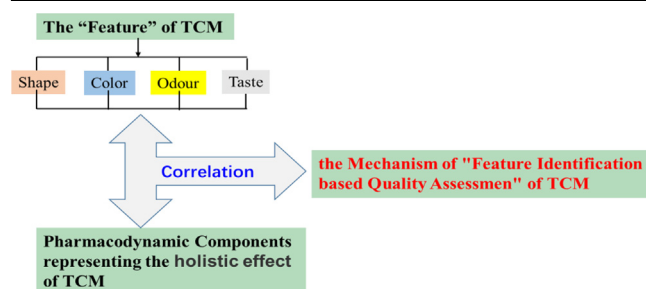




Research article

A new method for studying the mechanism of “Feature Identification based quality assessment” of Traditional Chinese Medicine, taking *Gastrodiae Rhizoma* as an exampleWang Bing^{a,1}, Sun Yan-tao^{a,1}, Pei Wen-han^b, Zhang Hui^{a,*}, Kang Ting-guo^{a,**}^a College of Pharmacy, Liaoning University of Traditional Chinese Medicine, Dalian, 116600, China^b Macau University of Science and Technology, Macau, 999078, China

GRAPHICAL ABSTRACT



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ABSTRACT

Aim of the study: The research group proposed that the mechanism of “Feature Identification based Quality Assessment” (FIQA) of Traditional Chinese Medicine (TCM) can be explained according to the relationship between the “Feature” of TCM and the Pharmacodynamic Components representing the holistic effect of TCM. *Gastrodiae Rhizoma* (GR) was selected as the research object to reveal the close relationship between “Feature” and the quality of TCM.

Materials and methods: In this study, the “Feature” such as “Shape”, “Color”, “Odour” and “Taste” of GR are quantified by the electronic nose, electronic tongue, and other instruments. Then, the Pharmacodynamic Components Group (PCG) of GR was determined which could reflect the holistic effect of GR by spectrum effect relationship analysis. By analyzing the correlation between the “Feature” and the content of PCG, the mechanism of FIQA of GR was determined.

Results: The quantitative results of “Shape”, “Color”, “Odour” and “Taste” of GR from different sources were significantly different. Six components were selected as the PCG, which can represent the holistic effect of GR in the aspect of the neuroprotective effect. There was a good correlation between the components in the PCG and “Feature”.

Conclusions: The quality of GR can be determined quantitatively according to its “Shape”, “Color”, “Odour” and “Taste”. The mechanism of FIQA of TCM can be explained according to the relationship between the Shape of TCM and the Pharmacodynamic Components representing the quality of TCM. The revelation of this mechanism reflects the holistic characteristics of the multi-component synergistic effect of TCM. This study provides a reference research method for revealing the mechanism of FIQA of TCM.

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1. Introduction

“Feature Identification based Quality Assessment” (FIQA) [1] is the traditional experience identification theory of Traditional Chinese Medicine (TCM) varieties. It is to judge the quality of TCM according to its “Feature”, such as “Shape”, “Color”, “Odour” and “Taste”, and to clarify the essence of its quality. It is the basic basis for evaluating the quality of medicinal materials in the current Chinese medicinal material market. However, there are few reports on the internal mechanism of FIQA, and FIQA has not been determined as the standard of quality identification of TCM by legal institutions, the fundamental reason is that its scientificity and mechanism still need to be studied.

Through literature review, it can be seen that the rationality of FIQA is explained by the correlation between appearance and the main chemical components of medicinal materials [2, 3]. However, the selection of its chemical components does not represent the overall efficacy of TCM and does not reflect the overall characteristics of the multi-component synergistic effect of TCM. The “Shape” in the theory of FIQA of TCM only includes macroscopic external characters. However, the research team's research on 20 aftertastes TCM [4, 5] found that the microscopic characteristics of TCM, such as the pollen grains of *Polygonum Multiflorum* [6], *Farfarae Flos*, and stone cells of *Arctium Lappa* [7], can also evaluate the quality of TCM [8]. So this team gives a new meaning to “Feature” in the theory of FIQA, that is, “Shape” includes macroscopic and microscopic characteristics. The screening of active ingredients that can represent the overall characteristics of the multi-component synergistic effect of TCM is the key to explaining the mechanism of FIQA. By analyzing the relationship between the “Feature” of TCM and the active components that represent the holistic effect of TCM, we can explain the essence of “FIQA”.

