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Cholesterol-lowering effect of astringent persimmon fruits (*Diospyros kaki* Thunb.) extracts

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Abstract This study aimed to investigate the effects of ethanol extract of astringent persimmon on antioxidant activity, cholesterol, 3-hydroxy-3-methylglutaryl (HMG)-CoA reductase activity, and mRNA expression of cholesterol metabolism-related genes in human hepatoma cell line (HepG2 cells). In the results, DPPH and ABTS radical scavenging activity showed that the different types cultivars of astringent persimmon was similar to Vitamin C as positive control. However, there are not significant differences among samples. In addition, our results showed that cholesterol amounts and HMG-CoA reductase activity were inhibited by astringent persimmon in HepG2 cells. Further, treatment with astringent persimmon upregulated the expression of LDL receptor and SREBP-2, and also increased the level of HDL-associated ABCA1. Taken together, our results indicate that astringent persimmon regulate cholesterol accumulation by inhibiting the oxidative stress and controlling the levels of LDL & HDL-associated gene.

Keywords: astringent persimmon, anti-oxidants activity, 3-hydroxy-3-methylglutaryl-CoA reductase, low-density lipoprotein, high-density lipoprotein

Introduction

To maintain the balance of cholesterol in cell, the biological regulatory system is always works. However, an excessive supply of cholesterol from food or no production of LDL receptor can cause high blood cholesterol, which increases the arteriosclerosis occurrence rate and causes a stroke, a myocardial infarction, an aneurysm, or gangrene. Thus, the control of blood cholesterol is very important (1,2). Atherosclerosis is a symptom that occurs when the wall of an artery, which transports blood from a heart to all body parts, becomes rough or less elastic and narrows because of the accumulation of fat-like substances such as cholesterol, cellulose, or calcium compounds, preventing the supply of sufficient blood to various organs (3). Furthermore, atherosclerosis occurrences are known to be associated with the oxidation products generated by reactive oxygen species (ROS), e.g. hydroxyl radical, superoxide radical, and hydrogen peroxide, which occur in an organism (4,5). Phenol compounds and flavonoids, which are found in various plants, have been consistently reported as being associated with diverse biological activities such as antioxidant, immune, and anticancer. An antioxidant substance from food enhances the biological defense system against ROS, inhibiting cholesterol accumulation (6,7).

Diospyros kaki Thunb, originated in Korea, China, and Japan, is a temperate fruit tree specifically found in East Asia including southern part of Korea, e.g. Yeongdong-gun, Chungbuk; Wanju-gun, Jeollanbuk-do; Boseong-gun, Jeollanam-do; Sangju-si, Uiseong-gun, and Yecheon-gun, Gyeongsanbuk-do; Sancheong-gun, Uiryeong-gun, Haman-gun, Gyeongsangnam-do, Korea (8). Persimmons are categorized into the sweet and astringent kinds. Sweet persimmons taste sweet when eaten raw, while astringent persimmons are further classified as soft, deastringent, and dried (9). Representative persimmons include the flat persimmons from Cheongdo-gun, the Sagok persimmons from Uiseong-gun, the Gojong or the Danseong persimmons from Sancheong-gun, the Sue persimmons from Goryeong-gun, the Gojong or the Godong persimmons from Wanju-gun, the Wolha persimmons from Nonsan-gun, the Muk persimmons from Imsil-gun, the Bidan persimmons from Changseong-gun, the Doong persimmons from Sangju-gun (Sangjudungsi). Persimmons are one of the three major fruits in Korea, along with grapes and apples. Persimmons, rich in saccharides, such as glucose and fructose, Vitamin A and C, inorganic salts, such as calcium and potassium, water soluble tannin, and dietary fiber (10).

