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Arsenic accumulation in *Scutellaria baicalensis* Georgi and its effects on plant growth and pharmaceutical components

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ABSTRACT

Scutellaria baicalensis Georgi is a traditional Chinese medicinal plant. The effects of arsenic (As) on the growth and the formation of pharmaceutical components of *S. baicalensis*, and the uptake and accumulation of As by *S. baicalensis* were investigated using a field pot-culture experiment. The results show that spiking low concentrations of As ($\leq$100 mg kg^{-1}) into soils can hasten the growth and development of the roots. High levels of As, however, reduced plant growth. The concentrations of five flavone components were not significantly affected by spiking low concentrations of As ($\leq$200 mg kg^{-1}) into soils. High levels of As inhibited the generation of baicalin and wogoninside, but facilitated the generation of baicalein, wogonin and oroxylin A in *S. baicalensis* Georgi. The concentration of As in each part of the plant was proportional to the concentration of As spiked into the soil. The application of phosphorus (P) to the soil promoted the uptake and accumulation of As in the roots of the plant, but this synergistic effect became weaker with the incremental addition of P. Dry biomass did not change in response to low levels of P addition ($\leq$200 mg kg^{-1}) to soils, but it increased significantly under high levels of P. Based on the results of both this pot-culture experiment and human health risk assessments, maximum safety limits of 2.0 mg kg^{-1} of As in the roots of *S. baicalensis* Georgi and 70 mg kg^{-1} of As in cultivated soils are suggested.

1. Introduction

Arsenic (As) is a naturally occurring toxic element in the environment. It can enter the environment through weathering, biological activity, and volcanic activity. Anthropogenic inputs from agricultural and industrial practices, such as the application of pesticides and chemical fertilizers, wastewater irrigation, precipitation from heavy coal combustion and smelter wastes and residues from metalliferous mining, increase the levels of As contamination in soil, ground water and surface water [1–4]. In China, the concentration of As typically varies from below 5 mg kg^{-1} in non-contaminated soils [5] to as high as 3800 mg kg^{-1} in contaminated soils near the tailing spots of arsenic sulphide mines [6]. Once in the soil, As can be absorbed by plants, including farm crops such as grains, vegetables and fruits, and the ingestion of these contaminated farm crops can have hazardous effects on human health. Chronic exposure to high levels of inorganic As has been found to result in a variety of adverse health effects, including skin and internal cancers and cardiovascular and neurological effects [2,7].

With the boom in the use of natural herbal medicine in the world, traditional Chinese medicine has been increasingly exported. Consequently, the quality and safety of traditional Chinese medicine has drawn more attention in the world. Quality and safety standards for herbal medicine that clearly stipulate the maximum allowable value of heavy metals in herbal medicine have been enacted and put into effect by many countries [8–10]. However, since most of these standards are determined by referring to the quality standards for foods, they are not based on the research on herbal medicine. Unlike nutritional components such as starch, lipids and amino acids, pharmaceutical components in medicinal plants (referring to original plant of traditional Chinese medicine hereafter) are commonly secondary metabolites, and some of them are produced in much higher concentrations under environmental stress [11,12]. The uptake and accumulation of heavy metals may have impacts on medicinal plants that are different from their impacts on farm crops. Moreover, the methods of processing and ingesting medicinal plants are also different from those for farm crops, which should translate into differences in human exposure to and health risks from heavy metals between medicinal plants and

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farm crops. Therefore, it is necessary to improve quality standards for herbal medicines by examining and revising the maximum allowable values of heavy metals in medicinal plants, using research based on medicinal plants.

To date, research on the safety of traditional Chinese medicine has mainly been focused on the development of methods for detecting heavy metals in traditional Chinese medicines [13–15] and the investigation of the level of contamination of traditional Chinese medicines by heavy metals [16,17]. Over 300 species of medicinal plants have been investigated. The results indicate that the concentration of heavy metals in medicinal plants differs significantly depending on the production area and that, even within the same plant, the concentrations of heavy metals may differ among parts of the plant [17]. There has been little research aimed at elucidating the uptake mechanisms, accumulation and partitioning of heavy metals in medicinal plants or at determining the response of medicinal plants to heavy metals in terms of their growth and their production of pharmaceutical components. However, as a reference, we can use the theories, methods and techniques taken for both studies on the hyper-accumulation of heavy metals in plants for soil remediation [18,19] and research on the harmful effects of heavy metals and their mechanisms in farm crops [20,21]. Furthermore, some research has examined the adequacy of environmental quality standards based on the results of risk assessments of human beings exposed to heavy metals in crops [22]. These studies could help inform us for the formulation and revision of quality and safety standards for medicinal plants.

