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Review article

A comparative study on chemical composition of total saponins extracted from fermented and white ginseng under the effect of macrophage phagocytotic function

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ABSTRACT

In this study, white ginseng was used as the raw material, which was fermented with *Paecilomyces hepiali* through solid culture medium, to produce ginsenosides with modified chemical composition. The characteristic chemical markers of the products thus produced were investigated using rapid resolution liquid chromatography coupled with quadrupole time-of-flight mass spectrometry (RRLC–QTOF–MS). Chemical profiling data were obtained, which were then subjected to multivariate statistical analysis for the systematic comparison of active ingredients in white ginseng and fermented ginseng to understand the beneficial properties of ginsenoside metabolites. In addition, the effects of these components on biological activity were investigated to understand the improvements in the phagocytic function of macrophages in zebrafish. According to the established RRLC–QTOF–MS chemical profiling, the contents in ginsenosides of high molecular weight, especially malonylated protopanaxadiol ginsenosides, were slightly reduced due to the fermentation, which were hydrolyzed into rare and minor ginsenosides. Moreover, the facilitation of macrophage phagocytic function in zebrafish following treatment with different ginseng extracts confirmed that the fermented ginseng is superior to white ginseng. Our results prove that there is a profound change in chemical constituents of ginsenosides during the fermentation process, which has a significant effect on the biological activity of these compounds.

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1. Introduction

Ginseng is a perennial herb that contains various bioactive substances, belonging to the genus *Panax* of the family Araliaceae [1]. It is considered an adaptogenic herb, which is used to enhance immunity, radioresistance, and the capability to cope with tumor and many other diseases [2,3]. Under most circumstances, ginsenosides are used as the main active ingredient during the treatment of various diseases by oral administration [4]. However, ginsenosides have poor water solubility and low bioavailability, which greatly affect their pharmacological action [5]. As a result, numerous studies have been conducted to modify the structural composition of ginsenosides to improve their activity, bioavailability, and their composition in every relevant aspect (e.g., chemical hydrolysis, biological conversion, hot working, semisynthesis) [6–8]. For example, hot processing of red ginseng produces a special ingredient of ginsenosides (Rg3, Rh2, and CK) that has a high development value and bright future for use in various clinical applications, due to its effect in reducing blood fat, antitumor activity, enhancing immune system as well as protecting the heart, liver, and blood vessels [9]. Fermented ginseng leavened by microbial actions has become a hot topic of research recently, and has been applied extensively in functional foods such as distiller's yeast-fermented ginseng wine and probiotics-fermented ginseng beverage [10,11]. Fermented ginseng is usually processed by methods such as mild acid hydrolysis, microbial conversion, or enzyme conversion, through which major ginsenosides would be transformed into minor ones [12]. Many studies have shown that the multiple probiotics in fermented ginseng change the chemical compositions of rare ginsenosides through biological transformation. Rae et al [13] confirmed that Korean ginseng fermented with mushroom mycelium bred by solid culture contained significantly more crude fat (4.66–12.02%) than raw Korean ginseng (white ginseng; 1.61%). Nevertheless, the effects of fermented

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ginseng on mitogenic activity were much greater than those of white ginseng.

In addition, some studies have indicated that the concentrations of total ginsenosides in red ginseng leavened by *Phellinus linteus*, *Cordyceps militaris*, and *Grifola frondosa* were particularly at higher levels in comparison with those in red ginseng [14,15]. However, the effect brought about by the biotransformation of ginsenosides and whether they had an impact on the functions of the fermented ginseng as well as the difference between fermented ginseng and white ginseng had been reported so far. Therefore, it is crucial to identify the chemical components in ginseng following fermentation with different forms of microorganism as well as consider the differences between fermented ginseng and white ginseng with respect to "chemical markers" to understand the chemical basis for their various pharmacological activities. Ginsenosides have an important pharmacological activity by enhancing immunity against potential pathogens or harmful bacteria [16]. Most currently used methods were confined to the determination of a few marker constituents and there are no systematical studies on the immunity effects that interfere with the content of ginsenosides of *Panax ginseng* treated by different methods, especially those in fermented ginseng and white ginseng.

In recent years, zebrafish has been used as an animal model to study the effects of various compounds on behavior, immunity, toxicity, etc. [17]. Besides, it has been one of the best animal models to study the effects of novel drugs on immune function, especially the function of macrophage. Because macrophages are important cells in immunological system that perform many functions such as presenting antigen, eliminating foreign body, and secreting various cytokines [18], they play an important role in specific cellular immunity and nonspecific immunity. Therefore, to evaluate the effect of various components on immunity, we investigated the improvement in the phagocytic function of macrophages in zebrafish. In this study, white ginseng was used as the raw material, and was fermented with *Paecilomyces hepiali* (China Center of Industrial Culture Collection, Beijing, China) through a solid culture medium, to produce ginsenosides with modified chemical composition. A rapid resolution liquid chromatography coupled with quadrupole time-of-flight mass spectrometry (RRLC–QTOF–MS) method has been developed for the identification of ginsenosides produced. Chemical profiling of white ginseng and fermented ginseng has been established and in combination with multivariate statistical analysis has been used for the systematic comparison of active ingredients in white ginseng and the fermented ginseng to understand the beneficial properties of ginsenoside metabolites. Therefore, the former steps would provide a research basis for further study as well as promote the development of ginseng product with an emphasis on its effects on the improvement of the phagocytic function of macrophages in zebrafish.