The function of persimmons in the human body has been reported colon contraction, secretagogue action, apoplexy and hematemesis treatment, and antipyretic action (11). The fruits and leaves of persimmons have also been reported recently as having the effects of lowering blood pressure, and preventing cardiac arrests, along with anti-mutagenic, antioxidant, anti-allergenic, and anti-cancer

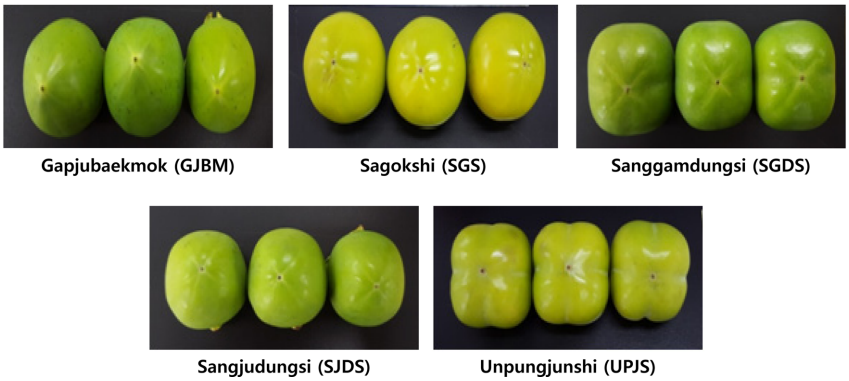

Fig. 1. Different types cultivars of astringent persimmon.

effects (12,13). Furthermore, studies are being conducted focusing on developing the processed foods using persimmons, e.g. the properties of persimmon makgeolli (14) and persimmon bread (15), effects of persimmon syrup on blood sugar levels (16).

However, these studies are focused mainly on ripe persimmons, and studies on the biological activity and mechanisms of astringent persimmons have not yet been clarified. In particular, few studies have been performed on the functions of different type's cultivars of astringent persimmon. Hence, this study evaluates the effects of astringent persimmons on antioxidant activation and cholesterol improvement for different type's cultivars and provides basic data for using astringent persimmons as functional foods.

Materials and Methods

Reagents 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) and dimethyl sulfoxide (DMSO), were purchased from Sigma-Aldrich Co. (St. Louis, MO, USA). Dulbecco's Modified Eagle Medium (DMEM), fetal bovine serum (FBS), penicillin-streptomycin (P/S), and phosphate buffed saline (PBS) were purchased by Gibco (Waltham, MA USA). For 2',7'-dichloro-dihydrofluorescein diacetate (DCFH-DA), the products purchased were made by Molecular Probes (Waltham, MA, USA). For other reagents for analysis were purchased from Sigma–Aldrich unless specified otherwise.

Sample preparation For astringent persimmons, five different types, i.e. Gapjubaekmok (GJBM), Sagokshi (SGS), Sanggamdungsi (SGDS), Sangjudungsi (SJDS), and Unpungjunshi (UPJS) (Fig. 1), were obtained from Gyeongsangbuk-do Agricultural Research and Extension Services, Sangju Persimmon Experimental Farm (Sanju, Korea). The persimmons were cleaned, removing all foreign materials and stems. The persimmons were sliced and dried in hot air at 50°C for 48 h, and pulverized. The pulverized persimmon (20 g) was added to 500 mL of 70% ethanol and stirred at 70°C for 6 h and being extracted twice. The extracts were filtered and concentrated using a vacuum concentrator (EYELA CCA-1110; Tokyo Rikakikai Co., Tokyo, Japan) and then lyophilized with a freeze dryer (Lyoph-pride Series Freeze Dryer; ilShinBioBase, Dongducheon, Korea) at −70°C.

Total phenol and tannin content analysis The total phenol content was analyzed using the Folin-Denis assay (17). First, 300 μL of distilled water was added to 1 mg/mL of 100 μL astringent persimmon ethanol extract, and 1 mL of Folin-Ciocalteu phenol reagent was added afterward. Then 1 mL of $NaCO_3$ saturated solution was mixed after 5 min and was left for an hour at room temperature before being measured for absorbance at 760 nm using a spectrophoto-meter (SpectraMax M5; Molecular Devices, LLC., Sunnyvale, CA, USA). The phenol compounds content was quantified after the analytical calibration curve was made using gallic acid concentration as the standard substance. The total tannin content was measured using the method of Bubba *et al*. (18). In other words, 15 mL of 0.5 M HCl solution was added to the 1 g astringent persimmon powdered specimen before being extracted in a shaking water bath at a constant-temperature of 60°C for 30 min. Next, 9.4 mL of distilled water was put into the 100 μL extract, and mixed with 300 μL Folin-Denis reagent and stirred, then placed in a dark place for an hour before being measured for absorbance at 760 nm. The measurements were quantified using gallic acid standard curve.

