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The establishment of comprehensive quality evaluation model for flavor characteristics of green Sichuan pepper (*Zanthoxylum armatum* DC.) in Southwest China

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A R T I C L E I N F O

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A B S T R A C T

In this study, the quality indexes and sensory evaluation of *Zanthoxylum armatum* DC. from the main production areas in Southwest China were analyzed. Further, correlation analysis (CRA), principal component analysis (PCA) and cluster analysis (CA) were used to comprehensively evaluate the quality characteristics of *Z. armatum*. The results showed that the sensory indexes and physicochemical indexes of *Z. armatum* were significantly correlated. Five principal component factors were extracted from 12 indexes by PCA, and a comprehensive evaluation model of quality was established with $Y = 0.2943Y_1 + 0.2387Y_2 + 0.1896Y_3 + 0.1679Y_4 + 0.1094Y_5$. On this basis, 21 producing areas were grouped into 4 groups and 3 groups by Q-type CA, respectively. R-type CA showed that the content of hydroxyl-sanshools, linalool content and b* value were the quality characteristic indexes of *Z. armatum* in Southwest China. This work provided an important theory and practice reference for *Z. armatum* quality evaluation and in-depth product development.

Introduction

Sichuan pepper (*Zanthoxylum*) belongs to the rutaceae family, there are more than 200 varieties in the world, and it has a long history of crop cultivation in China (Li et al., 2020; Guo et al., 2011). Southwest China is the main production area of Sichuan pepper. The main cultivated varieties are *Zanthoxylum bungeanum* Maxim. and *Zanthoxylum armatum* DC. (Li et al., 2016). Among them, *Z. armatum* is also called "Tengjiao" because its branches and leaves are spread out and pendulous, meanwhile its branches resemble vines. In addition, it is also known as "Qinghuajiao" regarding to its green fruit (Xu et al., 2020; Cheng et al., 2021). Green Sichuan pepper contains abundant bioactive compounds, including alkaloids (Gong et al., 2021), volatile oil (Rout et al., 2007), amides (Bryant and Mezine, 1999), and phenols (Sun et al., 2020a), etc. Meanwhile, it also has important medical value (Kaleeswaran et al., 2018). Furthermore, *Z. armatum* has the characteristics of long pungency taste and fresh smell, therefore, amides and volatile oil are the two main indexes to evaluate the quality of *Z. armatum*. The former is the main contributor to pungent taste, and the latter is the main substance for aroma (Song et al., 2022). Hydroxyl-α-sanshool, hydroxyl-β-sanshool, hydroxyl-γ-sanshool are the main amides in *Z. armatum* (Luo et al., 2022a; Feng et al., 2023a). Linalool and limonene accounted for the largest area and the highest contribution value of aroma in the volatile components of *Z. armatum* (Rout et al., 2007; Liu et al., 2020).

In China, *Z. armatum* is mainly distributed in Sichuan, Yunnan, Guizhou, Chongqing and other Southwest regions. The harvest time is concentrated in June to August every year. At present, *Z. armatum* is mainly used to make *Z. armatum* oil, *Z. armatum* flavor food, *Z. armatum* flavor dishes, etc. However, due to different origins and influenced by genetic and environmental factors, the content of bioactive compounds

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in *Z. armatum* varies, which leads to differences in product quality (Kaigongi and Lukhoba, 2021). According to the report, the distinctive pungent taste of *Z. armatum* is mainly caused by a series of unsaturated amides (sanshools) (Luo et al., 2022a). Zhuo et al. (2021) found that there were significant differences in the content of hydroxyl-α-sanshool (49.47–100.87 mg/g), hydroxyl-β-sanshool (4.26–10.70 mg/g) and hydroxyl-γ-sanshool (2.30–4.30 mg/g) in *Z. armatum* from 24 different regions. At the same time, in addition to the influence of genetic and environmental factors, volatile and instability of volatile oil will also lead to the differences in volatile compounds in *Z. armatum* (Rashed et al., 2021). Feng et al. (2021b) detected the volatile compounds in eight different geographical indications of *Z. bungeanum* and *Z. armatum*, the results showed that the volatile compounds in the samples differed in composition, with the linalool (1333.17–1474.24 mg/100 g) and limonene (408.57–1031.04 mg/100 g) contents of *Z. armatum* varying widely. With the exception of in the main flavors such as pungency and aroma, *Z. armatum* will also be due to the natural geographical environment, climate conditions and other effects of different degrees of color. The green color of *Z. armatum* is related to chlorophyll (Cheng et al., 2021). Gebregziabher et al. (2022) demonstrated that the difference in production area has a significant effect on chlorophyll accumulation.

At present, the quality evaluation of *Z. armatum* is mainly based on amides and volatile oil, etc, which is difficult to accurately qualitative and quantitative evaluation of complex components. Moreover, color evaluation is mainly based on sensory index, which is subjective and has obvious error. Recently, there is still a lack of research on quality characteristics identification and quality standardization evaluation of *Z. armatum*. Therefore, it is urgent to establish a set of *Z. armatum* standardized quality evaluation system based on quality characteristics index. In order to better develop and utilize *Z. bungeanum*'s leaves, Chen et al. (2019) confirmed that quercitrin and afzelin were key compounds for the quality control of *Z. bungeanum*'s leaves through similarity analysis (SA), principal component analysis (PCA), hierarchical cluster analysis (HCA) and discriminant analysis (DA), meanwhile established a standardized evaluation model, the leaves of 13 cultivars were divided into four groups according to the characteristic indexes. Sun et al. (2021) measured 23 quality indicators of 90 Chinese foxtail millet, divided all materials into five groups through cluster analysis (CA), extracted 7 principal components from the 23 indicators by PCA, and established a comprehensive quality evaluation system. On this basis, 8 kinds of high-quality millet were identified. So, rational use of analytical methods can not only simplify a large number of complex data, but also establish a standardized evaluation model, which is crucial to product quality control.

In this work, the color, physical and chemical indexes, pungency and aroma of *Z. armatum* from Southwest China were analyzed. The quality characteristics of *Z. armatum* were evaluated by correlation analysis (CRA), PCA and CA, building the appraisal standardized model, in order to provide reference for *Z. armatum* quality evaluation and in-depth product development.

Materials and methods

Plant material

Z. armatum samples were collected from 21 *Z. armatum* producing areas in Sichuan, Yunnan, Guizhou and Chongqing from June to July 2021. Specific sample information is shown in Table S1.