Gastrodiae Rhizoma (GR), an Orchidaceae plant, was selected as the research object in this experiment. GR is the dry tuber of the orchid plant *Gastrodia elata* BL. The Compendium of Materia Medica records that GR is mainly used to treat rheumatism, arthralgia, limb contracture, paralysis, dizziness, and headache. GR has been used as an important medicine for headache and dizziness by doctors of all ages. It has a significant effect on central nervous system and cardiovascular system diseases. Modern pharmacological studies have confirmed that its clinical efficacy is mainly related to the protection of nerve injury [9, 10]. The traditional standard of FIQA of GR is “big, yellow and white, solid and heavy, with the parrot mouth shape, bright cross-section, and strong smell” [11]. Based on the above, this experiment is based on the neuroprotective effect, through the artificial sensory intelligence technology, to quantify the “Shape”, “Color”, “Odour”, and “Taste” of GR. Secondly, HPLC and LC-MS were used to establish the fingerprint of GR and to identify and quantify the common components. On the premise of considering the overall characteristics of the multi-component synergistic effect of GR, the Pharmacodynamic Components Group (PCG) of GR was screened through the Spectrum-Effect relationship, and the equivalence with the overall pharmacodynamic effect of GR was verified. Through the correlation analysis of “Feature-Quantity-Effect”, the mechanism of “FIQA” of GR was explained. This study provides new ideas for the interpretation of the scientific connotation of the theory of “FIQA” and the standardization of characters, as well as new thinking modes and new technical routes for the quality evaluation of TCM.

2. Material and strategies

2.1. Material

30 batches of GR were collected. There were 5 batches from Guizhou. There were 4 batches from Yunnan, Sichuan, Shaanxi, and Anhui respectively. There were 3 batches from Hubei. There were 2 batches from Henan. There was one batch from Dabieshan, Jilin, Gansu, and Hunan respectively. They were identified as the dried tuber of *Gastrodia*

elata Bl. by Professor Zhai Yanjun from the teaching and Research Office of Liaoning University of traditional Chinese medicine.

2.2. Quantitative analysis of “feature” of GR

2.2.1. Quantitative analysis of “shape”

(1) Macro “Shape”

Each batch of GR was randomly taken into 6 pieces, and the longest, widest and thickest parts were measured with Vernier Caliper and weighed on an electronic balance. The results of each batch of GR were recorded and averaged.

(2) Microcosmic “shape”

The GR power passing No. 5 sieve was randomly selected and accurately weighed 200.0 mg in 6 parts. Chloral hydrate was added to the GR powder of each group, which was ground and transferred to a 10 ml volumetric flask many times. 7 ml of glycerol was added to each group of samples, and the volume was fixed to the scale with chloral hydrate. After fully shaking the sample solution before each sampling, 0.08 ml of the solution was accurately absorbed, 50 pieces were made in parallel, and the sclerenchyma cells were counted under the microscope (Olympus Co., Ltd., JAPAN).

Calculate the Microscopic Characteristic Index (MCI) according to the following formula.

$$MCI = (X \times V) / (V' \times W) \quad [12]$$

MCI (Quantity/mg)

X Number of microscopic characteristics of medicinal materials under each cover glass

V Total volume of quantitative drug suspension (mL)

V' Volume of drug suspension under cover glass (mL)

W Weight of medicinal materials (mg) (mg, Calculated by dry product)

2.2.2. Quantitative analysis of “color”

The GR powder passing No. 5 sieve was spread in a measuring dish for measurement, and the measured chromaticity values L^* , a^* , and b^* were recorded respectively by the SC-10 colorimeter (Shenzhen sanenchi Technology Co., Ltd, Shenzhen, CHINA). Where the L^* represents the brightness (light and dark) value, and the larger the L^* value, the brighter the sample is. The a^* represents the red green degree value. The larger the a^* value, the redder the color. The smaller the a^* value, the greener the color. The b^* represents the yellow blue degree value. The larger the b^* value, the yellower the color is. The smaller the b^* value, the bluer the color is. The Δa^* , Δb^* , ΔE^* are the difference between the chromaticity value of the standard sample and the measured chromaticity value of the test sample. The E^* is the total color difference between the sample to be tested and the standard sample.

$$\Delta E^* = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$$

The difference between the chroma value of the standard sample and the chroma value of the tested sample was the chroma value of the sample. The chroma value of appearance, section, and powder of GR was measured respectively.

