sucrose was not added. After incubation at 37°C for 12 h, the water-insoluble glucan synthesized in each mutansucrase reaction was collected by centrifugation, redissolved in 1 M NaOH, and spotted on a silica gel 60F254 TLC plate. The amount of insoluble glucan was quantitatively determined on a TLC plate using the AlphaEaseFC 4.0 Image Program.

9. Caloric analysis

Total non-digestible oligosaccharide concentration in the omija oligosaccharides sample was determined using the enzymatic gravimetric method of AOAC Methods 2009.01.

Enzymatic hydrolysis of sucrose and modified oligosaccharides was carried out with three kinds of enzymes: 7.5 U/mL α-amylase (Megazyme) in pH 6.9 at 37°C for 16 h for hydrolysis of the digestible oligosaccharides, 33 U/mL amyloglucosidase (Megazyme) in pH 4.8 at 60°C for 45 min, and 500 U/ml invertase (sigma) at 50°C for 3 hours for hydrolysis of the sucrose. The amounts of glucose and fructose produced in the reaction mixtures were determined using a K-FRUGL kit.

10. Melanogenesis inhibition assay using Zebrafish larvae

In 24 well plate, 5~10 larvae per well were treated with each sample: omija, omija oligosaccharide, and oligosaccharide. 0.1% (v/v) to 0.4% (v/v) of each sample as final concentration were applied to each well. The time after 38 hours post fertilization is proper time to check if melanogenesis occurs fully. At 48 hours post fertilization, zebra

larvae was checked on skin condition in this study. Positive control is 20 μM of PTU (phenylthiourea) treated zebrafish. Negative control is base medium as 0.2 mM of egg water.

Result

1. High-performance liquid chromatography analysis of oligosaccharides

Omija oligosaccharides were analyzed by HPLC using YMC polyamine ii column and detected by Evaporative Light Scattering detector (Waters 2424, USA). The peaks detected at 2.60-2.74 second were regarded as common developing solvent considering many previous analyses. Using yeast beads to consume monosaccharides, newly nine kinds of oligosaccharides could be obtained by acceptor reaction.