1.1. Experimental analysis

1.1.1. Chemicals and reagents

A total of 18 ginsenoside standards were obtained from Jilin University, Changchun, China: Rb1, Rb2, Rb3, Rc, Re, Rg1, Rf, Rd, Ro, 20(S)-Rg3, 20(R)-Rg3, 20(S)-Rg2, 20(R)-Rg2,20(S)-Rh1, 20(R)-Rh1, Rh2, Rk1, and Rg5 (Jilin Food And Drug Administration, Changchun, Jilin, China). All other chemicals were of reagent grade and obtained from local suppliers (TAKALA Co., Changchun Branch, Changchun, Jilin, China). The standard stock solutions of ginsenosides were prepared independently by dissolving 1 mg of the standard into 5-mL aqueous methanol, where the purification of methanol was approximately 80% (methanol:water, 80:20, v/v), to achieve a concentration of 0.2 mg/mL. Mixed standard solutions were prepared by combining the aliquots of each set of individual stock solutions and diluting them to the appropriate concentration.

1.2. Plant material and sample preparation

All the root samples from Ji'an (Jilin province) were harvested in fall and collected at the 5th year of their growth phase. White ginseng was dried at 42°C to achieve a constant weight. Strains of *P. hepiali* maintained on potato dextrose agar slants were obtained from China Center of Industrial Culture Collection, and the seed liquid was cultivated according to methods described previously [19]. The optimal conditions for fermentation, presented as follows, were created through single factors and orthogonal experiments. The optimal acetic fermentation conditions were set as follows: ginseng power (45 mesh), 10%; solid-to-liquid ratio, 2–3. For *P. hepiali*, the seed liquid was cultivated for 14 h with an inoculation amount of 10% at a culture temperature of approximately 26°C. The first batch of samples was collected and analyzed after 24 h of fermentation, and the process was continued for up to 96 h after every 24-h collection under the condition of a natural pH value. When fermentation is completed, the culture was dried.

Samples for RRLC–QTOF–MS analysis of white ginseng and fermented ginseng were grinded into powder. Each replicate contained 10 individual ginseng samples. Every 1.0-g powdered ginseng sample was weighed accurately and immersed in aqueous methanol at a concentration of 80% (methanol:water, 80:20, v/v) to be extracted in an ultrasonic bath at 20°C for 30 min. The extraction process was repeated three times using fresh aliquots of the solvent. When the combination of the three aliquots was achieved, the solution was centrifuged at 13,000*g* for 5 min, after which the supernatant was passed through a 0.45-μm filter. This process is subsequently followed by the RRLC–QTOF–MS analysis. All the aforementioned solutions were kept at 4°C during storage and brought to room temperature before utilization.

1.3. Detection conditions of RRLC–QTOF–MS

The extracts were qualified using the Agilent 1260 LC system using a Thermo Scientific Hypersil GOLD C_{18} column (3.0 × 100 mm × 1.8 μm), and the mobile phase consisted of water with 0.1% formic acid (A) and 100% acetonitrile (B). In brief, solutions were treated as follows: 0.00–3 min, 25–30% B; 3–5 min, 30–36% B; 5–9 min, 36–40% B; 9–12 min, 40–45% B; 12–13 min, 45–50% B; 13–14 min, 50–65% B; and 14–15 min 65–90% B. The flow rate was 0.3 mL/min and the volume of the injected sample was 10 μL. The column and sample managers were maintained at 30°C, respectively, and the peak was monitored at 203 nm.

The mass spectrometer was operated in the negative ion mode and the total ion chromatogram mode. The MS analysis conditions were as follows: desolvation gas flow, 600 L/h; desolvation temperature, 250°C; capillary voltage, 2,500 V; cone voltage, 35 V; and scan range, m/z 100–1,500. The fragment ions were obtained using collision energy of 35% through both MS2 and MS3 experiments.