Chemicals

Methanol, acetonitrile, *n*-hexane and anhydrous sodium sulfate were purchased from Chengdu Cologne Chemical Co., LTD.(Chengdu, China). The standard hydroxyl-α-sanshool, hydroxyl-β-sanshool and hydroxyl-γ-sanshool were purchased from Chengdu Pusi Biotechnology Co., LTD. (Chengdu, China, purity ≥ 98.00%). The standard linalool (The content is 98.8%) and limonene (The content is 96%) were purchased from National Center for Standard Substances.

Sensory measurement

Measurement of color difference

The samples were laid in a petri dish, and the hand-held color difference instrument (CR-10 Plus, KONICA MINOLTA, Japan) was used for determination in the room temperature and sunlight. Five different detection points were selected for each sample, with L*, a*, b*, △E values recorded and the average values were calculated.

Measurement of rate of depressed oil droplet and degree of uniformity

The rate of depressed oil droplet and degree of uniformity were detected by referring to the Forestry industry standards of China (SFA, 2005). The specific operations are as follows: First, 100 *Z. armatum* were obtained by quartering method, and the procedure was repeated three times to take a total of three samples. The obtained samples were placed on white A4 paper, and the number of grains in oil gland depression were visually inspected under indoor sunlight, which was recorded as "a". Then observe whether there were significant differences in color and size, which was recorded as "b". Rate of depressed oil droplet and degree of uniformity were calculated as per Eq. (1) (2):

$$Rate\ of\ depressed\ oil\ droplet(\%) = \frac{a}{100} \times 100\%$$

$$Degree\ of\ uniformity(\%) = \frac{100 - b}{100} \times 100\%$$

Measurement of pungency

Extraction of hydroxyl-sanshools

According to Cheng et al. (2021) hydroxyl-sanshools were extracted. The sample was crushed with a grinder (FD-15-F-500A, Zhejiang Yongkang Struggle Industry and Trade Co., LTD.), 5.00 g of crushed sample mixed with 50.00 mL methanol was placed in an ultrasonic cleaner (YM-040S, Shenzhen Fang ao Micro Electronics Co., LTD.) for 30 min. After filtering, the filtrate was transferred to a 50.0 mL brown volumetric flask and diluted 10 times with methanol. Then filtered through a 0.22 μm membrane, and stored at −20 °C for later use.

Analysis of hydroxyl-sanshools content by HPLC

The high performance liquid chromatography (HPLC) was used for the analysis of hydroxyl-sanshools, according to Cheng et al. (2021). The 10 μL samples were injected into 1260 HPLC (Agilent, USA) at a rate of 1 mL/min and separated on Eclipse Plus C 18 (4.6 mm × 150 mm, 5 μm) column. Using water (A) -acetonitrile (B) as mobile phase, the gradient program was set as follows: 0–5 min, 65 %A-35 %B; 5–10 min, 60 %A-40 %B; 10–30 min, 50 %A-50 %B; 30–35 min, 65 %A-35 %B. VWD was used as the detector, the wavelength was set at 268 nm, and the column temperature was maintained at 35 °C.

The standards of hydroxyl-α-sanshool, hydroxyl-β-sanshool, hydroxyl-γ-sanshool were prepared into 0.05–0.6, 0.001–0.01, 0.0001–0.003 mg/mL standard solution with methanol, and then injected into HPLC through 0.22 μm filter membrane.

The concentration of hydroxyl- sanshools were calculated from the standard curve, and the content of hydroxyl-sanshools were calculated as per Eq. (3):

$$Hydroxy - sanshools\ content(mg/g) = \frac{\mu_1 \times V \times X_1}{m_1}$$

where μ_1 is the concentration of hydroxyl-sanshools (mg/mL) ; V is constant volume (mL); X_1 is the dilution ratio; m_1 is the sample mass (g).

Measurement of aroma

Extraction of volatile oils

The volatile oils of *Z. armatum* were extracted by steam distillation according to the Standardization Administration of China (SAC, 2009). 100.00 g of untreated *Z. armatum* were added into 700 mL ultrapure water and extracted for 4 h to obtain volatile oils.

Analysis of linalool and limonene contents by GC–MS

Volatile oils were treated with reference to Cheng et al. (2021). The volatile oil was dissolved with *n*-hexane and diluted 50 times. Anhydrous sodium sulfate was added and stored overnight at −20 °C. The sample solution was filtered by 0.22 μm filter membrane before injection.

The contents of linalool and limonene were determined by Gas Chromatography-Mass Spectrometry (GC–MS), according to Cheng et al. (2021). The 1 μL sample was injected into 7890 A-5975C GC–MS (Agilent, USA) at a ratio of 10:1 and detected on HP-5 MS (30 m × 0.25 mm × 0.25 μm) elastic quartz capillary column. The inlet temperature and detector temperature were set at 250 °C and 260 °C, respectively. The heating procedure was set as follows: the initial column temperature was 50 °C for 2 min, then increased to 110 °C at 3 °C/min for 3 min, next increased to 140 °C at 3 °C/min for 10 min, and finally increased to 220 °C at 6 °C/min for 2 min. Helium was used as carrier gas. The solvent delay was 3 min. The mass spectrometry condition was to use EI as an ion source and set its temperature at 230 °C. The temperature of the four-stage rod was controlled at 150 °C. The samples were scanned in the *m/z* range of 40–550 AMu and retrieved in the NIST 11.1 standard spectrum library.

Linalool and limonene standards were prepared into standard solutions of 1.5–3.0 and 0.6–1.6 mg/mL by *n*-hexane, and then detected by GC–MS after filtered by 0.22 μm.

The concentrations of limonene and linalool in the sample were calculated from the standard curve, the contents were calculated as per Eq. (4):

$$Aromas\quad content = \frac{\mu_2 \times X_2}{m_2}$$

where μ_2 is the concentration of Linalool and limonene (mg/mL) ; X_2 is the dilution ratio; m_2 is the sample mass (g).

Statistical analyses

Statistical analysis was conducted by Origin 2021 (Origin Lab, USA) and SPSS 27 (SPSS Inc., Chi-cago, IL, US). Among them, the data is analyzed and plotted through Origin 2021. Then, SPSS 27 was used for significant difference analysis (SDA), correlation analysis (CRA), principal component analysis (PCA) and cluster analysis (CA). All results were expressed as mean ± standard deviation (SD) ($P < 0.05$).

