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Antioxidative and antiproliferative activities of ethanol extracts from pigmented giant embryo rice (*Oryza sativa* L. cv. Keunnunjami) before and after germination

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BACKGROUND/OBJECTIVES Oxidative stress is a major cause of cancer. This study investigated the effects of the ethanol extracts from germinated and non-germinated Keunnunjami rice, a blackish-purple pigmented cultivar with giant embryo, on selected human cancer cell lines and on the antioxidant defense system of mice fed with a high-fat diet.
MATERIALS/METHODS: High fat-fed mice were orally administered with either distilled water (HF) or extracts (0.25%, w/w) from brown (B), germinated brown (GB), Keunnunjami (K), and germinated Keunnunjami (GK) rice.
RESULTS: In comparison with the brown rice extract, Keunnunjami extract showed higher anticancer effect against cervical and gastric cell lines but lower anticancer activity on liver and colon cancer cells. Mice from the HF group showed significantly higher lipid peroxidation and lower antioxidant enzyme activities than the control group. However, the oxidative stress induced by high-fat diet markedly decreased in B, GB, K, and GK groups as compared with the HF group.
CONCLUSIONS: Germination may be an effective method for improving the anticancer and antioxidative properties of Keunnunjami rice and extracts from germinated Keunnunjami rice may serve as a therapeutic agent against cervical and gastric cancers and oxidative damage.

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Keywords: Rice, germination, cancer, antioxidant, nutritional and metabolic diseases

INTRODUCTION

Keunnunjami rice is a new blackish-purple pigmented cultivar with a giant embryo that was developed in Korea through conventional breeding. This pigmented rice was reported to contains high levels of antioxidant compounds and exhibit strong anti-obesity, hypolipidemic, and antioxidant properties [1-3]. Pigmented rice varieties, particularly those with dark-colored pericarps such as black and purple rice, are known for their health-promoting effects and functional activities, owing to the high contents of flavonoids, phenolic compounds, and anthocyanins [4,5]. Rice varieties with enlarged or giant embryos are similarly rich in bioactive compounds and nutrients such as γ-aminobutyric acid (GABA), γ-oryzanol, and tocopherol, which have various physiological properties, such as anticancer, anti-tumor, and antioxidative activities [6-8].

Germination is an effective and inexpensive process for enhancing the nutrient content and eating quality of brown rice [9]. Whole rice grains are soaked for a few days until the emerged radicle is approximately 2 to 5 mm [10]. This process induces biochemical changes like enzyme activation and release of free and bound materials, resulting in the softening of texture and increase in bioactive compounds and nutrient bioavailability [11,12]. Several studies have shown that germination significantly increased the amounts of GABA, γ-oryzanol, and tocols in brown rice [10,13]. Other cereal grains, such as barley, wheat, and oat, also exhibited enhanced levels of nutrients and bioactive compounds after germination [14,15]. The previous study reported that germinated rice could reduce the risk of various chronic diseases such as hyperlipidemia, cardiovascular disease, cancer, and diabetes [16].

Keunnunjami is a unique verity of rice that was bred to possess both a dark purple-colored seed pericarp and a giant embryo. It is expected to have better biofunctional properties than other pigmented varieties or non-pigmented rice with giant embryo. Our recent reports have demonstrated that

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Keunnunjami showed greater antioxidant capacity than non-pigmented giant embryo rice [17]. Furthermore, dietary feeding of germinated Keunnunjami rice powder has been found to ameliorate the lipid metabolism in ovariectomized rats [18]. To further explore the functional properties of Keunnunjami and the influence of germination on its biological activities, the present study carried out the antiproliferative and antioxidative effects of ethanolic extracts from germinated and non-germinated Keunnunjami rice, in comparison to those from non-germinated and germinated ordinary normal embryo brown rice.