1.4. Multivariate statistical analysis

With the Agilent Mass Hunter Workstation Software-Qualitative Analysis and Q-TOF Quantitative Analysis (Version B.04.00, Build 4.0.479.5, Service Pack 3, and Agilent Technologies, Inc. 2011), all HPLC–QTOF–MS/MS data were fully recorded to assess the quality comprehensively and explore the discriminated variables of fermented ginseng and white ginseng. Peak integration was calculated automatically and supplemented with manual operation. Components of different samples appeared the same when they showed similar retention times with a tolerance of 0.2 min, accurate mass

weights with the tolerance of 0.05 Da, and ion fragments. No specific mass or adduct was excluded except reagent used. The ion intensity provided sample codes that were aligned in a table and corrected manually. The table was then imported into SIMCA-P Software (Version 13.0.3). Peak intensities were treated as *X* variables, whereas the sums of the peak intensities were taken as *Y* variables. All the variables were normalized before principal component analysis and orthogonal partial least-squares discriminant analysis (OPLS-DA).

1.5. Ascertainment of maximum tolerable concentration of drug

The samples of zebrafish were placed in six-pore plates with ginseng extract concentrations of 62.5 μg/mL, 125 μg/mL, 250 μg/mL, 500 μg/mL, and 1,000 μg/mL, respectively, administered to each fish. Meanwhile, the control group was prepared in which the zebrafish will be treated with water. Each control group had 30 samples. The zebrafish was observed daily and the dead ones were removed each day. Subsequently, the number and toxicity of extracts to zebrafish in each control group were statistically analyzed after disposal to determine the maximum tolerable concentration of ginseng extract.

1.6. Establishment of animal models

Nano-activated carbon ($PM_{2.5}$) was injected at a concentration of 2.3 mg/mL into the veins of 2 dpf zebrafish (equivalent to human IV) with 10 nL for each fish. This step refers to the establishment of the zebrafish $PM_{2.5}$ phagocytosis model at the dose of 23 ng/zebrafish.

1.7. Facilitation of macrophage phagocytic function following treatment with different ginseng extracts

The concentration of phagocytosis function test is confirmed. According to the experimental results of maximum tolerance concentration, this was set at the medium value (166.7 μg/mL) for the normal control and modeling control groups adapted at the same time. After completion of all these procedures, the zebrafish is stained with neutral red solution; then, the macrophage phagocytosis facilitative rate determined by calculating the number of phagocytosed macrophages (*N*) was used to evaluate the effects of different ginseng extracts on the macrophages.

