Changes in the contents of prosapogenin in Red ginseng (*Panax ginseng*) depending on the extracting conditions

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**ABSTRACT**

This study compared the contents of prosapogenin depending on the extracting conditions of Red ginseng to provide basic information for developing Red ginseng-based functional foods. The content of ginsenoside Rg3 reached their maximum value at 24 h of extraction, followed by 36 h and 72 h of extraction at 100 °C.

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**Keywords:** conditions, extraction, ginsenoside Rg3, prosapogenin, Red ginseng

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Ginseng Radix (*Panax ginseng* Meyer), one of the important Oriental herbal medicines, has been used by people in Far Eastern countries to maintain their physical vitality for the past 2,000 y. *Shin-Nong-Bon-Cho-Kyung*, the oldest oriental medicine reference book, notes that ginseng can be used as a folk medicine to strengthen the activities of the five internal organs and vitalize stamina [1].

The extensive biological activities of ginseng were revealed through systematic pharmacological examinations of this plant including its effects on the cardiovascular system [2], immune system [3], and nervous system [4], along with its antidotal [5] and antidiabetic [6] functions and its effects as an antitumor agent or an antitumor adjuvant [7].

The main physiologically active substances of the ginseng are known to include ginsenosides, polyacetylenes, polysaccharides, ginseng proteins, and phenolic compounds [8–10].

The Shibata Group of University of Tokyo, Tokyo, Japan [9] has identified the chemical structure of ginsenoside and reported that ginsenosides exhibit a wide range of medical functions including anticancer, antioxidation, antifatigue, antistress, antiaging, and anti-inflammation activities. It prevents hardening of the arteries and hypertension, promotes liver functions, relieves hangover, and enhances memory. It is also effective in curing allergic disorders and promotes protein synthesis, among other things [8].

Red ginseng (*Ginseng Radix rubra*) in particular, a ginseng product variation prepared by steam-drying raw ginseng, which contains ginsenosides Rg2, Rg3, Rh1, and Rh2, is found to exhibit an anticancer function. It has been observed to suppress the growth of cancer cells [11,12], contain an antioxidant that helps lower blood pressure, protect brain cells [13], exhibit antiblood clot [14], and have antioxidation functions [11], thus making it pharmacologically superior.

In addition, high concentrated ginseng prosapogenin preparations are being developed using physical methods such as heat and pressure, and biochemical methods [15] using enzymes based on an artificial substance in the form of prosapogenin obtained by hydrolyzing ginseng saponin glucoside by heat, an ingredient unique to Red ginseng.

This study proposes to provide basic data required in developing highly concentrated, physiologically activated ingredients...
Ginsenosides composition of the concentrate was analyzed with HPLC, as suggested by Kim et al. [16]. The total ginsenoside content and ginsenoside composition of each sample were analyzed three times. The pure ginsenoside standards (99% pure) used in this experiment were purchased from Chromadex (Santa Ana, CA, USA) and Ambo Institute (Seoul, Korea).

The HPLC instrument used for the experiment was Waters 1525 binary HPLC system (Waters, Milford, MA, U.S.A.), and the column used was Eurospher 100-5 C18 (3 × 250 mm; Knauer, Berlin, Germany). The mobile phase was the mixture of acetonitrile (HPLC grade; J.T. Baker, Phillipsburg, NJ, USA) and distilled water (HPLC grade; J.T. Baker). The content of acetonitrile was sequentially increased from 17% to 25% (25 min), 30–40% (50 min), 40–60% (105 min), 60–100% (110 min), 100–100% (120 min), and finally adjusted from 100% to 17% (125 min, lasting for 10 min). The operating temperature was set at room temperature, and the flow rate was 0.8 mL/min. The elution profile on chromatogram was obtained using a UV/VIS detector at 203 nm (2487 dual λ absorbance detector; Waters).

Taking note of the fact that ginseng processing is no other than producing concentrated extracts, and that such trace ingredients as ginsenosides Rg3, Rg5, and Rk1 peculiar to Red ginsengs are produced [15] via hydrolysis by heat, the current study compared and analyzed the functional foods with Red ginseng roots by comparing and analyzing the contents of ginseng saponins depending on the time and temperature under which Red ginseng roots were extracted.

In this study, we used Red ginseng (Panax ginseng) purchased from Chung-buk-insam-chohap (Nam, Sung-Yop, President), Eumseong, Chung-cheong-buk-do, South Korea in May 2011. The product specimens were then kept at the Oriental Medical Food & Nutrition Research Laboratory, Semyung University, Jecheon, Korea (Fig. 1).

Fig. 1. Red ginseng (Panax ginseng).