The roots of *Scutellaria baicalensis* Georgi (Labiatae) are an important Chinese medicine that is used as a diuretic, laxative, febrifuge, an antipyretic and for hemoptysis, bloody stool, and nasal haemorrhaging when prescribed together with other herbs [10]. A major component of this plant, baicalein, has antibacterial, antiviral and lipoxygenase inhibitory activities [23,24]. Many Chinese medical preparations containing mainly *S. baicalensis* Georgi, such as “San Huang Pian”, “Ku Gan Chong Ji” and “Yin Huang Kou Fu Ye”, have been developed and used widely in China as drugs with antibacterial and antiviral functions, partly substituting for antibiotics. Over 300 species of plants belonging to *Scutellaria* Linn. grow throughout the world, except for the tropical areas of Africa. *S. baicalensis* Georgi is one species of *Scutellaria* Linn., and it is widely distributed in most provinces of China. Chengde in Hebei province and Chifeng in Inner Mongolia municipality are two of the largest areas of production of natural *S. baicalensis* Georgi. Shandong province also has major plantations of *S. baicalensis* Georgi [25].

The objectives of this paper are: to clarify the accumulation and partitioning of As in *S. baicalensis* Georgi and the influence of P in soil; to elucidate the responses of *S. baicalensis* Georgi to As in terms of both its growth and its production of five major active flavone components, baicalin, wogoninside, baicalein, wogonin and oroxylin A; and furthermore, to provide suggestions for the establishment of environmental safety standards for As in *S. baicalensis* Georgi.

2. Materials and methods

2.1. Field site and soil characterization

The field pot-culture experiment was conducted at the Fangshan Experimental Station (39°41′N and 116°03′E), which belongs to Beijing Normal University and is located in the suburb of Beijing. The altitude is 38 m. The experimental station is in the temperate zone with a semi-moist continental climate, an average annual temperature of 11.6 °C and an average annual precipitation of 611 mm.

Most soil samples were collected from the surface (0–20 cm) of a field in the station. This soil was a yellow sandy loam; total N, total P, total K and pH were 0.055%, 0.043%, 2.02% and 7.34, respectively. The concentrations of exchangeable Ca, exchangeable Mg, Al, S, Fe and Mn were 0.45%, 0.015%, 6.22%, 0.019%, 2.92% and 0.059%, respectively. The concentration of As in the soil was 12.2 mg kg^{-1}.

2.2. Experimental procedure

Eighty-five plastic pots (27.5 cm in height and 30 cm in diameter) were used. The same quantity of soil (20 kg) was collected from a field in the Fangshan Experimental Station and placed into each pot. Two-year old striking roots of *S. baicalensis* Georgi were purchased from the plantation in the Wafangdian, Liaoning province. In April, three seedlings of striking roots of *S. baicalensis* Georgi of similar sizes were transplanted into each pot before turning green. To simulate field conditions, the plants were grown under open field conditions and no fertilizer was added. Any loss in water was alleviated by using tap water to sustain the water holding capacity at 16%.

In this study, disodium hydrogen arsenate [$Na_2HAsO_4{\cdot}7H_2O$] and sodium dihydrogen phosphate (sodium phosphate) [NaH_2PO_4] were used without further purification for the As and P treatments. The arsenic and phosphate solutions were prepared by mixing the appropriate amount of disodium hydrogen arsenate and sodium dihydrogen phosphate into the suitable volume of purified water (no As detected) for each treatment. Preliminary tests were performed to determine the appropriate range of As concentrations to use for testing the plants sensitivity. The As addition only (nine levels of As at 0 (CK1), 10, 20, 40, 100, 160, 200, 400 and 600 mg kg^{-1}, no P addition) and the As and P addition (eight levels of P at 0 (CK2), 10, 20, 40, 100, 200, 400 and 600 mg kg^{-1}, all with As at 40 mg kg^{-1}) experiments were arranged to investigate the uptake and accumulation of As by *S. baicalensis* Georgi and the influence of the soil P concentration on the accumulation of As. The solutions were carefully irrigated into the soil to avoid trickling any onto the leaf. All treatments were replicated five times in the experiments.

