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Quantitative Analysis of Eight Compounds in Traditional Korean Medicine, Gongjindan Using HPLC, UPLC–MS/MS, and GC–MS/MS Systems

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Abstract: Gongjindan (GJD) is a traditional Korean medicine consisting of four herbal medicines and two animal-derived medicines, and is taken as a tonic in Republic of Korea. In this study, the goal was to develop and validate a simultaneous analytical method to quantify eight compounds in commercially available GJD samples using high-performance liquid chromatography (HPLC), ultra-performance liquid chromatography with tandem mass spectrometry (UPLC–MS/MS), and gas chromatography with tandem mass spectrometry (GC–MS/MS) systems. In HPLC and UPLC–MS/MS, seven components (gallic acid, 5-(hydroxymethyl)furfural, morroniside, loganin, nodakenin, decursin, and decursinol angelate) were separated and quantified using a distilled water–acetonitrile mobile phase system on a Capcell Pak UG80 C₁₈ column and an Acquity UPLC BEH C₁₈ column, respectively. Muscone was quantified using GC–MS/MS. The developed assays were validated by evaluating the linearity, limit of detection, limit of quantitation, recovery, and precision. In the regression equations of all compounds, the coefficient of determination was ≥ 0.9917 , showing good linearity. The recovery was 93.70–108.17%, and the relative standard deviation values in the precision test were all $<1.50\%$. Using the developed analysis methods, GJD samples were determined to contain the eight target compounds in concentrations from non-detected to 10.75 mg/g. The analytical assays developed and validated in this study can be used to obtain data for the quality control of commercially available GJDs and for the further expansion of efficacy and clinical studies.

Keywords: quantitative analysis; Gongjindan; HPLC; UPLC–MS/MS; GC–MS/MS



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1. Introduction

Herbal medicine preparations are usually composed of two or more types of herbal medicines, animal-derived medicines, and mineral medicines, and these types of medicines have long been used to treat and prevent various diseases, and to improve health. Gongjindan (GJD) is a traditional Korean medicine consisting of four herbal medicines (*Angelica gigas* Nakai, *Cornus officinalis* Siebold & Zucc., *Panax ginseng* C.A. Meyer, and *Rehmannia glutinosa* (Gaertn.) DC.) and two animal-derived medicines (*Moschus moschiferus* L. and *Cervus nippon* Temminck), and is one of the best-known tonics in Republic of Korea [1]. When GJD has been used in animal models, liver protection [1], antioxidant [2], neuroprotective [3], anti-fatigue [4], and Alzheimer’s-disease-protective [5] effects have been demonstrated.

Each of the herbal medicines that comprise GJD contain various ingredients, including coumarins (e.g., nodakenin, decursin, and decursinol angelate) from *A. gigas* [6], iridoids (e.g., loganin, morroniside, and cornuside) from *C. officinalis* [7], triterpenoid saponins (e.g., ginsenoside Rb₁ and ginsenoside Rg₁) from *P. ginseng* [8], miscellaneous compounds (e.g., 5-(hydroxymethyl)furfural; 5-HMF) from *R. glutinosa* [9], macrocyclic ketones (e.g., muscone and normuscone) and steroids (e.g., cholesterol, cholestane-3-ol, and lanosterol) from *M. moschiferus* [10–12], and peptidoglycans and gangliosides from *C. nippon* [13,14].

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