Results and discussions

Differences of main quality indexes of Z. armatum from different origin

Color difference of Z. armatum in different origin

The peel of fresh or dried fruit of *Z. armatum* is green, so a* (red/green) value is an important index to evaluate the color of *Z. armatum.* As shown in Table 1, a* values of *Z. armatum* were negative, it indicated that the color of *Z. armatum* was mainly green. According to the research, the green color of *Z. armatum* was related to chlorophyll (Cheng et al., 2021). The L* value shows that *Z. armatum* was dark, and b* indicates that *Z. armatum* was yellow, and $\triangle E < 2$ indicated that the color variation were within the acceptable range. In Table 1, the L*, a* and b* values of *Z. armatum* in the inspection area were different. Among them, EB had the smallest a* and the largest b* value, which proves that its green color was the most pure and the yellow was more obvious. LZ had the highest L* value, indicated that the color was brighter. By the way, the L*, a* and b* values of *Z. armatum* from the same origin were not all the maximum or minimum, which also demonstrated that the color of *Z. armatum* from different origin were different. The color of *Z. armatum* was influenced by chlorophyll, but also by its secreted peroxidase. The products were catalyzed by peroxidase and then polymerized, resulting in a change in the color of *Z. armatum*. Meanwhile, peroxidase caused browning and darkening of *Z. armatum* after contact with the substrate. In addition, its activity also affected the degradation of chlorophyll, which affected the color of the *Z. armatum* (Yang et al., 2023).

Table 1
Color test results of *Z. armatum.*

Abbreviations	L*	a*	b*	△E
HY-1	27.8 ± 3.63^{abc}	−4.4 ± 0.96^{bcde}	18.3 ± 1.75^{ab}	0.9 ± 0.78^{a}
HY-2	24.1 ± 2.05 ^{cd}	−3.8 ± 0.88^{abcd}	17.1 ± 1.05^{abc}	0.7 ± 0.71^{a}
RS	26.1 ± 2.72^{bcd}	−4.0 ± 0.51^{abcde}	16.9 ± 1.79^{abc}	0.6 ± 0.34^{a}
DL	24.3 ± 3.57 ^{cd}	−4.0 ± 0.77^{abcde}	16.4 ± 1.53^{bc}	1.2 ± 0.96^{a}
BX	27.2 ± 4.17^{abcd}	−4.2 ± 1.06^{bcde}	18.3 ± 1.81^{ab}	1.8 ± 2.22^{a}
HY	25.2 ± 3.22^{bcd}	−4.3 ± 0.62^{bcde}	16.4 ± 2.33^{bc}	0.5 ± 0.47^{a}
XC	26.2 ± 2.19^{bcd}	−4.3 ± 0.60^{bcde}	18.0 ± 0.87^{abc}	0.7 ± 0.39^{a}
QL	27.1 ± 1.74^{abcd}	−4.4 ± 1.93^{bcde}	17.3 ± 3.49^{abc}	1.2 ± 0.89^{a}
DY	25.4 ± 2.34^{bcd}	−4.5 ± 0.55^{bcde}	17.2 ± 2.06^{abc}	1.1 ± 0.88^{a}
JJ	26.8 ± 1.68^{abcd}	−4.9 ± 0.61^{e}	18.0 ± 0.71^{abc}	0.7 ± 0.56^{a}
QW	26.3 ± 1.35^{bcd}	−4.9 ± 0.41^{de}	17.8 ± 0.69^{abc}	1.6 ± 1.67^{a}
EB	28.9 ± 1.77^{ab}	−6.1 ± 0.43^{f}	19.3 ± 1.82^{a}	0.5 ± 0.31^{a}
MB	26.5 ± 4.24^{bcd}	−3.5 ± 1.14^{ab}	15.6 ± 2.24^{c}	1.7 ± 1.60^{a}
NJ	27.6 ± 2.55^{abc}	−3.0 ± 0.46^{a}	19.0 ± 1.25^{a}	0.5 ± 0.60^{a}
LZ	30.7 ± 5.26^{a}	−4.7 ± 1.00^{cde}	19.2 ± 1.75^{a}	0.9 ± 0.87^{a}
GA	23.2 ± 2.36^{d}	−3.9 ± 0.62^{abcde}	16.5 ± 0.99^{bc}	0.7 ± 0.59^{a}
BZ	23.7 ± 4.16 ^{cd}	−3.6 ± 0.73^{abc}	17.2 ± 2.94^{abc}	1.0 ± 0.51^{a}
DZ	26.1 ± 3.65^{bcd}	−4.7 ± 0.77^{cde}	16.9 ± 1.43^{abc}	1.3 ± 1.06^{a}
CQ	26.2 ± 4.68^{bcd}	−4.4 ± 1.15^{bcde}	17.2 ± 3.02^{abc}	1.1 ± 1.54^{a}
ZT	28.7 ± 0.75^{ab}	−4.4 ± 0.35^{bcde}	18.1 ± 0.97^{abc}	0.7 ± 0.59^{a}
ZY	26.0 ± 2.78^{bcd}	−4.5 ± 0.47^{bcde}	18.2 ± 2.06^{ab}	1.1 ± 1.17^{a}

Rate of depressed oil droplet and degree of uniformity difference of Z. armatum in different origin

The oil droplets of *Z. armatum* were related to the volatile oil content and aroma components, which were highly susceptible to destruction during growth, harvesting and transportation, thus affecting the aroma of *Z. armatum* (Chen et al., 2019). Therefore, the rate of depressed oil droplet is important for the quality control of *Z. armatum.* Fig. 1(a) shows the oil droplet depression rate of *Z. armatum* from 21 producing areas. In Fig. 1(a), the oil droplet of all *Z. armatum* had different depressions and damages, with the rate of depressed oil droplet was 8.67–33.00%. Among them, GA was the least, while NJ and ZY were the highest, with a maximum difference of 3.8 times. This result was related to picking and transportation around. When picking and transportation