2.2.3. Quantitative analysis of “Odour”

Accurately weighed 1.5 g GR powder passing No. 5 sieve was placed in a 20 ml electronic nose special headspace bottle. After automatic injection, each sample was tested 3 times, and each sample was tested 3 times in parallel by the Electronic Nose (Alpha MOS, FRANCE). Alpha soft V14.2 was used to process data. N-alkane standard solution (nC6–nC16) was used for calibration, the retention time was converted

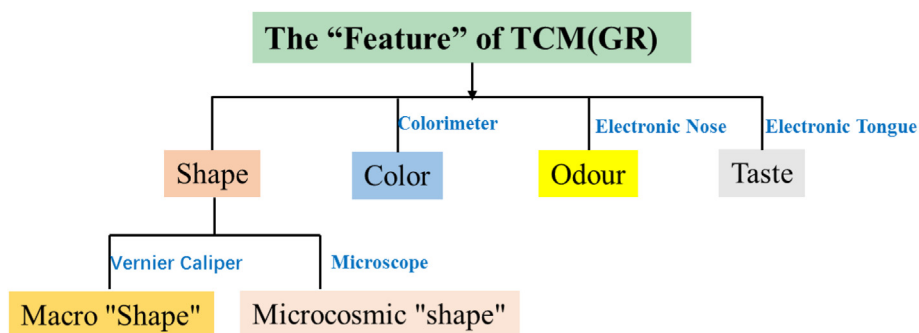


Figure 1. Quantitative analysis of "Feature" of GR.

to retention index, and then the compounds were qualitatively analyzed by the AroChemBase database. In this experiment, the dimensionality reduction method was used. According to the principal components obtained by SPSS software, the peak areas of 30 gas components from GR were expressed by D1, D2.....D30 respectively. As a result, 19 chromatographic peaks were reduced to 6 principal components, which were recorded as F1, F2... F6 in turn. The contribution rate was 85.194% and the maximum eigenvalue was 7.014. The principal component scoring function was obtained through the principal component load.

2.2.4. Quantitative analysis of "taste"

100 ml of 80 °C distilled water was added to 4 g of GR powder passing No. 5 sieve and dissolved. After cooling to room temperature, the upper solution was centrifuged at 3000 rpm for 5 min and then was directly placed in a special beaker (25 ml) for electronic tongue measurement. Each sample was tested three times, and the response values of seven sensors were obtained on the Asree Electronic Tongue (Alpha MOS, FRANCE) according to the test procedure.

2.3. Screening of PCG of GR

2.3.1. Discovery of active components of TCM (GR)

HPLC (Agilent Technology Co., Ltd., Beijing, CHINA) Gradient elution method was used to determine the common peaks of fingerprints from different sources of GR and the content of related effective components [13]. The following mass spectrometry conditions (Thermo Finnigan, USA) were used. The structure and content of the common components of GR were demarcated according to the results.

100 g of GR powder passing No. 5 sieve was weighed precisely, 50% methanol was added precisely, and it was allowed to stand for 12 h and then refluxed for 1 h. 30 % methanol was added to dilute to 100

ml to make GR extract. The cells were divided into blank control group, normal control group, model group, treatment group, and positive control group. According to the method of literature [13], the model of Oxygen Glucose Deprivation and Reoxygenation (OGD/R) was copied, and the chemical components of GR were separated and prepared into high, medium, and low concentration solutions respectively. The protective effect of common components on the injury of the Human Myeloneuroblastoma cell line (SH-SY5Y, provided by Shanghai cell bank of Chinese Academy of Sciences) OGD/R was determined.

Using the common peak areas of GR, and the neuroprotective efficacy, spss19.0 software was used to screen the effective components of GR through the Spectrum-Effect relationship.

2.3.2. Equivalence verification

(1) Cell experiment verification in vitro

By using the method of preparing the liquid phase, the PCG selected by "Spectrum Effect" analysis and other components (The other components were collected after the PCG was removed by the method of preparing the liquid phase.) were collected and prepared into the PCG solution with the concentration of 0.4, 0.08, 0.008, 0.004, 0.002, and 0.001 g/ml respectively. The protective effect of PCG and other components on the injury of SH-SY5Y OGD/R was determined.

(2) Experimental verification in vivo 1

The embryos were incubated with water at 28 °C. After Ao staining in vivo, the juveniles of Zebrafish were exposed to 0.64 mmol/L *tricine* methanesulfonic acid and killed under anesthesia. The procedure of

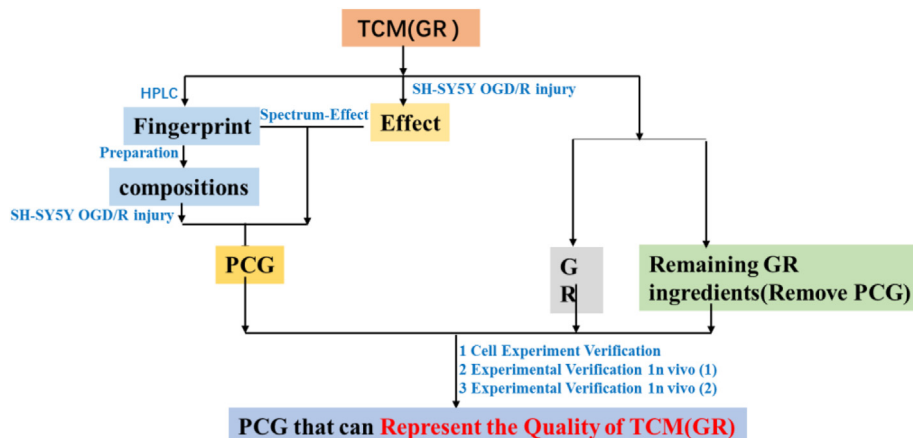


Figure 2. Screening of PCG of GR.