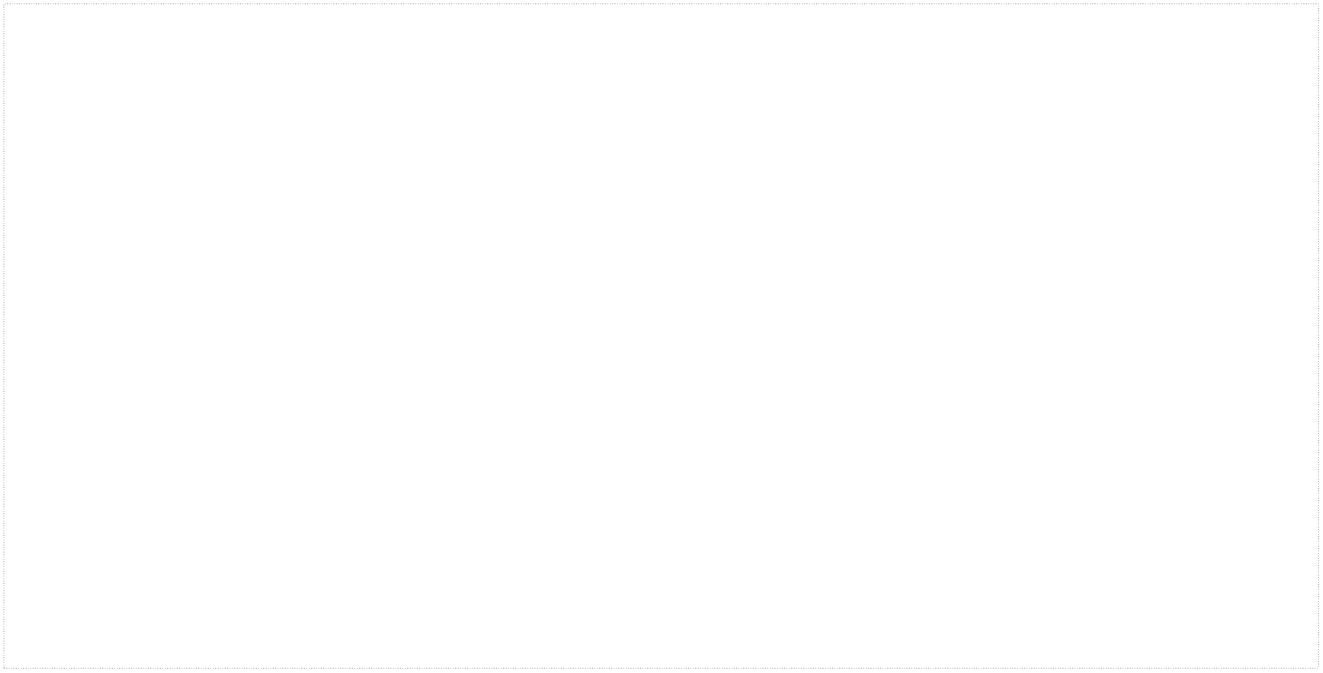

Fig 1. HPLC analysis of oligosaccharide in omija oligosaccharides

2. Thin Layer Chromatography and Maldi-Tof MS analysis in omija oligosaccharides

Degree of polymerization, or DP, is the number of monomeric units in a polymer or oligomer molecule. In TLC analysis, more than nine DP was confirmed. As TLC analysis result showed, nine kinds of oligosaccharides were specified in maldi-tof MS analysis. Every product could be described as this, $[n \cdot M-(n-1) \cdot H_2O+K^+]^+$. “n” is natural number which is more than two. “M” is the molecular weight of glucose. “K” is the molecular weight of potassium.

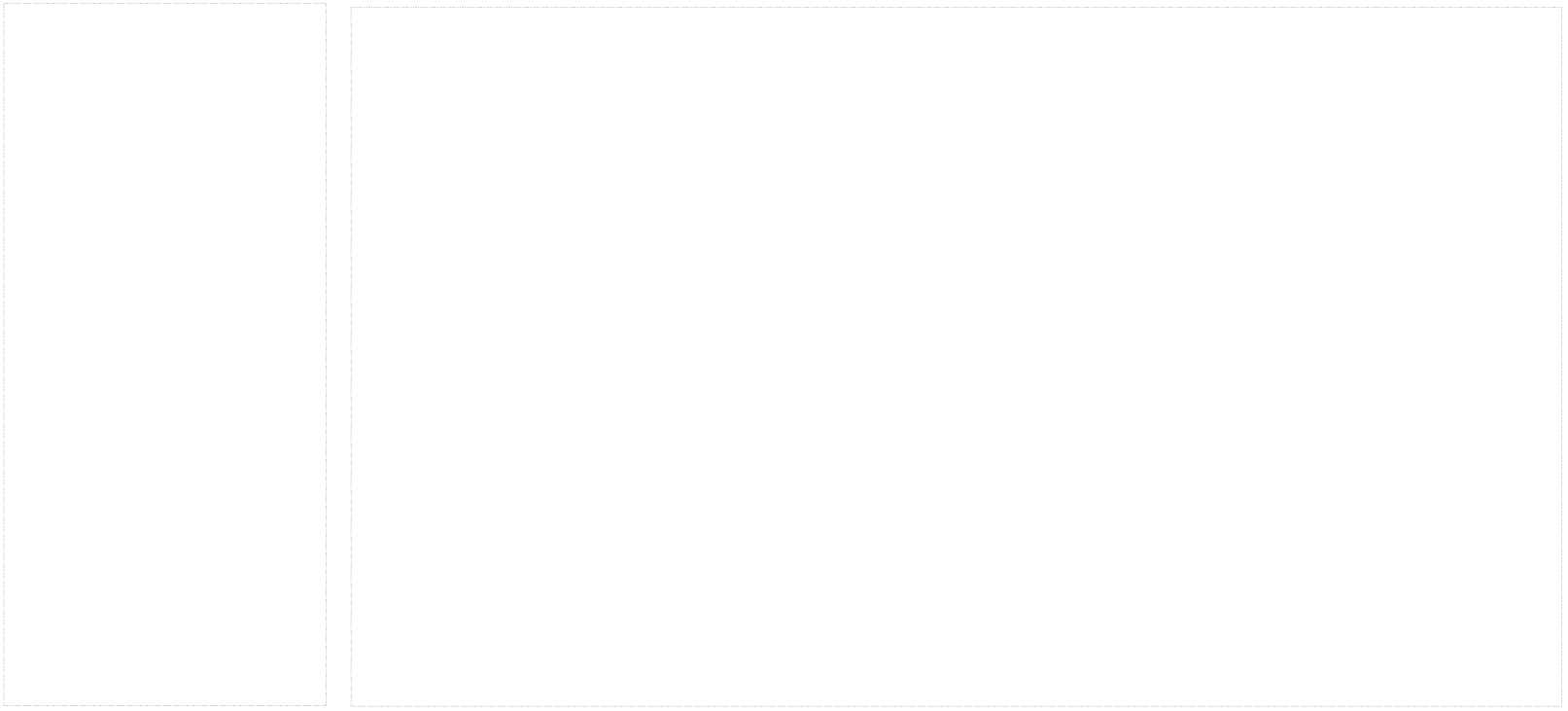

Fig 2. Nine kinds of synthesized oligosaccharides in Thin Layer Chromatography and Maldi-Tof MS analysis