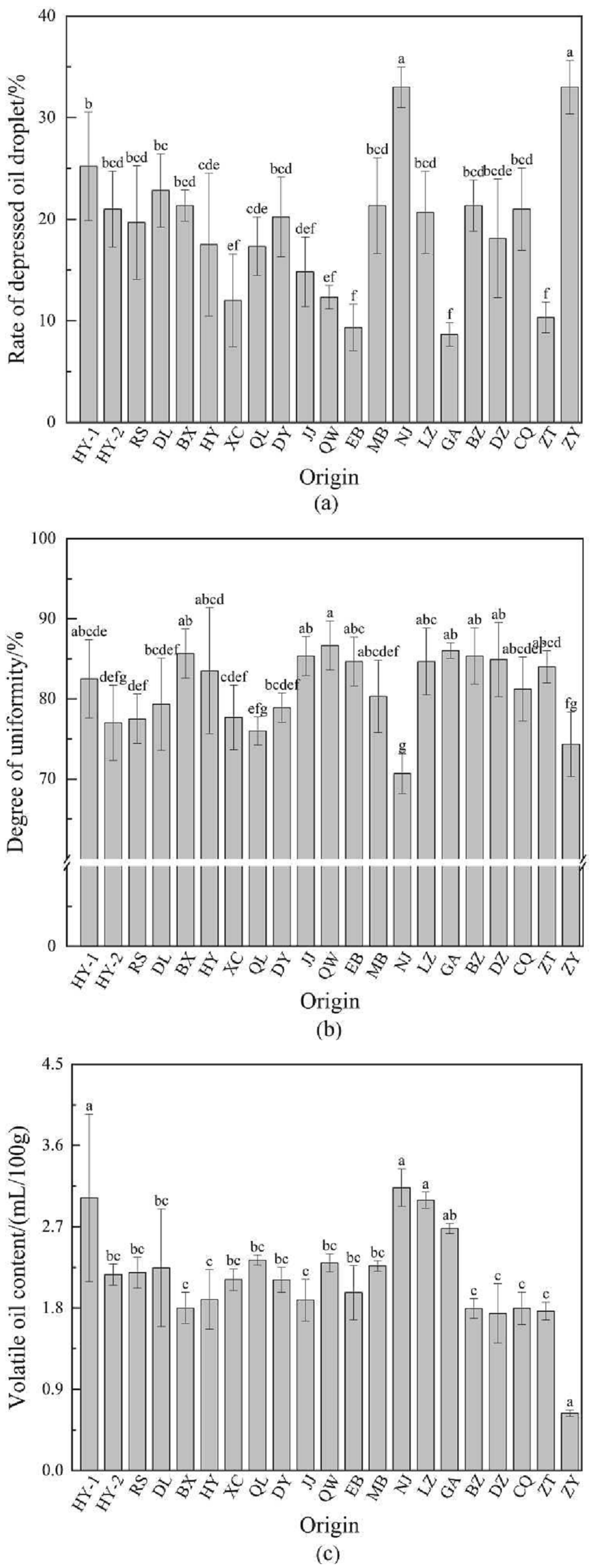

Fig. 1. Rate of depressed oil droplet (a), degree of uniformity (b) and Volatile oil content (c) in *Z. armatum.*

was improper, such as picking *Z. armatum* directly by hand, the fingers were easy to crush the oil droplet after force, as the oil droplet were rich in volatile oil, so its rupture would lead to the loss of volatile oil, thus forming "Paoyoujiao", after a certain time, *Z. armatum* color was dark brown, pungency was reduced and aroma became weaken (Chen et al., 2019).

The degree of uniformity was closely related to the climate, soil and cultivation management techniques of *Z. armatum* planting site, etc. After picking, there might be poor quality *Z. armatum* with inconsistent color and size mixed in, affecting the quality of *Z. armatum* (Zhang, 2011). Therefore, the degree of uniformity of *Z. armatum* needs to be detected to better control its quality. In Fig. 1(b), the degree of uniformity ranges from 70.67% to 86.67%, among which QW was the highest and NJ was the lowest. This indicated that there were significant differences in the uniformity of *Z. armatum* from different origins. Zhang. (2011) pointed out that the size and color of *Z. bungeanum* were mainly affected by natural factors such as temperature, illumination time and precipitation. On the same *Z. bungeanum* tree, the fruit size and color with good light condition were better than those with dark branches and leaves. Meanwhile, when the branches and leaves were too dense, the rain accumulated in rainy days couldn't be dried quickly, the fruit was easy to mold, which would affect its color. These fruits without full photosynthesis, their nutrient content would also be lost, thus affecting *Z. armatum* quality.

The content difference of hydroxyl-sanshools in Z. armatum from different origin

Sun et al. (2020b) had pointed out that hydroxyl-α-sanshool, hydroxyl-β-sanshool and hydroxyl-γ-sanshool were the most representative components responsible for pungent sensation. Therefore, the content of the above three hydroxyl-sanshools were mainly detected and analyzed in this study. Fig. S1(a) shows the HPLC chromatogram of *Z. armatum*. The three peaks labeled were identified as hydroxyl-α-sanshool, hydroxyl-β-sanshool, and hydroxyl-γ-sanshool based on the retention time of the samples and the standards. Fig. S1(b) shows the HPLC chromatogram of *Z. armatum* from 21 production areas, the retention time of sanshools was approximately the same in each production area, but the peak height and peak area varied greatly. Fig. 2 shows the content changes of hydroxyl -α -sanshool, hydroxyl -β -sanshool, hydroxyl -γ -sanshool and the total content of the three sanshools from 21 producing areas.

As shown in Fig. 2, the ranges of hydroxyl-α-sanshool, hydroxyl-β-sanshool hydroxyl-γ-sanshool and total sanshool content were 14.4897–33.7574 mg/g, 0.0763–0.5439 mg/g, 0–0.132 mg/g and 14.6321–34.3723 mg/g, respectively. These results suggested that hydroxyl-α-sanshool was the most highly amides in *Z. armatum*, and its variation in content determined the variation in the total value of sanshools, which was consistent with the research results of Machmudah et al. (2009). Fig. 2 showed that DY (33.7574 mg/g), GA (32.4728 mg/g) and ZT (32.2487 mg/g) had higher content of hydroxyl -α -sanshool, but ZY (14.4897 mg/g) and EB (17.7057 mg/g) were lower. Since the tingling sensation of *Z. armatum* was mainly caused by hydroxyl -α -sanshool (Zhuo et al., 2021), it indicated that *Z. armatum* from DY, GA, ZT had the most intense tingling sensation. However, the contents of hydroxyl -β -sanshool and hydroxyl-γ-sanshool were relatively low, and the content of hydroxyl-β-sanshool in ZT (0.5439 mg/g) was the highest, while EB (0.0763 mg/g) was the lowest. DY (0.132 mg/g) has the highest content of hydroxyl -γ -sanshool, but MB has no detectable hydroxyl -γ -sanshool content. In terms of the total content of hydroxyl-sanshools, the contents of DY (34.3723 mg/g), ZT (32.8972 mg/g) and GA (32.7925 mg/g) were higher. However, the contents of ZY (14.6321 mg/g) and EB (17.8361 mg/g) were lower. To sum up, there were great differences in hydroxyl-sanshools content of *Z. armatum* from different origins, which was similar to the research results of Chen et al. (2022).