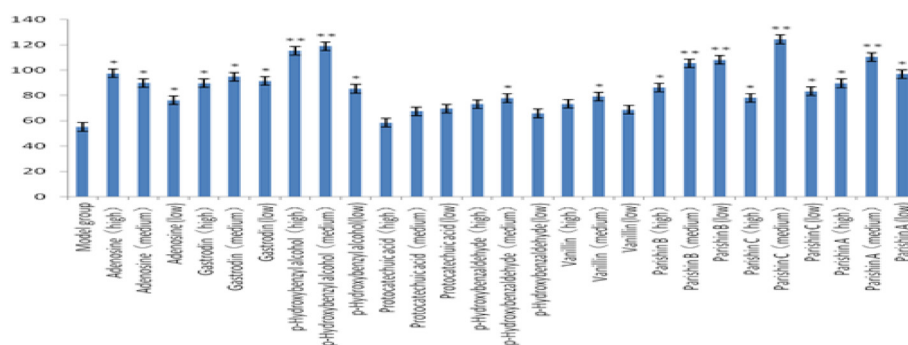


Figure 3. Protective effects of components in GR on OGD/R injury of SH-SY5Y. (*: $P < 0.05$ was a significant difference between treatment groups and the model group. **: $P < 0.01$ was a significant difference between treatment groups and the model group.)

anesthesia was to the requirements of the American Veterinary Association (AVMA). The model of Zebrafish embryo OGD/R injury was duplicated. *Gastrodia* Tuder Halimasch Tablets were ground and added with culture solution to prepare a solution of 0.005 g/ml. Ten 24 hpf Zebrafish were put into each hole of the 6-hole cell plate and randomly divided into normal control group, model group, and treatment group. According to the literature operation [14, 15], we used Image J image processing software to process the photos taken, count the fluorescence absorption intensity of the injured spinal nerve cells of Zebrafish, and calculated the fluorescence intensity and neuroprotection rate. The medicinal material was GR 5.

(3) Experimental verification in vivo 2

The model of persistent local cerebral ischemia induced by the middle cerebral artery in infarcted rats was duplicated [16, 17, 18, 19]. Investigations using experimental animals state in the Methods section that the research was conducted by the European Community guidelines (EEC Directive of 1986; 86/609/EEC). The ethics permit number for the use of animals in this study is SYXK (Liao) 2016–0009. 90 SD rats (half male and half female) were randomly divided into normal control group, model group, positive drug group, No. 5 GR extract group (high, medium, and low dose), and PCG group (high, medium, and low dose). After intraperitoneal injection of Pentobarbital Sodium (dose 50 mg/kg) for 15 min, the cervical vertebrae of mice were dislocated when the respiratory rate was reduced and the touch reaction was unresponsive. After the brain was taken out, thick sections were prepared, stained with hematoxylin and eosin, and photos were taken. The infarct area was calculated by Image J image processing software. Infarct volume was calculated according to the infarct area. Infarct volume = $t(A1 + A2 + \dots + An)$ (t is slice thickness, A is infarct area).

2.4. The mechanism of FIQA

The quantitative values of macroscopical “shape”, microcosmic “shape”, “color”, “gas”, “taste” and content of each component in the PCG of 30 batches of GR samples were input into SPSS statistical software respectively, and Pearson correlation analysis was used to reveal the mechanism of “FIQA” of GR.

2.5. Statistical treatment

The experimental results were analyzed by SPSS 19.0 software system, and the calculated data were expressed by mean \pm standard deviation. If the variance is uniform, the data were designed with a one-way ANOVA and LSD follow-up test. If the variance is not uniform, Welch was used for analysis, and Dunnett T3 was used for multiple comparisons. $P < 0.05$ was a significant difference.

3. Results

3.1. Results of quantitative analysis of “feature”

3.1.1. Determination results of “shape”

The results of macroscopic shape show that there were some differences in the length, width, and thickness of GR from different producing areas. The length was 86.1–116.0 mm, the width was 19.88–34.62 mm, and the thickness was 8.18–22.72 mm.

The results of microcosmic shape showed that there were significant differences in the microcosmic characteristic indexes of GR from different sources, among which the highest was No. 7, reaching 74.66, and the lowest was No. 14, 13.23. The results might be related to the quality of GR.