MATERIALS AND METHODS

Germination and rice extract preparation

Whole grains of Keunnunjami rice, a dark purple-pigmented cultivar with a giant embryo, and Ilpum rice, an ordinary non-pigmented cultivar, were used. The two rice varieties were of the same species and subspecies and obtained after harvest in October 2016 from the Department of Agricultural Science (Korea National Open University, Seoul, Korea). To rice germinate, The 50 g of gains set a stainless container overlaid with sterilized gauze after washing by distilled water and incubated 3 days at 30°C in an oven. The container was added with 100 mL distilled water and cover surface with plastic wrap with holes prior to incubation. The grains were checked regularly at 12 h interval to make sure there was no unpleasant odor and fungal growth. After 72 h, the germinated grains were dried at 50°C for 2 h, ground using a grinding machine (OCM-D1000PB, OCOO, Chungnam, Korea), passed through a 100 mesh sieve, and extracted with 70% ethanol (1:3 solid/solvent ratio) at 40°C with constant stirring for 2 h. The mixture was then filtered and the rice powder was extracted again with 70% ethanol (1:1 solid/solvent ratio) at 40°C with constant stirring. The process was repeated and the combined extracts were concentrated using a rotary evaporator (Eyela N-1000, Tokyo, Japan) to completely remove the ethanol, obtaining an extract yield of 5% (w/w). For the non-germinated rice extract samples, the rice grains (50 g) were washed, dried, ground, and extracted using the same method described above for the germinated grains. The concentrated extracts of the germinated and non-germinated rice samples were freeze-dried. The levels of bioactive components such as GABA, tocopherol, phytic acid, and polyphenol were determined using previously described methods [19-22] and the results are shown in Table 1. All chemicals used in this study were of analytical grade and purchased from Merck KGaA (Darmstadt, Germany) and Sigma-Aldrich, Inc. (Steinheim, Germany).

Cell culture and cell viability

The cancer cell lines AGS, HeLa, HEP3B, and HT-29 were obtained from the Korea Cell Line Bank, Seoul, Korea. The AGS and HT-29 cells were cultured in Roswell Park Memorial Institute (RPMI)-1640 (Hyclone Laboratories, Utah, USA) supplemented with 10% fetal bovine serum. HeLa cells were cultured in MEM (Hyclone Laboratories, Inc., Utah, USA) supplemented with 10% fetal bovine serum, while HEP3B cells were cultured in DMEM (Hyclone Laboratories, Utah, USA) supplemented with 10% fetal bovine serum in a 5% CO_2 atmosphere at 37°C. Cell viability was analyzed using an MTT assay kit (Roche Molecular Biochemicals, Mannheim, Germany) according to the manufacturer's instruction. Briefly, the treated cells were incubated with the MTT reagent for 4h and lysed with a lysis buffer. The absorbance was measured from 550 nm to 650 nm after an overnight incubation. The 50% inhibitory concentrations (IC50s) were calculated from four concentrations of the extracts (100, 250, 500, 750 μg/mL).

Animals and diets

Four-week-old male C57BL/6N mice (n = 48, 20 g each) were purchased from Central Laboratory Animal Inc. (Seoul, Korea) and housed individually in hanging stainless steel cages in a room maintained at 25 ± 2°C with 50% relative humidity and 12/12 h light-dark cycle. They were initially fed with a commercial pelletized diet and distilled water ad libitum for 1 wk. Following a 1 wk period, 40 animals were fed with a high-fat diet (45% calories from fat), purchased from Feed Lab (Guri, Gyeonggido, Korea) for 3 wks to induce obesity, while the remaining eight animals were given an AIN93G diet [23]. After 3 wks, the high-fat diet fed mice were divided into 5 dietary groups (n = 8) and orally administered 1 mL of distilled water (HF group) or ethanol extracts in distilled water (0.25%) of the non-germinated normal brown rice (B group), germinated brown rice (GB group), non-germinated Keunnunjami (K group), or germinated Keunnunjami (GK group). The control mice fed with AIN93G diet were orally administered distilled water (C group). For 4 weeks, daily oral gavage was carried out with either the sample rice extracts or distilled water. All mice had free access to water and food during this period. The blood samples gathered from the inferior vena cava (IVC) of anaesthetized mice through CO_2 inhalation. After samples transferred into ethylenediaminetetraacetic acid (EDTA)-coated tubes and centrifuged at 1,000 x g for 15 min at 4°C to get the plasma. The heart, liver, kidney, and white adipose tissues (perirenal, epididymal, and inguinal) were excised, washed with saline solution, and weighed. The collected samples were stored at