The content of amides in *Z. armatum* was greatly affected by natural

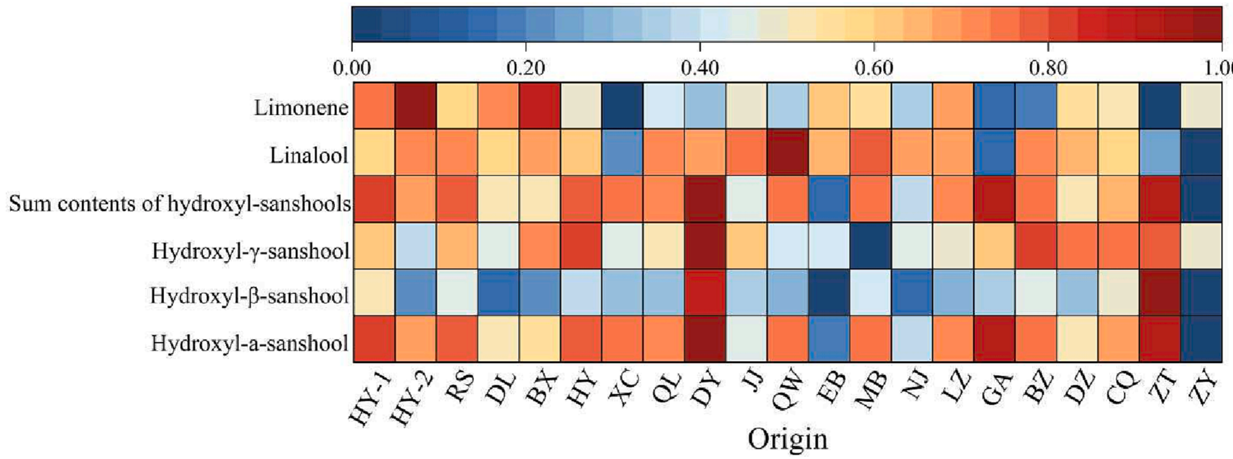

Fig. 2. Heat map of hydroxyl-sanshools, linalool and limonene contents in *Z. armatum.*

factors. Luo et al. (2022a) had indicated that the amides in *Z. armatum* were influenced by the origin. The content of amides in *Z. armatum* at high altitude and strong light was higher than at low altitude and weak light areas, sufficient light conditions were more conducive to the accumulation of active components in *Z. armatum*. Fresh *Z. armatum* had higher water content, which was prone to mold and reduced the content of sanshools. In addition, other studies had shown that amides in *Z. armatum* were extremely unstable, and their content would be reduced under high temperature, light and acidic conditions, therefore, their content would also be different during transportation and storage, thus affecting the quality of *Z. armatum* (Luo et al., 2022b).

Difference of volatile oil content in Z. armatum from different origin

The volatile oil extracted from *Z. armatum* by steam distillation was a light yellow translucent oily liquid with strong aroma, which was one of the main indicators to evaluate the quality of *Z. armatum* (Zheng et al., 2021). Fig. 1(c) shows the volatile oil content of *Z. armatum* from 21 producing areas. The content of volatile oil in *Z. armatum* was 0.64–3.13 mL/100 g, with an average value of 2.14 mL/100 g. In Fig. 1(c), NJ (3.13 mL / 100 g), HY-1 (3.02 mL / 100 g), LZ (2.99 mL / 100 g) of *Z. armatum* had higher volatile oil content, the ZY (0.64 mL / 100 g) content significantly lower. Therefore, the content of volatile oil of *Z. armatum* from different producing areas varies greatly. Ke et al. (2020) explained that volatile oil content was one of the important indexes reflecting *Z. armatum* aroma, whereas growth environment, cultivation management and genetic factors all affect its content. At the same time, when picking and transportation was improper, *Z. armatum* surface oil droplet would rupture and volatile oil content would also reduce, therefore affecting the quality of *Z. armatum* (Chen et al., 2019).

The content difference of linalool and limonene in Z. armatum from different origin

Feng et al. (2021b) calculated the relative content of aroma substances in *Z. armatum* and found that linalool and limonene were the main volatile substances. Therefore, the content of linalool and limonene was detected and analyzed in this study. Fig. S2(a) shows the GC–MS chromatogram of *Z. armatum*, and the two peaks were identified as limonene and linalool based on the retention times of the samples and the standards. Fig. S2(b) shows the total ion chromatogram of *Z. armatum* from 21 production areas, the peak height and peak area of limonene and linalool had some differences. Fig. 2 shows the contents of linalool and limonene in *Z. armatum* from 21 producing areas.

As shown in Fig. 2, linalool content was 0.1289–17.7212 mg/g, with an average content of 10.4927 mg/g, limonene content was 0.2711–12.3253 mg/g, with an average content of 6.0695 mg/g. The content of linalool in Leshan city was high, QW (17.7212 mg/g), MB (13.6470 mg/g), JJ (13.6070 mg/g) and EB (11.2773 mg/g), respectively. However, the linalool contents of ZY (0.1289 mg/g), GA (3.0263 mg/g), XC (3.8717 mg/g) and ZT (4.3207 mg/g) were all lower than the average, and the maximum difference was 137 times. Next, the limonene content of *Z. armatum* in Meishan city was relatively high, HY-2 (12.3252 mg/g), HY-1 (9.2180 mg/g), DL (8.7551 mg/g), RS (7.3033 mg/g), respectively. However, the limonene content in XC (0.2711 mg/g), ZT (0.4529 mg/g), GA (1.8958 mg/g) and BZ (2.4984 mg/g) was lower. In addition, according to studies, limonene had a citrus flavor, while linalool was floral and fragrant (Ji et al., 2019). Therefore, it could be inferred that *Z. armatum* in Leshan and Meishan presents different flavors. In summary, the contents of linalool and limonene from different sources were significantly different, which was similar to the results of Feng et al. (2021b).

Zheng et al. (2021a) showed that temperature, precipitation, sunshine duration could affect the accumulation of secondary metabolites in plants, and the content of volatile compounds in *Z. armatum* were correlated with climate factors to varying degrees. Even for the same variety, volatile compound content would vary significantly due to different growing environment. At the same time, the content of volatile oil was too low would affect the content of its compounds, so that the aroma of *Z. armatum* differences.