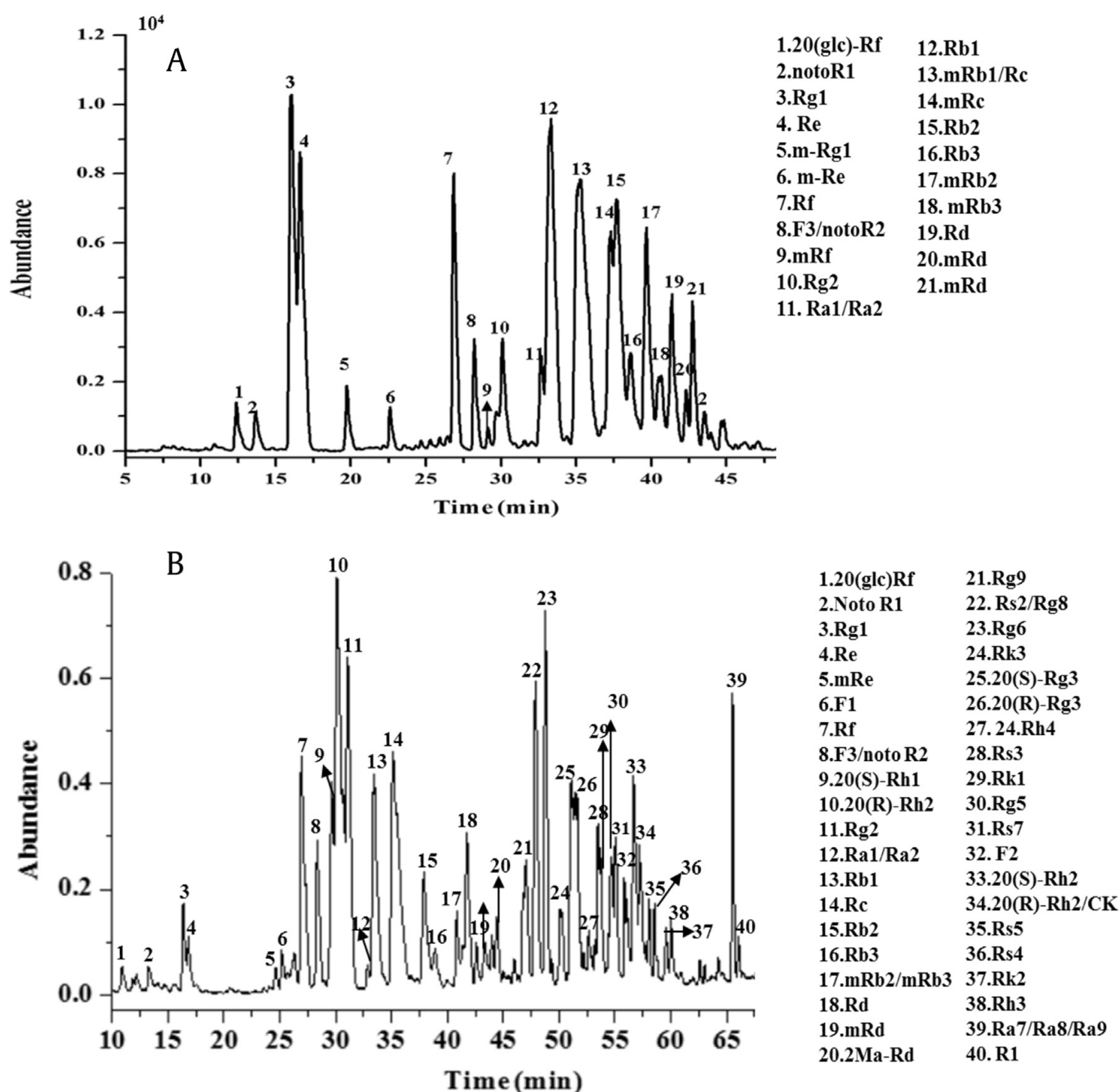

Fig. 1. Rapid resolution liquid chromatography coupled with quadrupole time-of-flight mass spectrometry total ion chromatogram of the extract of different kinds of ginseng acquired in the negative ion mode. (A) White ginseng; (B) fermented ginseng.

2. Results and Discussion

2.1. RRLC–QTOF–MS analysis of fermented ginseng processing product

All extracts of the samples were analyzed with the RRLC–QTOF–MS method. The typical profiles of different kinds of ginseng are shown in Fig. 1, according to which, the fermented ginseng contains more diverse ginsenoside compounds than white ginseng, with approximately 40 ginsenosides detected in fermented ginseng. Among the ginsenosides identified in fermented ginseng, 18 components were unambiguously authenticated as ginsenosides Rb1, Rb2, Rb3, Rc, Re, Rg1, Rf, Rd, Ro, 20(S)-Rg3, 20(R)-Rg3, 20(S)-Rg2, 20(R)-Rg2,20(S)-Rh1, 20(R)-Rh1, Rh2, Rk1, and Rg5 by comparing the retention times, m/z values, and fragment ions with those of the reference compounds, and 24 constituents were tentatively identified by analyzing specific MS/MS fragment ions, accurate mass, and isotopic ratio patterns. In addition, in MS/MS spectra, the glycoside bond of saponin components was broken by the collision-induced dissociation method to generate characteristic ions for further confirmation of the molecular structure. Fig. 2 shows the MS/MS spectra of representative types of ginsenoside. For example, Peak 1 under first mass spectrometry data showed the following: $[M-H]^-$ m/z for 799.4799, adduct ions $[M+CH_3COO]^-$ m/z for 845.6031, and the characteristic of MS/MS containing characteristic fragment ion m/z 475.3787, which indicated that this chemical compound belonged to the PPT group. The corresponding fragment ion originated from the break of the glycoside bond and included [M-H-Glc]-, [M-H-GlcGlc]-, [M-H-GlcGlc-C_6H_{12}]-, [GlcGlc-H_2O-H]-, [Glc-H_2O]-, [2.5A1β], m/z 391.2866 for [M-H-XlyGlc-C_6H_{12}]-, and m/z 161.0354 for [Glc-H_2O]- (Fig. 2A). Its elementary composition ($C_{42}H_{72}O_{14}$) was obtained using the MassLynx 4.2 Software. Built on the MS fragment pathways and structural composition of saponin, it could be inferred that Peak 1 refers to 20(Glc) -Rf. It can also be deduced that Peak 2 was NotoR1, which forms characteristic fragment ions such as [2GlcXly-H]- (m/z 475.3588), [Glc-H]-(m/z 179.0504), and others (Fig. 2B). In this experiment, the variational properties of ginsenosides in fermented ginseng could be identified as markers by the LC-MS scan mode and the selective ion mode.