DPPH radical scavenging ability measurement DPPH radical scavenging ability was measured using the method of Mensor *et al*. (19). The 0.3 mM DPPH solution was mixed with the astringent persimmon ethanol extract (50 and 100 μg/mL) or Vit C as positive control (20 μg/mL) and was incubated at room temperature for 30 min. And the absorbance measured at 518 nm using a spectrophotometer. The negative control was experimented using PBS instead of samples, and the radical scavenging ability was indicated in percentage (%).

$$\text{DPPH radical scavenging activity (\%)} = (1 - ABS_{sample}/ABS_{control}) \times 100$$

ABTS radical scavenging ability measurement ABTS radical scavenging

ability was measured by Re *et al*. (20) method. Seven mM ABTS solution and 2.45 mM potassium persulfate were mixed in a 1:1 ratio and were stored in a dark place at room temperature for 12-16 h to produce ABTS radical. ABTS radical was controlled at 732 nm for the absorbance to be 0.7±0.02 before use. The 1.8 mL ABTS solution was mixed with 0.2 mL of astringent persimmon ethanol extract (50 and 100 μg/mL) or Vit C as positive control (20 μg/mL) and was left at room temperature for 30 min and measured at 732 nm for the absorbance using a spectrophotometer (Molecular Devices, LLC.). PBS was used for the negative control, and the radical scavenging ability was indicated in percentage (%).

$$\text{ABTS radical scavenging activity (\%)} = (1 - ABS_{sample}/ABS_{control}) \times 100$$

Cell culture HepG2 cells, a human liver carcinoma cell line, were used for the experiment to investigate the effects of the astringent persimmon ethanol extract on the cholesterol control and the expressions of the genes related to production of HDL and LDL. HepG2 cells were purchased from the Korea Cell Line Bank (Seoul, Korea) and cultured using DMEM media includes 10% FBS and 1% P/S in a 5% CO_2 incubator at 37°C. The cultured cells were passaged biweekly while in use.

Cell cytotoxicity measurement HepG2 cells were plated into 96 well at 1×10^4 cell/well and were cultured for 4 h before being treated with samples in various concentrations, i.e. 50 and 100 μg/mL, and cultured for another 24 h. And then, MTT solution (5 mg/mL) was added to each well and the wells were re-incubated at 37°C for 4 h. The formazan produced from the process was dissolved in DMSO and was measured at 540 nm for the absorbance.

Suppression effect of cholesterol synthesis HepG2 cells were placed into 6 well at 3×10^5 cell/mL and cultured for 24 h before being treated with ethanol extract for different type's cultivars of astringent persimmon. The supernatant was then concentrated using centrifucal filter units (Vivaspin 4; Sartorius stedium, Göttingen, Germany). The total amount of cholesterol was measured using a cholesterol/ cholesterol ester quantification kit (Abcam, Cambridge, MA, USA). Briefly, the samples and reagents were mixed and incubated in a dark place at 37°C for 1 h before being measured using an ELISA reader.

HMG-CoA reductase activity assay HMG-CoA reductase activity assay kit (Sigma-Aldrich) was used in confirming 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA) reductase activity suppression of ethanol extract for different type's cultivars of astringent persimmon.