Determination and evaluation of characteristic quality indexes of Z. armatum

Correlation analysis of quality indicators

SPSS 27.0 was used for CRA of various indicators of *Z. armatum*, and the results are shown in Table 2. There was a significant positive correlation between L* and b* value, rate of depressed oil droplet and degree of uniformity, hydroxyl -α -sanshool content and hydroxyl -β -sanshool content ($P < 0.01$). Then, there was a significant positive correlation between a* value and rate of depressed oil droplet, volatile oil content and hydroxyl -β -sanshool content, hydroxyl -β -sanshool content and hydroxyl -γ -sanshool content ($P < 0.05$). Furthermore, a* value was significantly negatively correlated with degree of uniformity, and between hydroxyl-β -sanshool content and linalool content ($P < 0.05$). Positive correlation indicates that there are synergistic effects among these indicators, while negative correlation indicates that the indicators are mutually antagonistic (Yang et al., 2023).

The above results indicated there were different degrees of correlation between these quality indicators. Mainly due to the fact that both L* and b* values belonged to the color values of *Z. armatum*, and when the b* was larger (yellowish), its corresponding L* was also larger (brighter). Most of the time, there was a strong correlation between L* and b* except for a* value (Khan et al., 2009). In addition, the oil droplet

Table 2
Correlation analysis of quality indicators of *Z. armatum.*

	X_1	X_2	X_3	X_4	X_5	X_6	X_7	X_8	X_9	X_{10}	X_{11}	X_{12}
X_1	1											
X_2	−0.049	1										
X_3	0.054	−0.433	1									
X_4	−0.271	0.723**	0.418	1								
X_5	0.146	−0.005	0.497*	0.089	1							
X_6	0.249	0.064	−0.463*	−0.020	0.650**	1						
X_7	−0.19	0.233	0.271	0.135	−0.090	−0.035	1					
X_8	−0.037	−0.202	0.291	−0.433	−0.428	0.251	0.527*	1				
X_9	−0.043	0.018	0.108	−0.220	0.270	0.197	0.054	0.765**	1			
X_{10}	−0.054	−0.153	−0.114	0.022	−0.095	0.261	−0.236	0.302	0.546*	1		
X_{11}	0.303	0.109	−0.030	−0.067	0.014	0.195	0.356	0.148	−0.036	−0.101	1	
X_{12}	0.234	0.118	−0.087	0.021	0.382	−0.025	0.063	−0.314	−0.441*	−0.253	0.422	1

Note:(1) "*" was significantly correlated at the level of 0.05 (bilateral), "**" was significantly correlated at the level of 0.01 (bilateral).
(2) "X_1":△E, "X_2": "L*", "X_3": "a*", "X_4": "b*", "X_5": Rate of depressed oil droplet, "X_6": Degree of uniformity, "X_7": Volatile oil content, "X_8": Hydroxyl-α-sanshool content, "X_9": Hydroxyl-β-sanshool content, "X_{10}": Hydroxyl-γ-sanshool content, "X_{11}":Limonene content, "X_{12}": Linalool content.

of *Z. armatum* contained volatile oil. When they ruptured, the volatile oil flowed out and adhered to the surface of *Z. armatum*. After being left for a while, *Z. armatum* underwent browning, which was affecting its green color (Chen et al., 2019; Zhang, 2011). Then, hydroxyl-α-sanshool, hydroxyl-β-sanshool and hydroxyl-γ-sanshool were amides in *Z. armatum* to produce pungent taste, when the more intense the pungency of *Z. armatum*, indicating that the higher the content of them. Moreover, it has been demonstrated that the molecular weight of both hydroxyl-α-sanshool and hydroxyl-β-sanshool were 263 Da, these two types of sanshools could be converted to each other under certain conditions (including oxygen, light and acidic environment) (Luo et al., 2022a). The aroma substances of *Z. armatum* were mainly found in the volatile oil, which was associated with the oil droplet on the surface of *Z. armatum*. When the aroma of *Z. armatum* differed, its aroma component content would be different (Ji et al., 2019). The above indicators are the sensory, physical and chemical indicators of *Z. armatum*, which can reflect the main quality of *Z. armatum.*

Principal component analysis of quality indicators

SPSS 27.0 software was used for PCA on 12 indicators of *Z. armatum* from 21 producing areas, and the results were shown in Table 3. Table 3 shows the five principal components with their contributions of 24.164%, 19.592%, 15.566%, 13.785%, and 8.984%, respectively. The total contribution of these five principal components was 82.09%, while their eigenvalues were all greater than 1, which could reflect most information of *Z. armatum* quality. Therefore, these five principal components were selected to analyze the quality of *Z. armatum* (Zhao et al., 2023).

Table 3
Principal component analysis of quality indicators of *Z. armatum.*

Quality indicators	The principal components				
	1	2	3	4	5
△E	−0.075	−0.203	0.510	−0.506	0.233
L*	−0.339	0.651	0.151	0.441	0.212
a*	0.103	−0.871	−0.028	0.312	0.032
b*	−0.455	0.658	−0.178	0.414	0.200
Rate of depressed oil droplet	−0.574	−0.554	−0.126	0.161	0.516
Degree of uniformity	0.421	0.557	0.398	−0.381	−0.055
Volatile oil	0.062	−0.063	0.492	0.773	−0.227
Hydroxyl-α-sanshool	0.853	−0.169	0.314	0.286	0.004
Hydroxyl-β-sanshool	0.822	0.038	−0.015	0.218	0.373
Hydroxyl-γ-sanshool	0.552	0.187	−0.284	−0.081	0.627
Limonene	−0.581	−0.093	0.509	−0.168	0.227
Linalool	−0.074	−0.010	0.836	0.049	0.182
Eigenvalue	2.900	2.351	1.868	1.654	1.078
Variance contribution rate /%	24.164	19.592	15.566	13.785	8.984
Cumulative variance contribution rate /%	24.164	43.755	59.321	73.106	82.090

As shown in Table 3, hydroxyl -α -sanshool and hydroxyl -β -sanshool contributed more to the first principal component and had a larger positive load. The pungency of *Z. armatum* was mainly caused by hydroxyl-sanshools, in which hydroxyl -α -sanshool had a strong pungent sensation, so the first principal component mainly reflected the pungent taste of *Z. armatum* (Bryant and Mezine, 1999). Its function expression was:

Y_1 = $-0.044X_1-0.199X_2 + 0.060X_3-0.267X_4-0.337X_5 + 0.247X_6 + 0.036X_7 + 0.501X_8 + 0.483X_9 + 0.324X_{10}-0.341X_{11}-0.043X_{12}$.

The values of L*, a* and b* contributed a lot to the second principal component. Among them, L* and b* had a large positive load, while a* had a large negative load. These three indexes reflected different color attributes, so the principal component mainly reflected the color of *Z. armatum* (Khan et al., 2009). The function expression was:

$Y_2 = -0.132X_1 + 0.425X_2-0.568X_3 + 0.429X_4-0.361X_5 + 0.363X_6-0.041X_7-0.110X_8 + 0.025X_9 + 0.122X_{10}-0.061X_{11}-0.007X_{12}$.