Table 1. Bioactive compounds in germinated and non-germinated rice extracts

Bioactive compound (mg/100 g extract)	Brown rice		Keunnunjami rice	
	Non-germinated	Germinated	Non-germinated	Germinated
GABA	23.28 ± 2.65[a]	814.36 ± 9.87[c]	95.59 ± 3.87[b]	1,198.26 ± 10.55[d]
Tocopherol (α, β, γ)	0.02 ± 0.00[a]	0.07 ± 0.01[c]	0.06 ± 0.00[b]	0.16 ± 0.01[d]
Phytic acid	1.09 ± 0.02[a]	3.27 ± 0.04[c]	1.69 ± 0.01[b]	6.67 ± 0.05[d]
Polyphenol	2.49 ± 0.02[a]	6.04 ± 1.29[b]	11.58 ± 1.01[c]	18.44 ± 1.29[d]

Values are means ± SE (n = 3).
[a-d] Means in the same row with different letters are significantly different at $P < 0.05$, as analyzed by Tukey's range test.
GABA, γ-aminobutyric acid

-70°C before analysis. The study was approved by the Kyungpook National University of the Animal Care Committee. (Approval No. KNU-2016-0025).

Determination of lipid peroxidation and antioxidant enzyme activity

The amount of thiobarbituric reactive substances (TBARS) in the plasma and erythrocytes was determined following the method of Ohkawa *et al.* [24]. The activities of hepatic and kidney antioxidant enzymes superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), catalase (CAT), glutathione reductase (GR), and paraoxonase (PON) were determined based on the previous methods described by Chung *et al.* [17].

Statistical analysis

All data are presented as the mean ± standard error (SE). The data were performed by one-way analysis of variance (ANOVA) using IBM SPSS Statistics 23.0 software. (IBM Corporation, Armonk, NY, USA). Differences between the means were assessed using Tukey's test. Statistical significance was considered at $P < 0.05$.

RESULTS

Antiproliferative activitie

The half maximal inhibitory concentration (IC50) values were the lowest for the Extracts of from the germinated Keunnunjami rice and highest for the extract from the non-germinated brown rice in HeLa and AGS cancer cells (Table 2). On the other hand, the brown rice extract exhibited significantly lower IC_{50} value in Hep3B cancer cells than the Keunnunjami extract in both non-germinated and germinated group. The IC_{50} value for HT29 cancer cell was lowest after treatment with germinated brown rice extract and highest after treatment with non-germinated Keunnunjami extract. In general, germination substantially decreased the IC_{50} values of both brown rice and Keunnunjami extracts in all the cancer cells analyzed.

Body weight gain

The body weight gain and white adipose tissue weight significantly increased in HF mice compare to that of the control group (Table 3). However, oral administration of the rice extracts markedly decreased the gain in body weight and body fat in the high fat-fed mice, particularly the GK group. The feed intake and feed efficiency ratio were significantly higher in HF mice than the other animal groups. The HF mice also showed higher weights of liver and kidney than the control group. In comparison with HF mice, those fed with rice extracts, especially those from the GK group, showed a considerable decrease in liver and kidney weights.