3.1.2. Determination results of “color”

The results showed that the L^* of GR section was in the range of 36.21–56.60, a^* was in the range of 4.18–8.34, and b^* was in the range of 13.76–23.49. L^* of GR powder was in the range of 51.30–64.03, a^* was in the range of 1.32–3.37, and the b^* was in the range of 6.53–10.84. The results of the color analysis showed that the b^* value of GR was greater than a^* value, and the a^* value color was close to the standard white, indicating that the color of GR was yellow and white, which was consistent with the description of “surface yellow-white to yellow-brown” in Chinese Pharmacopoeia (2015 Edition). It showed that it was feasible to determine the color of GR by using the Color Difference Instrument.

3.1.3. Determination results of “Odour”

The results showed that there were 38 odor fingerprint peaks, among which 19 characteristic odor compounds were the main components that caused the odor difference of multiple batches of GR. These 19 compounds were Acetaldehyde, 2-Methyl-2-propanol, Butanal, 2-butanol, 1-penten-3-one, Methyl butanoate, (Z)-3-hexenal, 2-hexanol, (Z)-2-Hexen-1-ol, 2-methylbutanoic acid, Sabinene, Beta-phellandrene, Gamma-Terpinene, Nonan-2-one, Hexylcyclopentane, Tridecane, Tetradecane, Beta-ionone, Propanoic acid, and dodecyl ester respectively.

3.1.4. Determination results of “Taste”

The results showed that the response values of 7 sensors identified by the electronic tongue were from 5.19 to 6.35. It could be seen that the “Taste” of GR can be quantified by electronic tongue technology, the method was stable and feasible, and “Odour” could be used as the basis of “FIQA”.

3.2. Screening of PCG of GR

3.2.1. Discovery of active components of TCM (GR)

In the fingerprint experiment, 12 common peaks were identified, and 30 batches of common peak areas of GR from different sources were

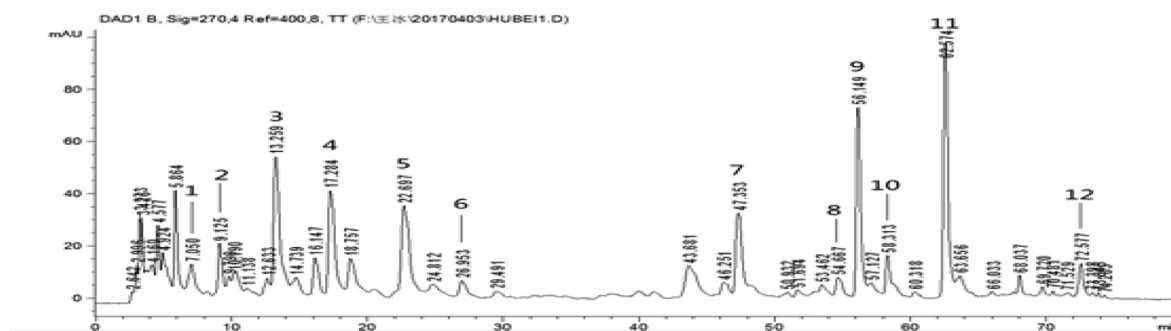


Figure 4. Schematic diagram of active components of GR (No. 1–12 were the identified 12 fingerprint common peaks).

obtained. According to the results of LC-MS and related literature [20, 21], 12 components were determined as Citric acid, Methyl Citrate, Adenosine, Gastrodin, p-Hydroxybenzyl alcohol, Protocatechuic acid, p-Hydroxybenzaldehyde, Vanillin, Parishin B, Parishin C, Parishin A and 4,4'-Dihydroxydibenzyl ether. The contents of the above 12 components were determined [13].

The results (Figure 3) of the protective effects of the common components on SH-SY5Y cells showed that 9 of the 12 common components had protective effects, which were Adenosine, Gastrodin, p-Hydroxybenzyl alcohol, Protocatechuic acid, p-Hydroxybenzaldehyde, Vanillin, Parishin B, Parishin C, and Parishin A, respectively (see Figures 1 and 2).

30 batches of GR fingerprint peak area and the results of SH-SY5Y protection [13] were analyzed by Spectrum-Effect relationship. The

results showed that Peaks 3, 4, 5, 7, 9, and 11 were the main components affecting the OGD/R injury of SH-SY5Y cells respectively (Figure 4). It was preliminarily determined that there were 6 effective components related to the neuroprotection of GR. The six components were Adenosine, Gastrodin, p-Hydroxybenzyl alcohol, Parishin A, Parishin B, Parishin C. The contents of six components were determined (Table 1).

3.2.2. Equivalence verification

(1) Cell experiment verification in vitro

The results showed that the selected PCG had a good OGD/R injury protection effect, which could represent more than 95% of the overall

Table 1. Content results of PCG (n = 3, mg·g⁻¹).