Linalool content contributed more to the third principal component and had a larger positive load. Linalool was the main aroma component in the volatile oil of *Z. armatum*, and contributed the most to the aroma of *Z. armatum* (Liu et al., 2020). Therefore, the third principal component mainly reflected the aroma of *Z. armatum*. Its function expression was:

$Y_3 = 0.373X_1 + 0.110X_2-0.020X_3-0.130X_4-0.092X_5 + 0.291X_6 + 0.360X_7 + 0.230X_8-0.011X_9-0.208X_{10} + 0.372X_{11} + 0.612X_{12}$.

The content of volatile oil contributed greatly to the fourth principal component and had a large positive load. Volatile oil content was an important index to analyze the quality of *Z. armatum*, and was related to the aroma of *Z. armatum* (Zheng et al., 2021). The function expression was:

$Y_4 = -0.393X_1 + 0.343X_2 + 0.243X_3 + 0.322X_4 + 0.125X_5-0.296X_6 + 0.601X_7 + 0.222X_8 + 0.170X_9-0.063X_{10}-0.131X_{11} + 0.038X_{12}$.

The hydroxyl -γ -sanshool contributed more to the fifth principal component and had a larger positive load. Although the content of hydroxyl -γ -sanshool in *Z. armatum* was not as high as that of hydroxyl -α -sanshool, but some studies have proved that the higher the content of hydroxyl -γ -sanshool, the stronger the tingling sensation (Feng et al., 2023a). Its function expression was:

$Y_5 = 0.224X_1 + 0.204X_2 + 0.031X_3 + 0.193X_4 + 0.497X_5-0.053X_6-0.219X_7 + 0.004X_8 + 0.359X_9 + 0.604X_{10} + 0.219X_{11} + 0.175X_{12}$.

According to the characteristic values of five principal components, a comprehensive evaluation model of *Z. armatum* quality was established:

$$Y = 0.2943\ Y_1 + 0.2387\ Y_2 + 0.1896\ Y_3 + 0.1679\ Y_4 + 0.1094\ Y_5$$

According to the above evaluation model, the comprehensive score and ranking of *Z. armatum* from all producing areas were calculated. The results are shown in Table S2. On the basis of the model score, the producing areas with the highest scores of the first, second, third, fourth and fifth principal components(1 Content of hydroxyl -α -sanshool, hydroxyl -β -sanshool; 2 L*, a *, b* values; 3 Linalool content; 4 Volatile oil content; 5 Hydroxyl-γ -sanshool content) were ZT, EB, QW, LZ and DY, respectively. Among them, the highest comprehensive score was ZT, followed by DY, and then LZ. From this score, the producing areas with high content of hydroxyl-sanshools or limonene and linalool have higher score, which indicated that amides and aroma components were the two main indexes to evaluate *Z. armatum*.

Cluster analysis based on characteristic quality indicators

According to the results of PCA, R-type CA was carried out on 12 indexes of *Z. armatum*. Q-type CA was conducted on *Z. armatum* from 21 producing areas. The results are shown in Fig. 3(a–c).

Fig. 3(a) shows the R-type CA of *Z. armatum*. When the distance was 5, the 12 indicators of *Z. armatum* were divided into 3 groups. First, the contents of hydroxyl -β -sanshool, hydroxyl -γ -sanshool, △E, volatile oil, a* value, limonene and linalool were in the first group. The second was L* value, hydroxyl -α -sanshool content, b* value and rate of depressed oil droplet. The third was degree of uniformity. Based on the above results, the multiple correlation coefficient of each indicator in these three groups were calculated separately, and the larger the multiple correlation coefficient, the closer the linear correlation between the indicators. Combined with related studies, it was reported that color, pungent taste and flavor quality were important quality indicators and commercial traits of *Z. armatum*, therefore, the hydroxyl-sanshools content, linalool content and b* value were determined as key indexes to evaluate the quality of *Z. armatum* (Luo et al., 2022b; Zheng et al., 2020). In addition, these indexes were the positive loads in the first principal component, second principal component, third principal component and fifth principal component respectively. This finding suggested that the larger the values of these indexes were, the higher the model evaluation score would be.

Fig. 3(b) shows the Q-type CA of producing area based on 12 indicators of *Z. armatum*. When the Euclidean distance was 10, the 21 producing areas clustered into 4 groups. Group 1 had 15 producing areas. The common characteristics were as follows: the content of hydroxyl -sanshools was close to the average value, and most of them were higher than the average value; Linalool content was high, the difference between L*, a* and b* was small. Moreover, except HY-2, DL and MB,