Lipid peroxidation

The lipid peroxidation significantly increased in HF mice, manifested by the elevated levels of plasma and erythrocyte

Table 2. IC_{50} values of the germinated and non-germinated rice extracts in different human cancer cell lines

Cancer cell	Brown rice extract (μg/mL)		Keunnunjami rice extract (μg/mL)	
	Non-germinated	Germinated	Non-germinated	Germinated
HeLa[1)]	486.59 ± 2.68^{d}	387.36 ± 4.68^{c}	343.93 ± 2.87^{b}	275.70 ± 8.25^{a}
Hep3B	297.74 ± 5.87^{a}	310.09 ± 7.91^{a}	435.26 ± 4.25^{c}	365.57 ± 6.98^{b}
HT-29	532.31 ± 5.34^{c}	391.92 ± 5.68^{a}	561.39 ± 6.88^{d}	495.43 ± 8.25^{b}
AGS	463.16 ± 1.68^{d}	386.59 ± 2.57^{c}	372.83 ± 6.87^{b}	106.42 ± 5.77^{a}

Values are represented as means ± SE (n = 3).
[a-d] Means in the same row with different letters are significantly different at $P < 0.05$, as analyzed by Tukey's range test.
[1)] HeLa, cervical cancer cell; Hep3B, liver cancer cell; HT-29, colon cancer cell; AGS, gastric cancer cell

Table 3. Body weight gain and weights of adipose tissue and organs in high fat-fed mice orally administered with germinated and non-germinated rice extracts

Parameter	Animal group					
	C[1)]	HF	HF-B	HF-GB	HF-K	HF-GK
Initial body weight (g)	20.65 ± 0.12^{ns}	20.69 ± 0.16	20.75 ± 0.17	20.63 ± 0.31	20.58 ± 0.10	20.63 ± 0.07
Final body weight (g)	27.73 ± 0.32^{a}	40.58 ± 1.87^{d}	35.11 ± 0.76^{c}	33.00 ± 0.59^{c}	31.28 ± 0.33^{c}	29.21 ± 0.43^{b}
Body weight gain (g)	8.12 ± 0.43^{a}	19.57 ± 0.35^{d}	15.43 ± 0.41^{c}	13.81 ± 0.06^{bc}	11.74 ± 0.38^{bc}	9.43 ± 0.51^{b}
Feed intake (g/day)	2.82 ± 0.01^{a}	3.27 ± 0.01^{b}	2.94 ± 0.10^{a}	2.97 ± 0.06^{a}	2.86 ± 0.05^{a}	2.91 ± 0.10^{a}
Feed efficiency ratio	0.09 ± 0.002^{a}	0.18 ± 0.001^{d}	0.17 ± 0.001^{cd}	0.17 ± 0.001^{bc}	0.16 ± 0.001^{bc}	0.12 ± 0.001^{ab}
White adipose tissue (g)	1.98 ± 0.07^{a}	4.99 ± 0.10^{e}	3.79 ± 0.10^{d}	3.46 ± 0.05^{c}	3.22 ± 0.07^{c}	2.99 ± 0.04^{b}
Epididymal	0.92 ± 0.02^{a}	2.49 ± 0.16^{e}	2.01 ± 0.21^{d}	1.62 ± 0.15^{c}	1.58 ± 0.14^{c}	1.26 ± 0.09^{b}
Perirenal	0.53 ± 0.03^{a}	1.28 ± 0.09^{d}	1.01 ± 0.08^{c}	0.89 ± 0.03^{b}	0.84 ± 0.02^{b}	0.57 ± 0.02^{a}
Inguinal	0.49 ± 0.01^{a}	1.15 ± 0.04^{c}	0.99 ± 0.02^{b}	0.94 ± 0.03^{b}	0.92 ± 0.02^{b}	0.72 ± 0.02^{a}
Liver (g)	1.24 ± 0.01^{a}	1.50 ± 0.01^{d}	1.42 ± 0.01^{c}	1.36 ± 0.01^{bc}	1.35 ± 0.01^{bc}	1.32 ± 0.03^{b}
Heart (g)	0.15 ± 0.01^{a}	0.17 ± 0.01^{a}	0.16 ± 0.01^{a}	0.15 ± 0.02^{a}	0.15 ± 0.02^{a}	0.14 ± 0.01^{a}
Kidney (g)	0.35 ± 0.01^{a}	0.42 ± 0.02^{c}	0.38 ± 0.01^{b}	0.38 ± 0.01^{b}	0.38 ± 0.01^{b}	0.36 ± 0.01^{a}