## Table 1

| Ginsenosides | Red ginseng extract (% w/w) |
|--------------|-----------------------------|
|              | RGEE-12 | RGEE-24 | RGEE-36 | RGEE-48 | RGEE-60 | RGEE-72 |
| Rb1          | 1.905 ± 0.081 | 0.814 ± 0.005 | 0.285 ± 0.017 | 0.030 ± 0.003 | 0.200 ± 0.058 | 0 |
| Rb2          | 1.196 ± 0.049 | 0.516 ± 0.005 | 0.248 ± 0.015 | 0.037 ± 0.004 | 0.155 ± 0.039 | 0.173 ± 0.005 |
| Rd           | 0.459 ± 0.014 | 0.301 ± 0.005 | 0.173 ± 0.014 | 0.066 ± 0.002 | 0.132 ± 0.042 | 0.028 ± 0.002 |
| Re           | 0.419 ± 0.009 | 0.046 ± 0.005 | 0 | 0 | 0 | 0 |
| Rf           | 0.245 ± 0.021 | 0.181 ± 0.004 | 0.178 ± 0.016 | 0.131 ± 0.026 | 0.140 ± 0.031 | 0.099 ± 0.003 |
| Rg1          | 0.215 ± 0.010 | 0.033 ± 0.004 | 0 | 0 | 0 | 0 |
| Rg2          | 0.491 ± 0.023 | 0.610 ± 0.003 | 0.482 ± 0.028 | 0.320 ± 0.006 | 0.366 ± 0.096 | 0.273 ± 0.005 |
| (20S) Rg3    | 1.319 ± 0.031 | 2.666 ± 0.017 | 2.797 ± 0.148 | 2.283 ± 0.012 | 2.138 ± 0.611 | 2.572 ± 0.019 |
| (20R) Rg3    | 0.610 ± 0.024 | 0.503 ± 0.022 | 0.353 ± 0.019 | 0.208 ± 0.021 | 0.283 ± 0.124 | 0.253 ± 0.007 |
| Rg5          | 0.762 ± 0.047 | 1.614 ± 0.020 | 1.796 ± 0.111 | 1.505 ± 0.024 | 1.408 ± 0.373 | 1.743 ± 0.012 |
| Rg6          | 0.051 ± 0.003 | 0.077 ± 0.002 | 0.072 ± 0.005 | 0.060 ± 0.001 | 0.048 ± 0.028 | 0.069 ± 0.002 |
| Rc + Rk1     | 2.230 ± 0.083 | 1.476 ± 0.012 | 0.966 ± 0.060 | 0.561 ± 0.004 | 0.726 ± 0.191 | 0.509 ± 0.006 |
| Rh4          | 0.065 ± 0.005 | 0.089 ± 0.001 | 0.100 ± 0.006 | 0.082 ± 0.001 | 0.079 ± 0.020 | 0.098 ± 0.003 |
| Rk1          | 0.487 ± 0.030 | 0.103 ± 0.010 | 1.139 ± 0.069 | 0.937 ± 0.020 | 0.893 ± 0.233 | 1.070 ± 0.013 |
| Rk3          | 0.026 ± 0.005 | 0.033 ± 0.003 | 0.039 ± 0.000 | 0.031 ± 0.003 | 0.033 ± 0.007 | 0.038 ± 0.001 |
| F4           | 0.214 ± 0.008 | 0.320 ± 0.003 | 0.297 ± 0.019 | 0.232 ± 0.002 | 0.226 ± 0.064 | 0.261 ± 0.008 |
| (20S)Rg3 + (20R)Rg3 | 1.929 | 3.169 | 3.150 | 2.490 | 2.420 | 2.825 |
| Rk1 + Rg5    | 1.249 | 2.651 | 2.935 | 2.442 | 2.303 | 2.813 |
| Total saponin | 10.694 | 10.317 | 8.924 | 7.044 | 7.555 | 7.695 |

**Note:** Sum total of individual ginsenoside contents. Values represent the mean ± SE (n = 3).
analyzed the contents of ginseng saponin depending on the time taken to extract Red ginseng by means of the HPLC method (Table 1, Fig. 2). Ginsenoside content was analyzed to determine the reproducibility of the results by repeating the experiment three times for each sample compared to standards.

As shown in Table 1, a comparison and analysis of the contents of ginseng saponins hour by hour while extracting Red ginseng in distilled water (100°C) indicates that in the case of prosapogenin of the protopanaxadiol group, the content of (20S + 20R) ginsenoside Rg3 reached 3.169% when extracted for 24 h. The content increased according to the time of extraction that elapsed, reaching its maximum of 3.150% when extracted for 36 h, followed by 2.825% when extracted for 72 h.

At the same time, the content of ginsenosides Rg5 and Rk1 reached its peak of 2.935% when extracted for 36 h, followed by 2.813% when extracted for 72 h. Accordingly, our findings confirmed that the content of ginsenoside Rg3, known as a unique ingredient of Red ginseng [8], reached its maximum when extracted for 24 h, and in the case of ginsenosides Rg5 and Rk1, known as unique ingredients of Black ginseng [17], an extract containing a highly concentrated ginseng prosapogenin ingredient was obtained when extracted for 36 h. The study confirmed the development of a composition containing prosapogenin (ginsenosides Rg5 and Rk1) through the optimization of the extraction condition of Red ginseng. Prosapogenins are known to be good for the absorption of cells than the saponins present in the natural state, and to enhance pharmacological potency [18]. Accordingly, high function ginseng containing prosapogenin substances (ginsenoside Rg3, Rg5 and Rk1) can be produced by adjusting ginseng extracting conditions, expectedly making it possible to develop functional foods.

**Conflicts of interest**

The authors declare no conflicts of interest.
Acknowledgments

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