2.2. RRLC–QTOF–MS-based chemical profiling coupled with multivariate statistical analysis of ginseng

The related data were processed by unsupervised principal component analysis and supervised OPLS-DA with SIMCA-P Software and used to study the differences between fermented ginseng and white ginseng, and to further explore their potential characteristic markers. From the result of analytical methods, we could observe that white ginseng and fermented ginseng were clustered into two groups (Fig. 3), which indicated that the metabolite profiles of the two materials were distinct, and different samples of

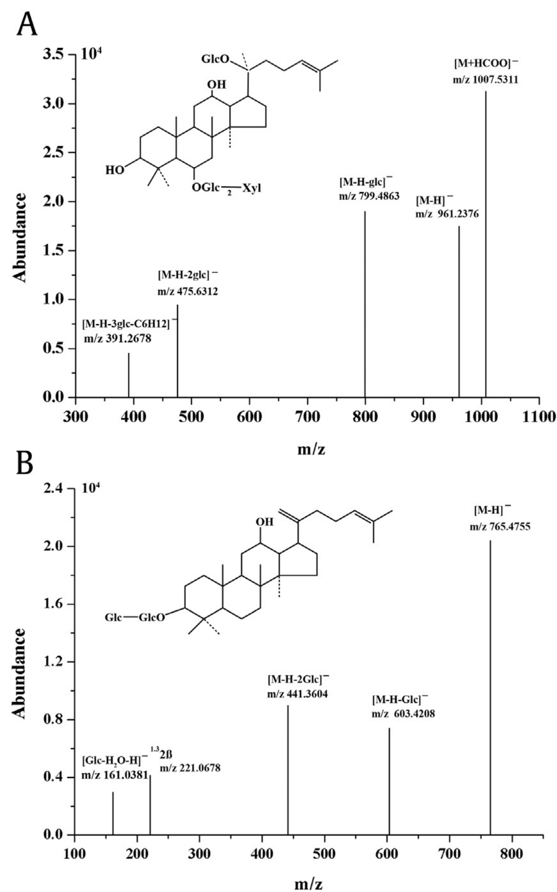

Fig. 2. Electrospray ionization–quadrupole time-of-flight–MS/MS spectra of ginsenosides. The mass spectrometer was operated in the negative ion mode and the total ion chromatogram mode. The fragment ions were obtained using collision energy of 35% through both MS^2 and MS^3 experiments. (A) 20(Glc)-Rf; (B) Rk1.

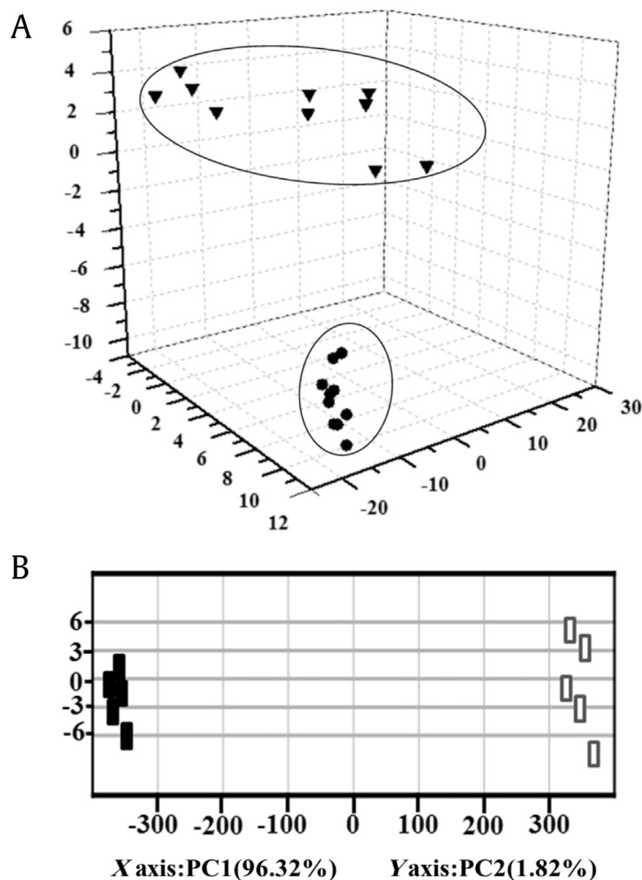

Fig. 3. (A) Principle component analysis scores plot of extracts from fermented ginseng and white ginseng, respectively. Using white ginseng as the raw material, various ginseng products were produced by fermentation with *Paecilomyces hepiali* through solid culture. The extracts of fermented ginseng are represented by closed triangles, whereas those of white ginseng are represented by closed circles. (B) Orthogonal partial least-squares discriminant analysis of white ginseng and fermented ginseng.