3. Calorie analysis using D-fructose and D-glucose assay procedure

Newly synthesized Omija oligosaccharides juice and regular omija juice adding 50% sucrose were analyzed in the calorie contents in triplicate. Lyophilized each sample was measured to 2g and dissolved in 2 mL of water. After adjusting to optimal pH of various enzyme to hyrolyze sucrose and other carbohydrates, Invertase, α-amylase, and amyloglucosidase were treated step by step. After digestion, calorie analysis was performed. Omija oligosaccharides juice contains 122.65 mg of monosaccharides in 1 g of powder. In Omija with 50% of sucrose, 315.53 mg of monosaccharides was calculated in 1 g of powder. Omija oligosaccharides juice contains 19.62 kcal/200 mL. Omija sucrose juice contains 50.48 kcal/200 mL.

Fig 3. Estimated calorie analysis in 200 mL of omija juice.

Omija sucrose; Omija sucrose juice before acceptor reaction, Omija oligosaccharides; Omija oligosaccharides juice after acceptor reaction.

4. Decrease in insoluble glucan formation from mutansucrase

Mutan is one of the products that cause dental plaque and caries. Mutan, insoluble glucan is formed by mutansucrase using sucrose in teeth. As inhibitors to prevent from forming plaque, omija oligosaccharides, omija, and oligosaccharides were tested. At the final concentration of 500 mg/mL of each sample, omija oligosaccharides, omija, and oligosaccharides showed 96%, 88%, and 71% inhibition effect against forming insoluble glucan comparing to positive control. With the treatment of 250 mg/mL as the final concentration, each sample has 78%, 63%, and 85% inhibitory effect, respectively. Treating with concentration of 125 mg/mL of each resulted in 45%, 35%, and 78% decrease in forming mutan. In 62.5 mg/mL of addition, 46%, 36%, and 25% decrease was shown in insoluble glucan formation.

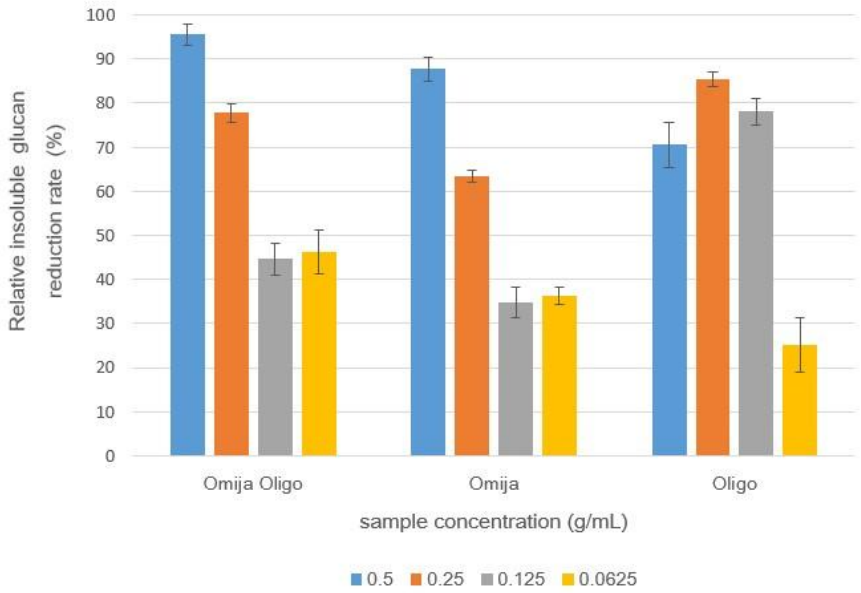

Fig 4. Relative insoluble glucan reduction rate treating omija oligosaccharides, omija, and oligosaccharides

(Omija Oligo; Omija oligosaccharides after acceptor reaction, Omija; Omija sample, Oligo; oligosaccharides)

5. Melanogenesis inhibition activity of Omija and oligosaccharides

At 48 hour, the degree of melanin formation was observed. Omija was so strong for zebrafish larvae that all of them were dead under the lower concentration of omija. The higher concentration of oligosaccharides was treated, the more transparent larvae were. Also, The denser concentration of omija oligosaccharides were treated, the more whitening effect larvae had comparing to negative control.