No.	Adenosine	Gastrodin	p-Hydroxybenzyl alcohol	Parishin B	Parishin C	Parishin A
1	0.0180	0.867	0.0893	0.904	0.0649	9.17
2	0.0138	0.980	0.0864	0.737	0.0519	8.20
3	0.0205	1.38	0.0408	0.843	0.0495	8.71
4	0.0191	1.29	0.111	1.12	0.0736	13.2
5	0.0131	1.26	0.0541	0.744	0.0520	8.07
6	0.00891	1.08	0.0751	0.934	0.0813	12.6
7	0.0144	1.28	0.0669	0.774	0.0701	7.22
8	0.0127	0.962	0.0847	0.724	0.0527	7.81
9	0.0116	1.57	0.0822	1.15	0.0813	15.7
10	0.0113	1.11	0.0799	0.739	0.0488	8.34
11	0.00973	0.694	0.0654	0.628	0.0476	7.81
12	0.0129	0.966	0.0416	0.658	0.0468	6.95
13	0.0112	0.759	0.0604	0.619	0.0513	8.32
14	0.00927	0.432	0.0828	0.492	0.0474	4.94
15	0.0200	1.25	0.183	1.44	0.113	15.1
16	0.0103	1.41	0.0554	0.834	0.0734	9.08
17	0.0105	1.18	0.0559	0.765	0.0688	9.30
18	0.0152	1.20	0.0486	0.754	0.0648	9.62
19	0.0138	1.65	0.0974	1.27	0.113	15.3
20	0.0150	1.84	0.103	1.14	0.0991	16.7
21	0.0226	1.88	0.0455	1.06	0.0923	11.7
22	0.0107	1.09	0.0870	0.805	0.0784	12.1
23	0.0151	1.13	0.102	0.969	0.0668	11.9
24	0.0186	1.28	0.0931	1.26	0.0865	18.8
25	0.0182	0.922	0.0896	0.888	0.0622	8.55
26	0.00981	1.99	0.0402	1.34	0.0682	11.3
27	0.00716	1.97	0.0991	1.35	0.0299	19.5
28	0.0142	0.697	0.0742	0.794	0.623	10.3
29	0.0133	1.06	0.0825	0.825	0.0719	10.5
30	0.0202	1.20	0.0458	0.852	0.0753	9.94

Remarks: 1–30 were GR from different sources.

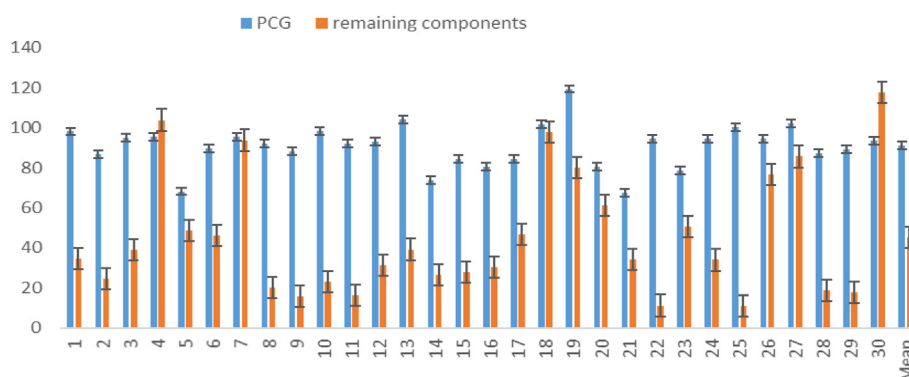


Figure 5. Results of neuroprotective efficacy (%), Comparative value of composition and overall efficacy of GR of PCG and remaining components of 30 batches of GR in vitro.

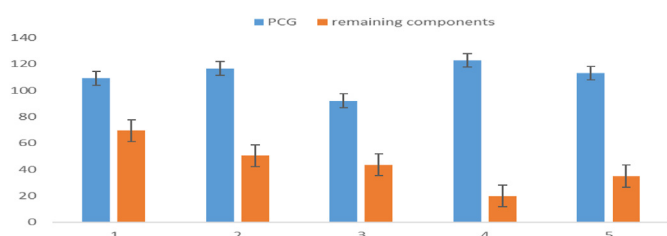


Figure 6. Neuroprotective efficacy (%), Comparative value of composition and overall efficacy of GR) results of PCG and remaining components of GR 5 (1–5 are the concentration of the extract 0.04, 0.08, 0.008, 0.004, 0.002 g/mL, respectively.).