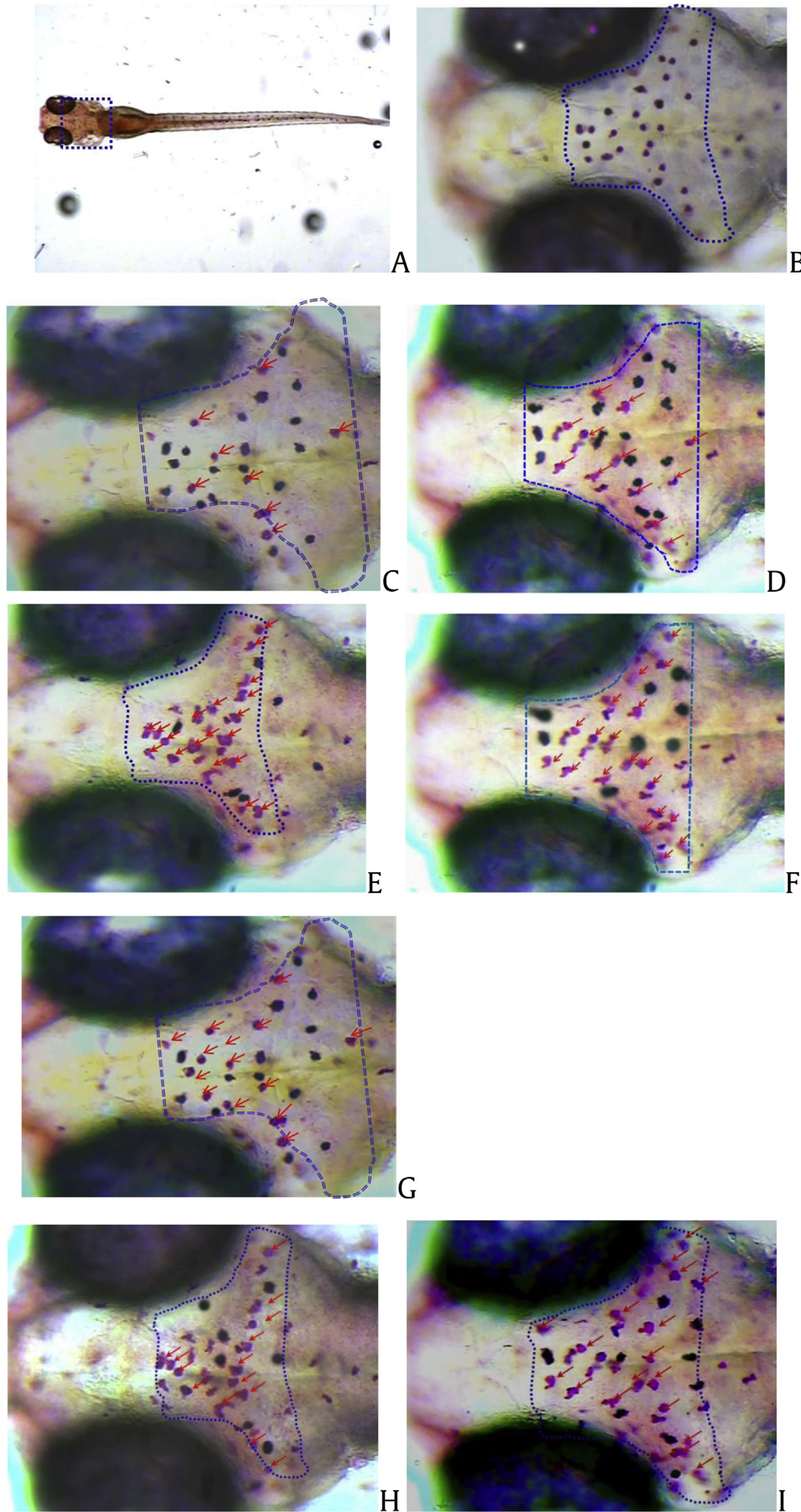

Fig. 4. The phenotype map of macrophages engulfing nano-activated carbon. The part painted with blue was used as the observation site, and the red arrows represent macrophage engulfing nano-activated carbon. (A) The whole zebrafish. (B) The normal control group. (C) The control model group. (D–F) The test group with the medium dosage of extract from fermented ginseng with doses of 55.6 μg/mL, 166.7 μg/mL, and 500 μg/mL, respectively. (G–I) The test group with the medium dosage of extract from white ginseng with doses of 55.6 μg/mL, 166.7 μg/mL, and 500 μg/mL, respectively.

each material were consistent with each other. The following S-plot generated by OPLS-DA and presented in the no S-form indicated that the synactic multicomponents were responsible for the chemical differences between the two groups rather than only a few characteristic constituents (Fig. 4). Some components, namely, malonyl-Rb1/Rc (Peak 13), malonyl-Rc (Peak 14), malonyl-Rb2/Rb3 (Peaks 17 and 18), malonyl-Rd isomer (Peaks 21 and 22), Ra1/Ra2 (Peak 11), and malonyl-Rg1 (Peak 5), are abundantly present in white ginseng, whereas the same components could hardly be found in the fermented product. At the same time, some components such as ginsenosides F1 isomer (Peak 6), 20(S)-Rh1/20(R)-Rh1 isomer (Peaks 9 and 10), Rg9 (Peak 21), Rs2/Rg8 (Peak 22), Rg6 isomer (Peak 23), Rk3 isomer (Peak 24), 20 (S)-Rg3/20(R)-Rg3 (Peaks 25 and 26), Rh4 (Peak 27), Rs3 (Peak 28), Rk1 (Peak 29), Rg5 (Peak 30), and the other 10 components [Rs7, F2, 20(S)-Rh2, 20(R)-Rh2/CK, Rs5, Rs4, Rk2, Rh3, Ra7/Ra8/Ra9, and R1] were prominent in the fermented product rather than in the white ginseng. The concentrations of these core components were obtained by calculating the peak area ratios of individual ginsenoside and were totally dependent on the RRLC–QTOF–MS chemical profiling data of white ginseng and fermented ginseng mentioned earlier.