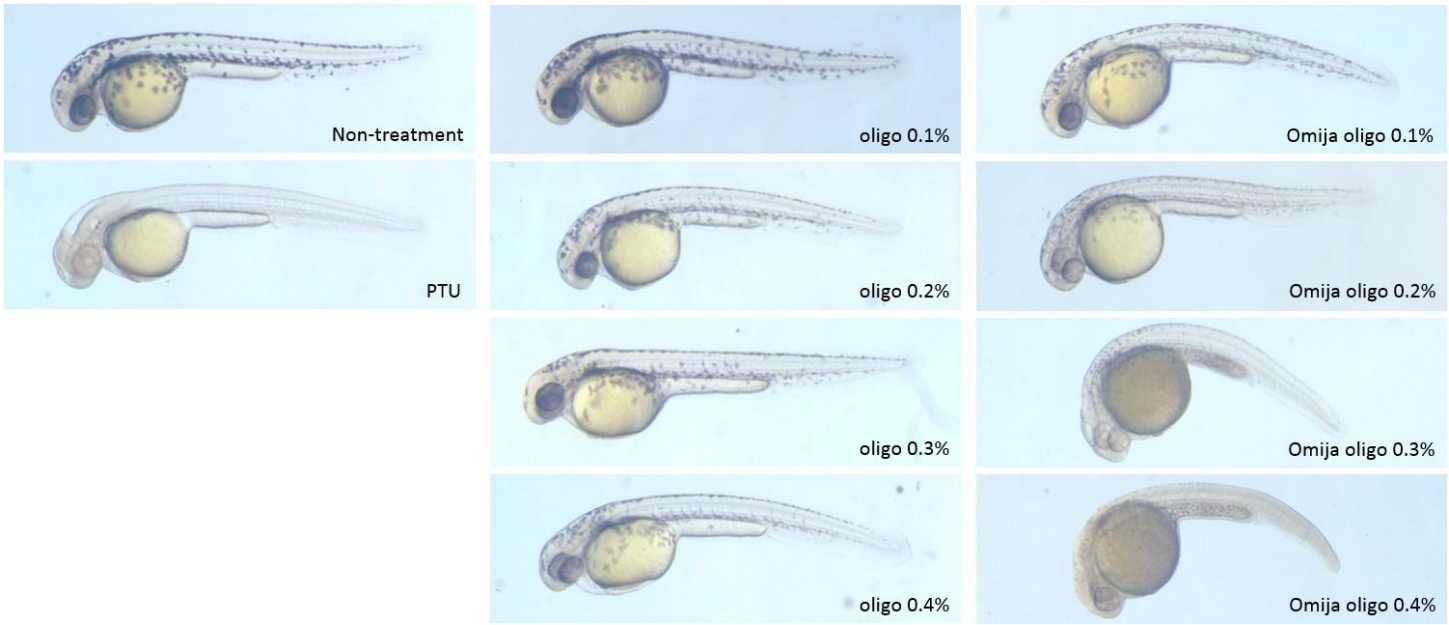

Fig. 5. Zebrafish larvae treated with various concentration of Oligosaccharides and Omija oligosaccharides.

Discussion

Omija oligosaccharides juice contains 122.65 mg of monosaccharides in 1 g of powder. In Omija with 50% of sucrose, 315.53 mg of monosaccharides was calculated in 1 g of powder. 1 g of each powder was dissolved in 1 mL of water. Considering daily consumed beverage volume, Omija oligosaccharides juice contains 19.62 kcal/200 mL. Omija sucrose juice contains 50.48 kcal/200 mL. 61.1% reduction in calorie was shown in Omija oligosaccharides juice than regular omija juice with sucrose.