Real-time RT-PCR HepG2 cells were placed into 12 well at 5×10^5 cell/well and incubated for 2 h before being treated with ethanol extract of astringent persimmon and incubated for another 24 h. The total RNA in the cells was extracted using an RNeasy mini kit (Qiagen, Hilden, Germany), and cDNA was synthesized using a reverse transcription system kit (Promega, Madison, WI, USA). The mRNA expressions were then analyzed in Real-Time PCR Detection System (CFX96™; Bio-Rad Laboratories, Hercules, CA, USA) using SYBR Green mix (Qiagen). All results were normalized to glyceraldehyde 3-phosphate dehydrogenase (GAPDH) expression. The following primer sequences were used for real-time RT-PCR: GAPDH, 5'-GAG CCA AAA GGG TCA TCA TC-3' (forward), 5'-TAA GCA GTT GGTGGT GCA GG-3' (reverse); LDL receptor, 5'-GAG TAC ACC AGC CTC ATC C-3' (forward), 5'-GCT GAT GAC GGT GTC ATA G-3' (reverse); SREBP-1, 5'-GCG CCT TGA CAG GTG AAGTC-3' (forward), 5'-GCC AGG GAA GTC ACT GTC TTG-3' (reverse); SREBP-2, 5'-TAG ACC GCT CAC GGA TT-3' (forward), 5'-AGG CAT CAT CCA GTC AAA C-3' (reverse); ABCA1, 5'-AAT GTC AAG GTG TGG TTC AATA-3' (forward), 5'-CTG CTG CTT GGT GAG AT-3' (reverse); SR-B1, 5'-AGC TCA ACA ACT CCG AC-3' (forward), 5'-GCT GTA GAA CTC CAG CGA-3' (reverse) and HMGR, 5'-TAC CAT GTC AGG GGT TAC GTC-3' (forward), 5'-CAA GCC TAG AGA CAT AAT CAT C-3' (reverse).

Table 1. Total polyphenol and tannic acid contents of astringent persimmons (unit: mg/g)

Sample	Total polyphenol content	Tannic acid content
Gapjubaekmok	116.59±2.34	13.16±0.56
Sagokshi	92.01±3.59	17.17±0.96
Sanggamdungsi	95.06±2.70	7.58±0.16
Sangjudungsi	88.78±1.23	15.76±0.55
Unpungjunshi	79.44±1.22	19.80±0.88

Statistical analysis Statistical analyses were performed with SPSS v12.0 (SPSS Inc., Chicago, IL, USA). Data are represented as the mean ±SEM from three independent experiments, unless stated otherwise. The differences between the treatments were analyzed using one-way ANOVA (Analysis of Variation), and were verified for significant difference at $p<0.05$ level using Duncan's multiple range test.

Results and Discussion

Total phenol and tannin content of the ethanol extract for astringent persimmons The total phenol content of the ethanol extract of astringent persimmon was showed in Table 1. The results showed GJBM being the highest with 116.6 mg/g, followed by SGDS with 95.1 mg/g, SGS with 92.0 mg/g, SJDS with 88.8 mg/g, and UPJS showing the lowest content with 79.4 mg/g. These are higher than the study results reported by Denev and Yordanov (21), which ranged from 223.0 to 916.8 mgGAE/100 g. Furthermore, the tannin content for different type's cultivars of astringent persimmon resulted in GJBM with 13.2 mg/g, SGS with 17.2 mg/g, SJDS with 15.8 mg/g, with SGDS being the lowest with 7.6 mg/g and UPJS being the highest with 19.8 mg/g. It is assumed that these differences in the phenol compounds and tannin content of the same breed come from being cultivated in different regions and harvested at different