As shown in Table 1, the contents of four protopanaxadiol ginsenosides (Rb1, Rc, Rb2, and Rb3) in fermented ginseng were higher than those in white ginseng on average. Besides, as same as the average content of five malonylated protopanaxadiol ginsenosides (mRb1, mRc, mRb2/mRb3, and mRd), the content of protopanaxatriol ginsenosides (Rg1, Re, and Rf) was much lower than those in white ginseng on average. It seems that malonylated protopanaxadiol ginsenosides (mRb1, mRc, mRb2, and mRd), which are abundant in white ginseng, are more likely to be transformed into small molecular weight and rare ginsenosides (Rg8, Rg9, Rh3, etc.). These changes in ginsenosides during fermentation were similar to those observed during red ginseng processing. For example, hydrolysis of glycosidic bond at C-(20) and isomerous reaction of C-(20) are the main reactions of dammarane-type ginsenosides. The transformation in the PPD group maybe that ginsenosides Rb1, Rc, and Rb2 hydrolase could only hydrolyze the glycosidic bond at the C-20 position into ginsenoside Rd. However, upon further hydrolysis of ginsenoside Rd into Rg3, Rg3 removed the C-(20) hydroxyl group, formed a double bond, and produced ginsenoside Rk1 or Rg5. However, some unique changes were noted in our study. For example, the content of ginsenoside Rd in fermented ginseng was 1.5 times higher than that in white ginseng. Many studies have demonstrated that the ginsenoside Rd in red ginseng is reduced during processing [20]. However, further exploration and research concerning transformation mechanism are needed.

Table 1
Relative content of ginsenosides of PPD, PPT, malonyl, and transformed types in white and fermented ginsengs

Group	Ginsenosides	White ginseng	Fermented ginseng
PPD group	Ra1	1.86%	0.43%
	Ra2	1.99%	0.32%
	Rb1	10.23%	4.47%
	Rb2	5.96%	3.04%
	Rb3	4.76%	2.43%
	Rc	5.81%	1.34%
	Rd	2.42%	3.63%
	Rg3	1.45%	7.39%
PPT group	Rg1	10.69%	4.78%
	Re	5.78%	3.54%
	Rf	6.14%	3.32%
	Noto-R1	1.44%	0.72%
	20(glc)-Rf	0.68%	0.43%
	F3/NotoR2	1.05%	1.91%
	20(s)-Rg2	2.47%	6.92%
Malyonyl	mRb1	6.31%	0
	mRb2	7.43%	0
	mRb3	2.23%	0
	mRc	4.68%	0
	mRd	3.56%	0.83%
	mRe	1.51%	0.3%
	mRg1	2.80%	0
	2MRd	3.01%	1.09%
Transformed	Rg6	0	2.47%
	Rh4	0	0.87%
	Rs2/Rg8	0	3.47%
	20(R)-Rh2/CK	0	2.24%
	20(R)Rg3	0	4.73%
	20(R)Rg2	0	2.74%
	Rk1	0	3.68%
	Rg5	0	3.12%
	Rh3	0	2.27%
	Rh1	0	6.78%
	F1	0	0.79%
	Rs3	0	2.67%
	R1	0	1.43%
	Ra1/Ra8/Ra9	0	2.74%
	Rs7	0	3.14%
	Rk2	0	1.07%
	F2	0	2.14%
	Rg9	0	3.01%

PPD: protopanaxadiol ginsenosides; PPT: protopanaxatriol ginsenosides.

2.3. *Comparison of the facilitation of macrophage phagocytic function following treatment with different ginseng extracts*

To understand the effect of these components on enhancing immunity, we compared the facilitation of macrophage phagocytic function following treatment with different ginseng extracts. According to the model characteristics, the zebrafish brain is used to observe the function of macrophage phagocytosis. To fulfill this, total ginsenosides were calculated as the sum of ginsenoside fractions. None of the zebrafish died in any of the control groups at concentrations of 62.5 μg/mL, 125 μg/mL, 250 μg/mL, and 500 μg/mL, whereas all zebrafish died in the group in which the set point of concentration was 1,000 μg/mL. Therefore, the maximum tolerable concentration for zebrafish from the ginseng extract was determined to be 500 μg/mL based on these results.