Omija has been reported to have antibacterial activity against *Streptococcus mutans* (Heo, Choi & Hwang, 2013). In this study, not antibacterial activity against *Streptococcus mutans*, but inhibitory effect against the enzyme, mutansucrase that *S. mutans* produce was firstly suggested by the phenomenon that formation of the insoluble glucan, mutan decreased. Oligosaccharides have been described to reduce soluble and insoluble glucan that mutansucrase produces (LEE, NGUYEN & KWAK, 2017). By consuming omija oligosaccharides juice, dental plaque and caries could be prevented from synergy effect between omija and oligosaccharides. 500 mg/mL of omija oligosaccharides showed 96% of inhibition effect against forming insoluble glucan comparing to positive control. Considering that omija has 88% of prevention effect, Omija oligosaccharides has approximately

10% more inhibitory activity owing to oligosaccharides. The concentration from 500 mg/mL to 125 mg/mL of oligosaccharides showed 71%, 85%, and 78% respectively. The reason that the highest concentration of oligosaccharides did not show the largest inhibitory effect was the existence of alternan and dextran. Besides oligosaccharides, alternan and dextran were also the product of alternansucrase and dextransucrase. In TLC analysis, alternan and dextran are dyed dipping in the sulfuric acid. To get rid of the background color of alternan and dextran, actual oligosaccharide concentration showed rather diminished seemingly regardless the substance.

At 48 hour, the degree of melanin formation was observed. Omija was so strong for zebrafish larvae that all of them were dead under the lower concentration of omija. However, Omija oligosaccharides has less toxicity than Omija sample. This is because oligosaccharide has ability to decrease toxicity (Lambert, McIntyre, & Gauthier, 1991). Firstly, oligosaccharides was elucidate to have potential whitening effect by observing zebrafish larvae in this study. Omija oligosaccharides were expected to have synergy effect between them. Oligosaccharides could increase bioavailability of Omija (Lu, Lin, & Yang, 2017).

Conclusion

Omija oligosaccharides juice from acceptor reaction by *Leuconostoc mesenteroides* B-512FMCM and B-1355CF10 glucansucrase could be healthful beverage in reducing calorie. It would prevent from many kinds of disease that are caused by too much consumption of sugar. By having this newly synthesized omija oligosaccharides juice, dental plaque and caries could be prevented by reducing formation of insoluble glucan, mutan. In this study, Omija was shown to have anti-insoluble glucan forming activity at the first time. Oligosaccharides and omija were expected to have synergy effect to prevent from forming plaque. Omija have reported to show melanogenesis inhibitory effect previously. However, the activity of Omija was so strong that zebrafish larvae could not survive. The presence of oligosaccharides helped to reduce toxicity of Omija. In this regard, Omija oligosaccharides could be applied in food, pharmaceutical, and cosmetic field with simple food process.

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Abstract in Korean

오미자의 학명은 *Schisandra chinensis Turcz. (Baill.)*로 우리나라, 중국, 러시아에서 많이 재배되어 약용 식물로서 이용되어왔다. 오미자에서 생리활성을 보이는 주요 화학성분을 dibenzocyclooctadiene 리그난이라 부른다. 그 중에서도 Schisandrin, Gomisin N, Schiznadrin C, Schisandrin A, Gomisin A와 Gomisin G가 주요 dibenzocyclooctadiene lignan 성분이다. 우리나라에서는 전통적으로 오미자에 설탕을 50% 넣어 오미자 주스를 만들어 마시는데, 본 연구에서는 여기에 *Leuconostoc mesenteroides* B-512FMCM 와 B-1355CF10, 두 유산균이 분비하는 글루칸수크레이스 효소를 첨가하여 올리고당화 하였다. 설탕을 올리고당으로 전환시키므로서 열량을 감소시켜 건강에 좋은 오미자 주스를 섭취할 수 있다. 9가지 이상의 올리고당이 Thin-Layer Chromatography, High-Performance Liquid Chromatography, Maldi-tof MS를 통해 합성되었음을 확인하였고, 오미자 올리고당 주스는 기존의 오미자 주스보다 61.13% 칼로리가 감소되었다. 새로 만들어진 오미자 올리고당 주스 500 mg/mL의 농도에서, 치석 형성 원인균의 효소인 뮤탄수크레이스가 생산하는 불수용성 글루칸을, 샘플을 추가적으로 처리하지 않은 비교군보다 96% 감소시켰다. 오미자 올리고당 주스는 멜라닌 형성을

억제시키는 효과도 지브라피쉬 치어를 통해 확인할 수 있었다. 이는 오미자의 효과 뿐 아니라, 올리고당에서도 멜라닌 형성 억제 물질이 있다는 것을 관찰하였다. 오미자 올리고당은 식품으로서 뿐 아니라 약리학적 기능성과 화장품 소재로서의 가능성도 있을 것으로 기대된다.

Keywords: Omija; Dibenzocyclooctadiene liganan; *Leuconostoc mesenteroides*; Less-digestible oligosaccharides; glucansucrase; Mutansucrase inhibition; Bioavailability.