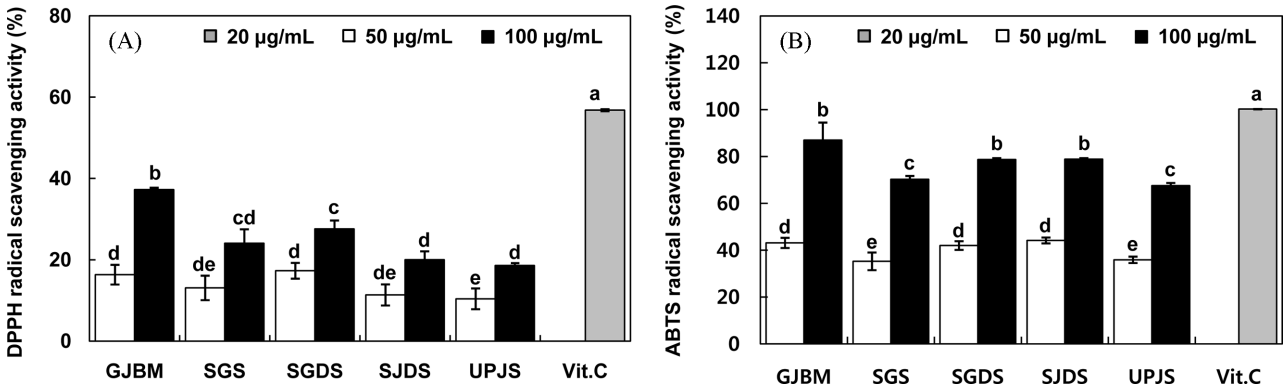

Fig. 2. Antioxidant activities of different type's cultivars of Astringent Persimmon. DPPH radical scavenging activity (A), ABTS radical scavenging activity (B). GJBM, Gapjubaekmok; SGS, Sagokshi; SGDS, Sanggamdungsi; SJDS, Sangjudungsi; UPJS, Unpungjunshi. Values are the Mean±SEM of three replications (n=3). Values expressed by different letters are significantly different at p<0.05.

times. The study results showed significant differences in the total phenol and tannin contents for different type's cultivars of astringent persimmon and can be used as a basic data for predictions of the biological activity of persimmons.

Antioxidant activity of the ethanol extract of astringent persimmons A removal of free radicals has been reported as a very important antioxidant mechanism in suppressing lipid oxidation in a human body, and thus suppressing disease and aging (22). DPPH radical scavenging ability is a method for measuring the reduction and antioxidant abilities of materials by the degrees of reduction. It is one of the antioxidant activity measurement methods made possible by the characteristic of hydrazyl reactive to antioxidant materials, that is, its nitrogen atoms easily accept hydrogen atoms in their unstable state (19). The ABTS radical scavenging activity method measures the $ABTS^{+}$ scavenging activity of the extract in absorbance values when ABTS and potassium persulfate were left in a dark place to produce $ABTS^{+}$, which is removed by the extract's antioxidant activity, and the bluish-green color that is a characteristic of the radicals gets bleached (20). DPPH radical scavenging activity of different types cultivars of astringent persimmons showed at 100 μg/mL GJBM 37.3%, SGS 24.1%, SGDS 27.6%, SJDS 20.03%, UPJS 18.6%, respectively, with GJBM being the highest (Fig. 2A). ABTS radical scavenging activity of the astringent persimmon extract showed antioxidant activity in the order of SJDS, SGDS, SGS, and UPJS, from high to low, with GJBM being the highest at 86.9%. Vit C as positive control at Both measurement results in DPPH and ABTS radical scavenging activity showed high antioxidant activity in all kinds of astringent persimmons, but the differences between the kinds were insignificant (Fig. 2B). The differences between the two radicals are assumed to be caused by the different reactive elements, on which consistent studies are needed.

ROS plays an important role in the accumulation of cholesterol. Gesquière *et al*. (23) were explained the presumed processes of oxidative stress on cholesterol homeostasis. First, ROS lead to increasing the cellular free cholesterol and cholesteryl esters by a stimulation of the cellular uptake of oxidative LDL. Second, ROS was stimulated the HMG-CoA reductase activity result in enhancing the cholesterol biosynthesis. Third, ROS was increased the cellular cholesteryl esters content via interrupting a cycle of transesterification and hydrolysis via the acyl coenzyme A: cholesterol acyl transferase (ACAT) and the neutral cholesteryl ester hydrolase (NCEH). In other words, CEs increased by increasing the ACAT activity and inhibiting the NCEH activity. Finally, ROS diminished the cell removal of free cholesterol to HDL. Therefore, enhancing biological defense system against ROS can prevent cholesterol accumulation. Taking the natural foods that contain abundant biological activity materials increases the antioxidant activity level in a human body, playing a key role in cholesterol accumulation prevention and disease suppression (22). Taken together, the astringent persimmons, an excellent source of antioxidant activity, can be used in the natural antioxidant product development and contribute to either the lipid metabolism or cholesterol control.

Cell cytotoxicity of the ethanol extract of astringent persimmons Cell cytotoxicity of the ethanol extract of the astringent persimmons was found in HepG2 cells. Cell cytotoxicity was not observed in the extract at 50 and 100 μg/mL concentrations (Fig. 3). Thus, the concentration of the extract was set at 100 μg/mL to investigate its effect on regulating cholesterol in HepG2 cells.