The numbers of phagocytosed macrophages for activated carbon nanoparticles are 13, 15, and 20 in the control group with the concentration of white ginseng at 55.6 μg/mL, 166.7 μg/mL, and 500 μg/mL, and comparing these values with those in the modeling control group (p <0.001), the rates of facilitated function for phagocytosis of zebrafish macrophages are 85.7%, 114.3%, and 185.7%, respectively. The numbers of phagocytosed macrophages for activated carbon nanoparticles are 14, 18, and 23 in the control group with the concentrations of fermented ginseng at 55.6 μg/mL, 115.7 μg/mL, and 500 μg/mL, which present a contrasting example with those in the modeling control group ($p < 0.001$). Furthermore, the rates of facilitated function for phagocytosis of zebrafish macrophages are 100%, 157.1%, and 228.7%, which are in contrast to those of the low concentration group (55.6 μg/mL, $p < 0.001$), in which the white ginseng is used (Table 2). Besides, significant differences exist in both high (500 μg/mL) and medium concentration groups (166.7 μg/mL; Fig. 5). Different kinds of ginseng extract could effectively facilitate the macrophage phagocytosis of zebrafish and show a concentration correlation; however, the facilitated function of fermented ginseng is better than that of white ginseng. It is further confirmed that the rare ginsenoside generated during the fermentation process helps make the phagocytosis of macrophage in fermented ginseng more effective compared with that in white ginseng.

Several studies have shown that Rg3 strengthens the body's response to restrain cancer cell by making the immune system

Table 2
The quantitative result of the phagocytic function of macrophages in zebrafish brain ($n = 10$)

Group	Concentration (μg/mL)	The number of macrophages engulfed the nano-activated carbon (mean ± standard devaition)	Macrophage phagocytic rate (%)
Normal control	—	0	—
Model control	—	7 ± 1	—
Fermented ginseng	55.6	13 ± 1 *	85.7 *
	166.7	15 ± 1 *	114.3 *
	500	20 ± 1 *	185.7 *
White ginseng	55.6	14 ± 1 *	100 *
	166.7	18 ± 1 *	157.1 *
	500	23 ± 1 *	228.7 *

* $p < 0.001$ (compared with the model control group).

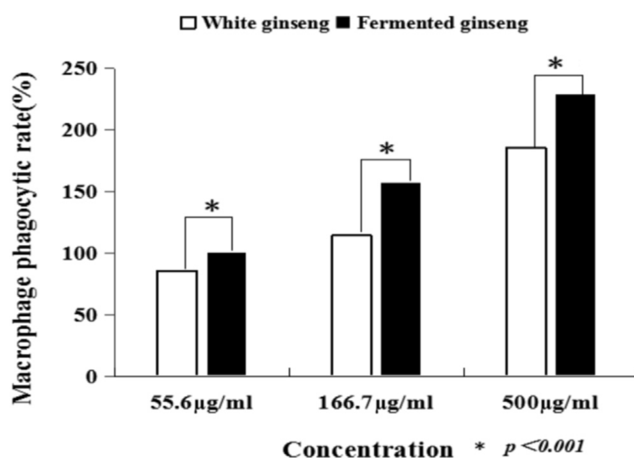

Fig. 5. The macrophage phagocytic rate of white ginseng and fermented ginseng. The macrophage phagocytic rate was calculated by the ratio of the number of macrophages swallowing nano-activated carbon in the test group and the model control group. Data are depicted as the mean ± standard deviation in the rate of macrophage phagocytic function between the two groups ($\bar{x} \pm s$, $n = 10$). indicate differences versus in the model group and p values indicate differences between the group of white ginseng and fermented ginseng. *$p < 0.001$.

more effective. Rg3 is gradually used in adjunctive therapy and provides a new pathway for cancer therapy. Our results show that the content of Rg3 is increased during hydrolysis through fermentation. Therefore, our results provide a novel way to improve Rg3 production by microbial fermentation. Our results also have important implications in medical research and improve our understanding of the reference values to be used while adapting fermented ginseng in medical applications.

3. Conclusions

In this study, we intended to confirm the fermentation process with *P. hepiali* obtained by solid culture, and measure ginsenosides composition using RRLC–QTOF–MS chemical profiling of white ginseng and fermented ginseng without microbial strains. A total of 40 ginsenosides were identified and the specific chemical components in fermented ginseng were elucidated approximately. The chemical compositions and the ratio of the component of ginsenosides differed widely between the fermented ginseng and the white ginseng, suggesting that the total amount of saponins in ginseng has generated rare and small molecular weight ginsenosides through unique biological fermentation. The comparison results of the facilitation of macrophage phagocytic function in zebrafish following treatment with different ginseng extracts indicate that fermented ginseng is superior to white ginseng. Fermented processing may well become a research hot spot, opening a new avenue for ginseng development in the future.

Conflicts of interest

The authors have no conflicts of interest to declare.

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