Values are means ± SE (n = 8); NS, not significantly different
[a-d] Means in the same row with different letters are significantly different at $P < 0.05$, as analyzed by Tukey's range test.
[1)] C, AIN93G diet; HF, high-fat diet; B, HF + normal brown rice extract; GB, HF + germinated normal brown rice extract; K, HF + Keunnunjami rice extract; GK, HF + germinated Keunnunjami rice extract

Table 4. Plasma and erythrocyte lipid peroxidation in high fat-fed mice orally administered germinated and non-germinated rice extracts

Parameter	Animal group					
	C[1)]	HF	HF-B	HF-GB	HF-K	HF-GK
Plasma TBARS (nmol/mL)	5.81 ± 0.32[a]	9.89 ± 0.19[e]	8.82 ± 0.06[d]	7.96 ± 0.05[c]	8.14 ± 0.02[d]	7.29 ± 0.09[b]
Erythrocyte TBARS (nmol/g Hb)	8.27 ± 0.08[a]	12.54 ± 0.08[e]	11.42 ± 0.03[d]	10.93 ± 0.03[c]	10.84 ± 0.10[c]	9.32 ± 0.08[b]

Values are means ± SE (n = 8).
[a-e] Means in the same row with different letters are significantly different at $P < 0.05$, as analyzed by Tukey's Range Test.
[1)] C, AIN93G diet; HF, high-fat diet; B, HF + normal brown rice extract; GB, HF + germinated normal brown rice extract; K, HF + Keunnunjami rice extract; GK, HF + germinated Keunnunjami rice extract; TBARS, thiobarbituric acid reactive substance

Table 5. Antioxidant enzyme activity in high fat-fed mice orally administered with germinated and non-germinated rice extracts

Enzyme	Animal group					
	C[1)]	HF	HF-B	HF-GB	HF-K	HF-GK
Hepatic enzyme activity (nmol/min/mg protein)						
SOD[2)]	2.34 ± 0.05[e]	0.76 ± 0.02[a]	1.24 ± 0.02[b]	1.64 ± 0.02[c]	1.63 ± 0.01[c]	1.91 ± 0.04[d]
GSH-Px	5.30 ± 0.03[f]	3.52 ± 0.01[a]	3.93 ± 0.10[b]	4.22 ± 0.01[c]	4.34 ± 0.01[d]	4.89 ± 0.12[e]
CAT	1.49 ± 0.03[e]	0.81 ± 0.01[a]	0.94 ± 0.01[b]	1.13 ± 0.02[c]	1.08 ± 0.03[c]	1.26 ± 0.01[d]
PON	5.46 ± 0.04[f]	3.22 ± 0.01[a]	3.68 ± 0.04[b]	4.76 ± 0.02[c]	5.02 ± 0.12[d]	7.18 ± 0.06[e]
Nephritic enzyme activity (nmol/min/mg protein)						
SOD	1.81 ± 0.01[e]	1.05 ± 0.02[a]	1.34 ± 0.05[b]	1.28 ± 0.03[b]	1.40 ± 0.05[c]	1.69 ± 0.06[d]
GSH-Px	2.46 ± 0.02[e]	1.06 ± 0.03[a]	1.33 ± 0.02[b]	1.74 ± 0.04[c]	1.68 ± 0.03[c]	2.07 ± 0.01[d]
CAT	0.75 ± 0.02[e]	0.42 ± 0.01[a]	0.48 ± 0.01[b]	0.57 ± 0.01[c]	0.55 ± 0.01[c]	0.65 ± 0.02[d]
PON	0.63 ± 0.02[d]	0.41 ± 0.01[a]	0.47 ± 0.01[b]	0.53 ± 0.02[c]	0.56 ± 0.02[c]	0.61 ± 0.01[d]