Suppression effect of the ethanol extract of astringent persimmons on cholesterol synthesis and HMG-CoA reductase activity As discussed above for ROS mediated modifications of the cholesterol homeostasis, we next sought to confirm if astringent persimmons inhibit which step of cholesterol metabolism.

To investigate the cholesterol-synthesis suppression effect of the ethanol extract of astringent persimmons, an experiment was conducted using HepG2 cell. In comparing the extract at 100 μg/mL for different types cultivars, the cholesterol-synthesis suppression

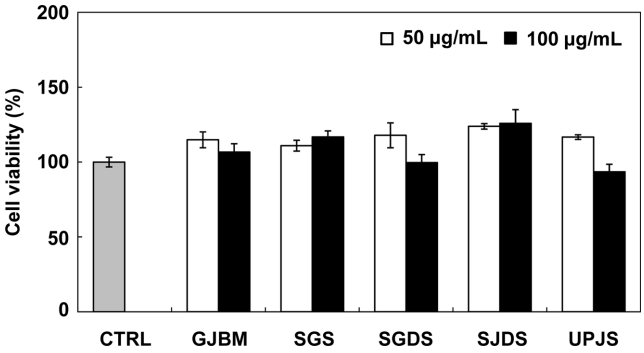

Fig. 3. Effects of different type's cultivars of Astringent Persimmon extracts on cell viability in HepG2 cells. GJBM, Gapjubaekmok; SGS, Sagokshi; SGDS, Sanggamdungsi; SJDS, Sangjudungsi; UPJS, Unpungjunshi. Values are the Mean±SEM of three replications (n=3). *p<0.05 compared to control.

effect was observed highest in SGDS, and also high in SJDS and SGS (Fig. 4A). Furthermore, the activity suppression effect of HMG-CoA reductase, a cholesterol production enzyme, was verified. Pravastatin was used as a control group. Similar to the cholesterol-synthesis suppression results, the suppression effect of HMG-CoA reductase was highest in SGDS ethanol extract, showing similar effects in the SJDS and SGS extracts as well. According to Tian *et al*. (24), tannins from persimmons effectively reduced the lipid oxidation induced from H_2O_2 or Fe^{2+}/ascorbic acids, increased antioxidant enzyme activity of superoxide dismutase (SOD), glutathione peroxidase (GPx), etc. of the mouse that decreased by bromobenzene injection, and decreased the malondialdehyde (MDA) level. Furthermore, the tannin administration of obese patients decreased the blood cholesterol level and contributed to arteriosclerosis suppression (25). In this study, the tannin content was highest in UPJS but, unlike the above mentioned study results; it showed the lowest cholesterol reduction effect. However, the phenol compound content showed a similar pattern, and thus, it is assumed that antioxidant activity and cholesterol-synthesis reduction effect of the ethanol extract of astringent persimmons were caused by other phenol compounds of the astringent persimmons.

Effects of the ethanol extract of astringent persimmons on LDL and HDL genes To investigate the effects of astringent persimmon extract on gene expression related to cholesterol metabolism in HepG2 cells, HepG2 cells were treated with the astringent persimmon extract and measured mRNA, gene related to LDL and HDL using RT-PCR. The results confirmed that the mRNA expression in ABCA1, LDL acceptor, significantly increased in SGDS, SGS, and SJDS extracts compared to the control cells. The gene expression of SREBP1, SREBP2, and HMGCR increased in SGDS and SJDS extracts. On the other hand, SR-B1 was not affected much in the extract treatment (Fig. 5). Consistent with these results, protein expression related to cholesterol metabolism were confirm in HepG2 cells (data not shown).

Cholesterol gets synthesized in a human body by two methods: one is from food; the cholesterol supplied to humans from food intake gets absorbed in the small intestines and gets transformed to the cholesterol ester by ACAT. Cholesterol ester bonds with apolipoprotein and other lipids to become chylomicron, which gets supplied through the peripheral blood vessels to adipose tissues or muscles and the remaining residues flow into the liver through the acceptors. The liver tissues take in cholesterol into the cells by absorbing LDL mainly through the LDL acceptors, and the cholesterol transportation from the peripheral tissues to the liver gets carried out by HDL (26). The other method is the synthesis at the liver. The biosynthesis of cholesterol at the liver starts from acetyl CoA and undergoes a process of HMG-CoA and mevalonic acid and biosynthesized mediated by HMG-CoA reductase and various enzymes. Such cholesterol is used in producing cell membrane, steroid hormones, and bile acids (27).