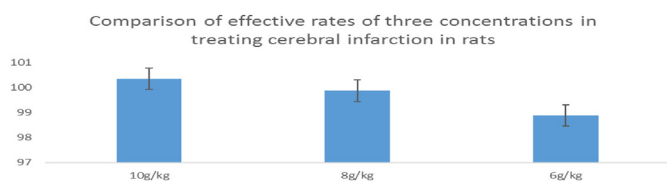


Figure 7. Comparison of the neuroprotective effect between the PCG and GR on cerebral infarction in rats of GR 5 (%).

Table 2. Correlation between the “Shape” of GR and PCG.

Component	Macro “Shape”		Microcosmic “Shape”	
	Pearson correlation	Significance (P)	Pearson correlation	Significance (P)
Adenosine	-0.096	0.046	0.031	0.036
Gastrodin	-0.344	0.026	0.842**	0.000
p-Hydroxybenzyl alcohol	-0.093	0.038	-0.053	0.045
Paliscin B	-0.404	0.027	0.620*	0.047
Paliscin C	0.419*	0.021	-0.161	0.028
Paliscin A	-0.345	0.042	0.444*	0.020

Remarks: *and** indicate that $P < 0.05$ and $P < 0.01$ respectively. Gastrodin was highly correlated with the MCI of thick-walled cells of GR, while Paliscin B was moderately correlated. According to the MCI, the contents of Gastrodin and Paliscin B can be predicted.

efficacy of GR (Figure 5). However, the efficacy of the remaining components of the PCG was only 46.4% (Figure 6), indicating that the protective effect of the PCG on OGD/R injury of SH-SY5Y cells could represent GR (see Figure 7).

(2) Experimental verification in vivo 1

The results (Figure 5) showed that the effective rate of neuroprotection of the PCG was more than 92%, while the effective rate of the remaining components was less than 40%, which proved that the PCG could represent GR in the protection of Zebrafish spinal injury.

(3) Experimental verification in vivo 2

The results showed that the PCG and GR extract had a significant difference from the model group ($P < 0.05$). The effective rate of each concentration group of PCG was more than 97%, indicating that the protective effect of PCG on cerebral infarction in rats could represent GR. The contents of six components (PCG) were determined (Table 1).

3.3. The result of the mechanism of FIQA

3.3.1. The results of correlation between “Shape” and PCG

The results of Table 2 showed that the six PCG were related to the macroscopic “shape”. Gastrodin, Paliscin B, Paliscin C and Paliscin A were moderately related to the “Shape” of GR. Adenosine and p-hydroxy benzyl alcohol had a low correlation with “Shape”.

3.3.2. The results of correlation between “Color” and PCG

Table 3 showed that the brightness, yellow-blue value, and total color difference of GR appearance were moderately correlated with the content of Paliscin C. The brightness, red-green degree, and yellow-blue degree of GR cross-section were all moderately correlated with the content of Paliscin C. The brightness, red-green value, and the total color difference of GR powder were correlated with the content of Gastrodin.

3.3.3. The results of correlation between “Odour” and PCG

The results are shown in Table 4. It could be seen that the main components F2, F3, F4, F5, and F6 correlate well with Paliscin C. The main components F2, and F3 were high positive correlations with Paliscin C, F6 was a high negative correlation with Paliscin C, and F4, F5 were medium correlations with Paliscin C.

3.3.4. The results of correlation between “Taste” and PCG

The results are shown in Table 5. It could be seen that Gastrodin, Paliscin B, and Paliscin A had good correlation with all “taste” indexes.

4. Discussion

4.1. Quantitative analysis of “feature” of GR

The four key elements of TCM characters are “Shape”, “Color”, “Odour”, and “Taste”, especially the latter three are the direct

Table 3. Correlation between the “Color” of GR and PCG.

form	Response value	1	2	3	4	5	6
surface	ΔL^*	0.192	-0.069	0.022	-0.135	-0.386*	-0.017
	Δa^*	0.174	0.047	-0.265	-0.175	-0.256	-0.211
	Δb^*	0.071	0.027	-0.184	-0.23	-0.329*	-0.221
	ΔE^*	-0.153	0.079	-0.083	0.08	0.323*	-0.044
section	ΔL^*	0.032	0.075	0.051	-0.047	-0.430*	-0.141
	Δa^*	0.162	0.192	0.056	0.029	-0.331*	-0.183
	Δb^*	0.151	-0.079	0.095	-0.174	-0.358*	-0.213
	ΔE^*	0.052	-0.133	-0.042	0.005	0.474*	0.101
powder	ΔL^*	-0.113	0.322*	-0.159	-0.049	-0.189	-0.09
	Δa^*	-0.076	0.467**	-0.055	0.157	-0.137	0.194
	Δb^*	-0.134	-0.312*	0.12	-0.017	-0.015	-0.012
	ΔE^*	-0.129	0.491**	-0.015	0.211	-0.093	0.268

Remarks: 1–6 were Adenosine, Gastrodin, p-Hydroxybenzyl alcohol, Paliscin B, Paliscin C, Paliscin A respectively, *and** indicate that $P < 0.05$ and $P < 0.01$ respectively.