Values are means ± SE (n = 8).
[a-e] Means in the same row with different letters are significantly different at $P < 0.05$ by Tukey's Range Test.
[1)] C, AIN93G diet; HF, high-fat diet; B, HF + normal brown rice extract; GB, HF + germinated normal brown rice extract; K, HF + Keunnunjami rice extract; GK, HF + germinated Keunnunjami rice extract
[2)] SOD, superoxide dismutase; GSH-Px, glutathione peroxidase; CAT, catalase; PON, paraoxonase

TBARS, relative to the control group (Table 4). However, addition of the rice extracts in the high-fat diet markedly decreased the TBARS content. Animals that were orally administered with germinated rice extracts showed considerably lower lipid peroxidation than those administered with non-germinated rice group. Between the two germinated rice extract-administered groups, the GK mice exhibited significantly lower TBARS level than the GB group.

Antioxidant enzyme activity

The activities of the hepatic and nephritic enzymes SOD, GSH-Px, CAT, and PON were lowest in HF mice and highest in the control group (Table 5). Compared with the HF group, all rice extract-administered groups showed significantly higher activities of the antioxidant enzymes. Among the groups administered with the rice extracts, the GK mice demonstrated the highest antioxidant enzyme activities, followed by the K and GB groups.

DISCUSSION

Here, we investigated the cytotoxic effects of the ethanolic extracts from germinated and non- germinated Keunnunjami, a pigmented giant embryo rice, and Ilpum, a non-pigmented normal embryo rice, against various cancer cell lines. Mice under high-fat diet regimen were orally administered the ethanolic extracts of these rice varieties to analyze the effects of the extract on the antioxidative defense system of animals. Results showed that germination significantly increased the antiproliferative activities of both the brown rice and Keunnunjami rice extracts. The germinated Keunnunjami extract exhibited greater antiproliferative effect against HeLa cervical cancer and AGS gastric cancer cell lines, as evident from its lower IC_{50} value, than the germinated brown rice extract. On the other hand, the germinated brown rice extract showed markedly higher anticancer activity against Hep3B liver cancer and HT29 colon cancer cell lines compared to germinated Keunnunjami extract. Oral administration of the rice extracts considerably decreased the high fat diet-induced body weight gain and TBARS level, an indicator of lipid peroxidation and oxidative stress in laboratory animals. Furthermore, the rice extracts substantially increased the activities of hepatic and nephritic antioxidant enzymes such as SOD, GSH-Px, CAT, and PON in obese mice. Among the groups fed with rice extracts, the GK group, exhibited the lowest TBARS content and highest antioxidant enzyme activities.

Keunnunjami rice extract contained higher amounts of GABA, tocopherol, phytic acid, and polyphenol than the ordinary brown rice extract. Germination further enhanced the levels of these bioactive compounds, especially GABA, in both rice samples. This elevation in the bioactive components is probably due to the breaking down of cell walls that surround various substances and activation of dormant enzymes during germination, which then leads to an increase in nutrients and generation of bioactive compounds in rice grains [11,12,25]. The GABA, tocopherol, phytic acid, and polyphenol have previously shown to have antiproliferative effects against different cancer cell lines such as liver, breast, ovarian, colon, lung, and gastric