Cholesterol control as well as cholesterol biosynthesis takes place at the liver, that is, when a cholesterol concentration decreases within cells, the LDL receptor expression increases, binding LDL cholesterol with LDL receptor before cholesterol flows into the cells. On the other hand, when a cholesterol concentration increases

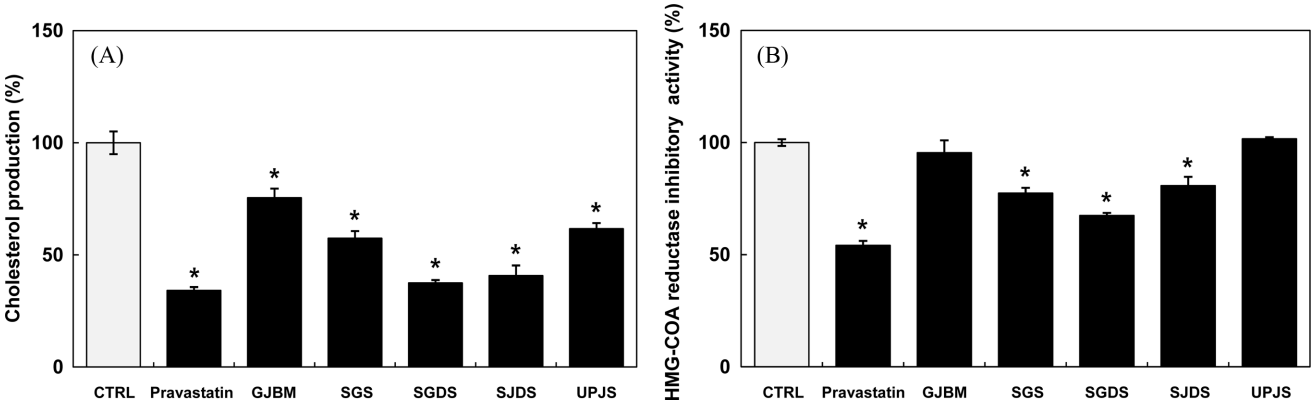

Fig. 4. Effects of different type's cultivars of Astringent Persimmon extracts on cholesterol synthesis inhibition (A) and HMG-CoA reductase inhibition activity (B) in HepG2 cells. GJBM, Gapjubaekmok; SGS, Sagokshi; SGDS, Sanggamdungsi; SJDS, Sangjudungsi; UPJS, Unpungjunshi. Values are the Mean±SEM of three replications (n=3). *p<0.05 compared to control.