Table 4. Correlation between the “Odour” of GR and PCG.

components	F1	F2	F3	F4	F5	F6
Adenosine	0.354	0.028	0.093	-0.131	0.197	-0.079
Gastrodin	-0.111	-0.269	-0.28	0.247	-0.144	0.100
p-Hydroxybenzyl alcohol	0	-0.017	0.02	0.070	-0.156	-0.077
Paliscin B	-0.022	-0.170	-0.106	0.116	-0.076	-0.084
Paliscin C	-0.136	0.747**	0.87**	-0.411*	0.551*	-0.736**
Paliscin A	-0.079	-0.107	-0.078	0.069	-0.119	-0.160

Remarks: F1–F6 were the six principal components of GR flavor by principal component analysis respectively, *and** indicate that $P < 0.05$ and $P < 0.01$ respectively.

Table 5. Correlation between the “Taste” of GR and PCG.

No.	Adenosine	Gastrodin	p-Hydroxybenzyl alcohol	Paliscin B	Paliscin C	Paliscin A
1	-0.106	-0.370*	-0.030	-0.412*	-0.257	-0.424*
2	0.183	0.415*	0.086	0.491**	0.283	0.518**
3	0.102	0.338*	0.035	0.416*	0.285	0.437*
4	-0.204	-0.360*	-0.063	-0.438*	-0.281	-0.442*
5	0.194	0.358*	0.069	0.440*	0.272	0.451*
6	0.209	0.363*	0.094	0.466**	0.272	0.464**
7	-0.081	0.303*	0.057	0.347*	0.239	0.404*

Remarks: 1–7 were AHS_sourness, PKS, CTS_saltiness, -NMS_umami, CPS, ANS, SCS respectively, *and** indicate that $P < 0.05$ and $P < 0.01$ respectively.

expression of chemical components in the characters. In addition, the standard of “FIQA” of GR is closely related to the above four elements. Therefore, the research object of this study is “Shape”, “Color”, “Odour”, and “Taste”.

Among the microscopic characteristics of GR, including sclerenchyma, needle crystal, gelatinized polysaccharide, etc., sclerenchyma is the special characteristic of GR, which is the main difference between GR and counterfeit. In addition, the microscopic characteristics of sclerenchyma are obvious, and the measured microscopic characteristic index is stable, which is not found in common counterfeit products. Therefore, sclerenchyma is selected as the microscopic “Shape” of GR.

4.2. Determination of PCG

On the premise of considering the overall characteristics of the interaction of TCM, the PCG is selected through the PCG of TCM, the remaining part after knocking out the PCG, and the pharmacodynamic comparison of all components of TCM. They are equivalent to the pharmacodynamic effects of the overall components of TCM and can

represent more than 90 % of the pharmacodynamic effects. The above experiments verified that the PCG was reasonable.

4.3. Analysis of the mechanism of GR's “FIQA”

It could be seen that macroscopical “Shape” and microcosmic “Shape” can reflect the quality of medicinal materials. Most of the indexes of GR appearance “Color” were moderately correlated with the content of Paliscin C. The “Color” index of GR cross-section was negatively correlated with the content of Paliscin C. The four indexes of powder color of GR have a low correlation with Gastrodin. The “Odour” index of GR had a strong correlation with the group of effective components, mainly reflected in the good correlation between the main components 2, 3, 4, 5, 6 and the content of Paliscin C. The “Taste” of GR had a good correlation with the PCG, which was embodied in the fact that the acid, salty, sweet, and bitter tastes of GR were mostly related to Gastrodin, Paliscin B, and Paliscin A. Among them, acid, salty and sweet had a medium correlation with the three components, while bitter had a low correlation with the three components.

5. Conclusion

There is a significant correlation between the “Shape”, “Color”, “Odour” and “Taste” of GR and the PCG, which can fully demonstrate the mechanism of “FIQA”. By analyzing the relationship between the characteristics of TCM and the content of pharmacodynamic components that can represent the quality of TCM, it provides a new idea for the study of the theory of FIQA of TCM.

Declarations

Author contribution statement

Kang Ting-guo: Conceived and designed the experiments.

Wang Bing: Performed the experiments.

Zhang Hui: Analyzed and interpreted the data.

Pei Wen-han: Performed the experiments; Contributed reagents, materials, analysis tools or data.

Sun Yan-tao: Performed the experiments; Wrote the paper.

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Data availability statement

Data included in article/supp. material/referenced in article.

Declaration of interest's statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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