cancer cells [26-29]. These bioactive compounds may have been partly responsible for the anticancer activities of the sample rice extracts. Moreover, Keunnunjami rice contains high levels of cyanidin-3-glucoside [1], an anthocyanin compound that have strong anticancer, anti-tumor, and antioxidant properties [30,31]. Past studies have also revealed that extracts from black and red rice have anticancer activities against breast and liver cancer cells due to high amounts of phytochemicals; in particular, an anthocyanin rich extract from black rice suppressed breast tumor (MDA-MB-453) growth [32,33]. Similarly, germinated brown rice extract with enhanced levels of bioactive compounds has reported to inhibit the development and proliferation of cancer cells [7,16]. Kim *et al.* [34] also accounted that germination for 4 days significantly increased the total polyphenol content and antiproliferative effect of rough rice on stomach and colon cancer cell lines.

Chronic consumption of a high fat diet promotes the formation of free radicals and reactive oxygen species, which then results in lipid peroxidation and oxidative stress [35]. The rice extracts were able to inhibit the high fat diet-induced lipid peroxidation, as evidenced by the lower levels of TBARS in B, GB, K, and GK groups relative to that of the HF mice. Moreover, the activities of the antioxidant enzymes were significantly higher in rice extract-administered groups compared to that of the HF mice. The antioxidant enzymes protect the cells from oxidative damage by catalyzing free radical-quenching reactions. For instance, superoxide radicals are converted by SOD enzyme into hydrogen peroxides, which are then degraded into non-toxic substances by GSH-Px and CAT enzymes [36]. The enzyme PON, on the other hand, hydrolyzes oxidized phospholipids and destroys lipid hydroperoxides [37]. This increase in the antioxidant enzyme activities and reduction in TBARS content suggest a significant improvement in the antioxidant defense system in mice orally administered with the rice extracts, making them less susceptible to oxidative damage caused by the high fat diet. The Keunnunjami rice extract showed greater antioxidative effect in mice than the ordinary brown rice. A previous study on Keunnunjami rice also showed that it has greater antioxidant capacity and higher phenolic and flavonoid contents than non-pigmented giant embryo brown rice [3]. Germination further enhanced the antioxidative property of both brown rice and Keunnunjami extracts which could be attributed to the increased amount of antioxidant compounds in the germinated grains. Dietary feeding of germinated non-pigmented giant embryo rice powder has also been shown to significantly lower lipid peroxidation and increase antioxidant enzyme activities in hypercholesterolemic rats, relative to those fed with non-germinated giant embryo rice and ordinary brown rice powders [38]. Also, increased phenolic compounds and anthocyanin contents from germinated rice were involved in the biosynthesis of proteins related to antioxidant activities [39]. Interestingly, the GB and K groups exhibited generally similar TBARS levels and antioxidant enzyme activities, suggesting that the non-germinated Keunnunjami extract has comparable antioxidant potential to germinated brown rice. This is probably due to the relatively high levels of polyphenol in Keunnunjami rice even in non-germinated form. Polyphenols are powerful antioxidant compounds that modulate oxidative stress through control of antioxidant enzymes [40,41]. In conclusion, ethanol extracts from Keunnunjami, a pigmented rice with giant embryo, have greater anticancer effect on cervical and gastric cancer cell lines compared to those from ordinary brown rice. On the other hand, the brown rice extract showed higher antiproliferative activity against liver and colon cancer cell lines than the Keunnunjami extract. Oral administration of the rice extracts in high fat-fed mice markedly decreased the lipid peroxidation and increased the activities of antioxidant enzymes. Germination for 72 h further enhanced these antiproliferative and antioxidative effects of both rice samples that may have been probably because of increased bioactive compounds. The germinated Keunnunjami extract exhibited greater antioxidative potential than the germinated brown rice extract. The results demonstrate that germination could be an effective method in enhancing the antiproliferative and antioxidant properties of Keunnunjami rice and that extracts from germinated Keunnunjami may potentially be useful as a natural therapeutic agent against cervical and gastric cancers and high fat diet-induced oxidative damage.

CONFLICT OF INTEREST

The authors declare no potential conflicts of interest.

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