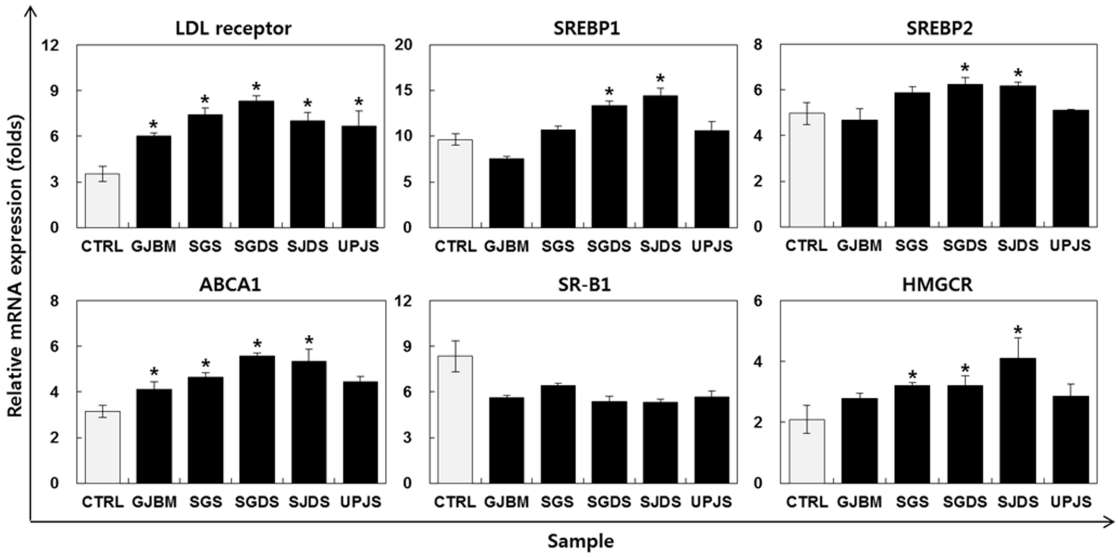

Fig. 5. Effect of different type's cultivars of Astringent Persimmon extracts on expression of cholesterol-related mRNA in HepG2 cells. GJBM, Gapjubaekmok; SGS, Sagokshi; SGDS, Sanggamdungsi; SJDS, Sangjudungsi; UPJS, Unpungjunshi. Values are the Mean±SEM of three replications (n=3). *p<0.05 compared to control.

within cells, the opposite phenomenon occurs, suppressing LDL receptor expression and failing to remove blood cholesterol. Such LDL acceptor manifestation is controlled by SREBP because of SRE-1 existing in LDL acceptor genes. In other words, when a sterol concentration decreases within cells, SREBP gets divided and enters into a nucleus to join with SRE-1 existing in the SREBP-target gene promoters, thus activating nuclear gene transcription of the enzymes that are in charge of cholesterol synthesis (27,28).

Furthermore, the expression of ABCA1 genes, which is a transporter participating in cholesterol efflux are also very important in cholesterol metabolism. Reverse cholesterol transport, which is a process of transporting the excessive cholesterols accumulated on the blood vessels to the liver, prevents arteriosclerosis by expediting cholesterol efflux. Genes that involve in the reverse cholesterol transport are known to be diverse, the most representative of which is ABCA. ABCA1 plays a role in cholesterol efflux by secreting cholesterol out of the cells, which then turns them into the bile acids at the liver (29,30). SR-B1, a HDL receptor and another very important gene in HDL metabolism, plays a mediating role in cholesterol absorption from the cells and is distributed most abundantly in the liver among lipid metabolism tissues (31,32). SR-B1 genes have been reported for their action mechanism to suppress arteriosclerosis by participating in discharging cholesterol from the body through the liver (33).

According to Matsumoto *et al*. (34), the C57BL/6.KOR-ApoE^{shl} mice that ate immature persimmons for ten weeks showed significant decrease in blood LDL-cholesterol and neutral lipid, confirming the mechanism of increasing SREBP-2 gene expression at the liver. Furthermore, there have also been reports that treating the functional elements of natural products, e.g. polyphenol or isoflavone, with cells increases LDL acceptor or SREBPs gene manifestation, thus lowering blood cholesterol level (35,36). The above-mentioned results confirmed that cholesterol synthesis decreased in the cells that the astringent persimmon extract was added. It was able to confirm that the result was caused by increasing manifestation of LDL acceptor mRNA and SREBP-2 mRNA, which controls LDL acceptor mRNA. Moreover, the astringent persimmon extract increases the manifestation of mRNA of ABCA1 gene, which is related to HDL synthesis, and the increased HDL protein indicates the cholesterol reduction effect.

In conclusion, using the astringent persimmon ethanol extract and HepG2 (human hepatoma cell line), this study confirms the extract effects in antioxidant activation, suppression of cholesterol and HMG-CoA reductase activity, and the molecular mechanism related to LDL and HDL. As a result, the extract showed high DPPH and ABTS radical scavenging activity, with no significant differences for different type's cultivars. Furthermore, all extracts showed suppression effects in HMG-CoA reductase activity as well as cholesterol synthesis. In particular, high activity suppression effects were confirmed in SGDS, SJDS, and SGS (p<0.05). The extract is also assumed to increase LDL acceptors and SREBP-2, which controls LDL receptor, increases expression of ABCA1, a cholesterol transporter, decreasing LDL and increasing HDL. These results prove that persimmons, a long-time traditional natural product, have antioxidant and cholesterol prevention effects and can contribute in preventing the related metabolic syndromes.

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Disclosure The authors declare no conflict of interest.

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