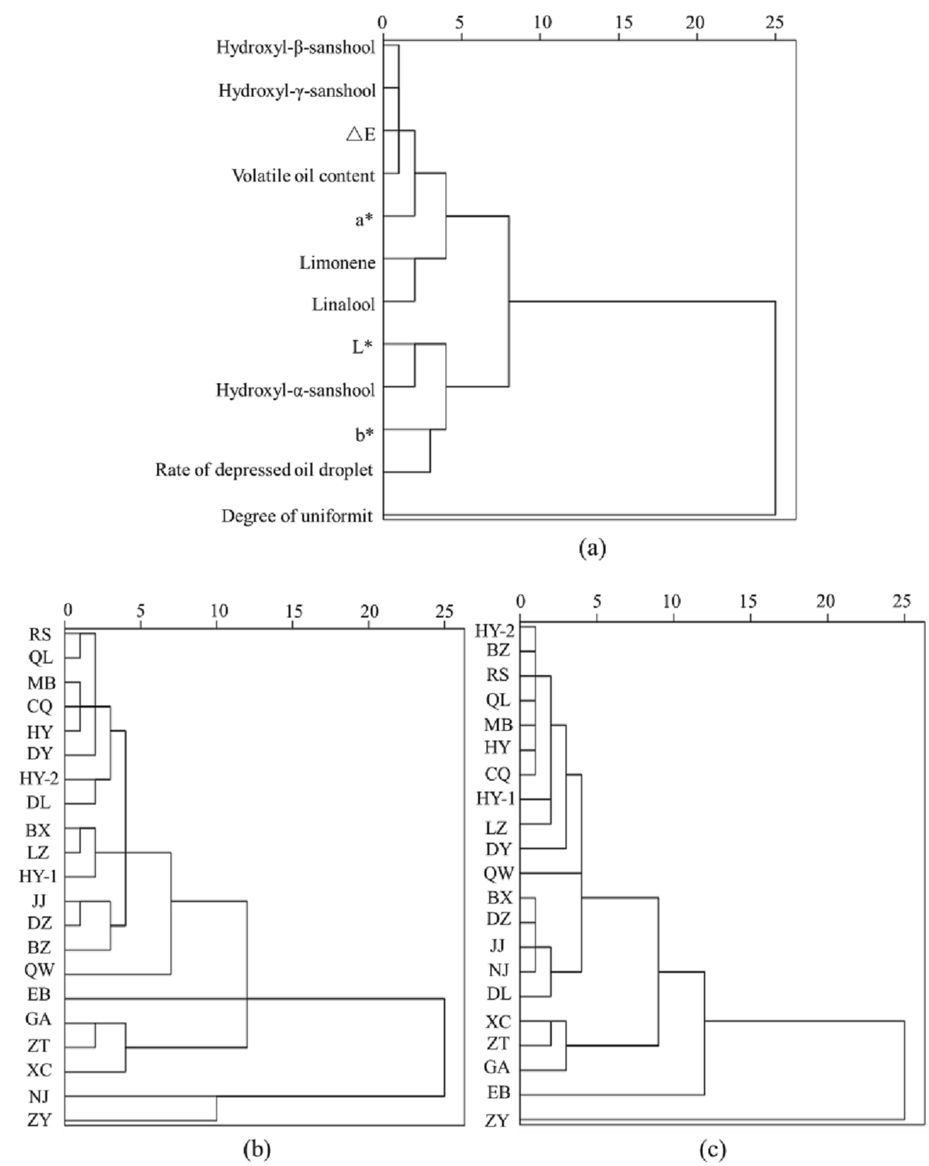

Fig. 3. R-type and Q-type cluster analysis of *Z. armatum*. (a): R-type CA of *Z. armatum* index, (b): Q-type CA based on 12 indicators of *Z. armatum*, (c): Q-type CA based on the 5 extracted principal components.

the comprehensive quality score of other producing areas ranked high and the quality was good, the three producing areas had low scores due to high rate of depressed oil droplet and △E. The second group was EB, the content of hydroxyl -α -sanshool, hydroxyl -β -sanshool, hydroxyl -γ -sanshool from this producing area were obviously different from that of other producing areas, and were lower than the average value. Therefore, it was grouped into a separate category. The third group was GA, ZT and XC. The general characteristics were hydroxyl -α -sanshool content, hydroxyl -β -sanshool content, rate of depressed oil droplet and △E were relatively close. Therefore, these three producing areas were clustered into one group. However, their principal component scores varied considerably, ranking 7th, 1st and 16th, respectively. It was due to the high content of hydroxyl-sanshools in GA, but the low content of linalool, limonene, and L* and b* values. Then, the content of linalool, limonene and hydroxyl -γ -sanshool in XC were low, therefore, the overall scores were lower. The fourth group was NJ and ZY, ranking 19th and 21th in overall scores. They were characterized by lower content of hydroxyl-sanshools, the same rate of depressed oil droplet (33%), the comprehensive quality was poor.

Fig. 3(c) shows the Q-type CA of the producing area based on the five principal components extracted. When the Euclidean distance was 10, the 21 producing areas were clustered into 3 groups. Thereinto, *Z. armatum* of ZY was clustered into one group because of the lowest content of hydroxyl-sanshools, volatile oil and linalool. EB was grouped into one group because the content of hydroxyl-sanshools was only higher than ZY, and the values of L*, b* were the highest and the values of a* were the lowest. Other producing areas were in one group.

Comparing Fig. 3(b) and (c), the former divided the 21 producing areas into four groups, while the latter divided them into three groups. Since Fig. 3(b) involved more quality indicators when performing CA, it could not accurately group the producing areas with better overall quality of *Z. armatum* into one group. For example, the third group of three production areas differed greatly in their overall quality ranking, being 7th, 1st, and 16th, respectively. However, Fig. 3(c) was mainly based on the five principal components extracted by PCA, which were consistent with the key indexes identified by the Q-type CA. Therefore, it could more accurately and effectively distinguish the producing areas with poorer main quality indexes in *Z. armatum*.

Conclusions

In this study, the quality indexes of *Z. armatum* in 21 production areas were examined. The results showed that the quality indexes of *Z. armatum* all had different degrees of variation, and there was a certain correlation between the indexes, such as L* and b* value, rate of depressed oil droplet and degree of uniformity, hydroxyl -α -sanshool and hydroxyl -β -sanshool content were significantly positive correlation ($P < 0.01$). In addition, five principal components were extracted from the 12 indexes of *Z. armatum* by PCA (1 Content of hydroxyl -α -sanshool, hydroxyl -β -sanshool; 2 L*, a*, b*; 3 Linalool content; 4 Volatile oil; 5 Hydroxyl-γ -sanshool content), and their contribution rate reached 82.09%. Meanwhile, a standardized evaluation model was constructed as $Y = 0.2943Y_1 + 0.2387Y_2 + 0.1896Y_3 + 0.1679Y_4 + 0.1094Y_5$. Through this model, *Z. armatum* from ZT, DY and LZ were ranked higher. On this basis, hydroxyl - sanshools content, linalool content and b* value were determined to be the key indexes for evaluating the quality of *Z. armatum* by R-type CA, which were consistent with the five principal components extracted by PCA. Then, Q-type CA was performed on 21 production areas based on 12 indicators of *Z. armatum* and 5 principal components extracted by PCA, respectively. The results showed that the latter could distinguish the quality of *Z. armatum* more precisely. To sum up, the evaluation model established in this study was convenient and effective, and could provide a reference for *Z. armatum* quality evaluation and in-depth product development.

CRediT authorship contribution statement

Jiahui Liu: Resources, Methodology, Data curation, Writing – original draft. **Junzhe Wan:** Methodology, Data curation. **Yu Zhang:** Methodology, Data curation. **Xiaoyan Hou:** Methodology, Formal analysis. **Guanghui Shen:** Software, Data curation. **Shanshan Li:** Software, Data curation. **Qingying Luo:** Supervision, Software. **Qingye Li:** Supervision, Software. **Man Zhou:** Visualization, Investigation. **Xingyan Liu:** Visualization, Investigation. **Chenggang Wen:** Conceptualization. **Xiang Zhu:** Conceptualization, Funding acquisition, Supervision. **Zhiqing Zhang:** Funding acquisition, Supervision, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The authors do not have permission to share data.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fochx.2023.100721.

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