The experimental treatments were conducted three and a half months after the seedling transplantation. Sixty days later at the end of September, the plants were harvested, when both the aboveground parts and the roots experienced their peak growth periods and reached their physiological maturity. The whole plant was scooped out. The attached soil was gently removed from the roots by a soft brushing. The roots and the aboveground parts were dried in an oven at 60 °C for 72 h until completely dry. The fresh weight and dry weight (after 72 h at 60 °C) of *S. baicalensis* Georgi were measured.

2.3. Plant and soil analysis

The dried plant samples were ground in a mill. After subsampling for the analysis of flavone components, the ground plant samples were sifted through a 50-mesh (0.30 mm) sieve. 0.1 g of plant samples were then digested in 4 ml of pure HNO_3, using a microwave digestion system (WX-8000). The soil samples were air-dried and ground to sift through a 100-mesh (0.15 mm) sieve. 0.2 g of soil samples were then digested with an 8 ml solution of 1:3 HNO_3: HCl, using the WX-8000 microwave digestion system. The As concentrations were determined by using hydrogen generation atomic fluorescence spectroscopy (AFS-830). The subsample of the dried plant samples were sieved through a 40-mesh (0.42 mm) sieve. 0.1 g of plant subsamples were ultrasonically extracted in a conical breaker with 25 ml of 70% ethanol for 40 min, and then the extracts were separated by filtering them through 0.45 μm membrane filters. The concentrations of five flavone components (baicalin, wogoninside, baicalein, wogonin and oroxylin A) in *S. baicalensis* Georgi were determined by using high performance liquid chromatography (Waters1525 HPLC/2487PDA detector/Mellennium32 server).

2.4. Statistical analysis

The means and standard deviations (SD) were calculated by the Microsoft Office Excel 2003. Correlation analysis, regression analysis and one-way analysis of variance (ANOVA) were carried out with SPSS13.0. In ANOVA procedure, when a significant ($P < 0.05$) difference was observed between treatments, multiple comparisons were made by the LSD test.

3. Results and discussion

3.1. Response of plant growth to different treatments of As

Fig. 1 shows the effects of soil As treatments on the dry biomass of *S. baicalensis* Georgi. The results show that low concentrations of As ($\leq$100 mg kg^{-1}) spiked into soils can stimulate the growth and development of the roots (100 mg kg^{-1}, $P < 0.05$), but they have no significant effect on the growth of the shoots (the aboveground parts, including stems and leaves). When the As treatment concentration was increased to 200 mg kg^{-1}, the shoots first displayed a decrease in biomass, while the biomass of the roots was still greater than that of the CK1 (the control). When the As treatment concentrations exceeded 400 mg kg^{-1}, however, adverse effects on the growth of both shoots ($P < 0.05$) and roots were observed. The total biomass of *S. baicalensis* Georgi increased at lower concentrations of As, but it then decreased significantly with the incremental increase of As spiked into the soil (400 and 600 mg kg^{-1}, $P < 0.05$). When the concentration of added As exceeded 400 mg kg^{-1}, plants were wilted and stunted, with chlorosis and necrosis at the leaf tips and margins, and they died, resulting in a 53.7% and 71.7% reduction in total dry matter production at the As treatment levels of 400 and 600 mg kg^{-1}, respectively, compared to that of the CK1.

This result agrees with those of some studies on crops and fruits. Chen and Liu [26] found that low levels of As in the soil could stimulate the growth and development of rice and increase the yield, while high levels of As impaired growth and development significantly. Similar results were reported by Jian et al. [21]: their pot experiment showed that low concentrations of As boosted the growth of *Brassica chinensis*, but beyond a certain limit, production fell rapidly until death occurred. The limits for As exposure varied according to the soil type used for cultivation. It is possible that arsenate additions may displace phosphate from the soil in certain situations, and thus, cause a resultant increase in plant-available P [27]. However, Carbonell-Barrachina et al. [28] also observed a positive growth response in their hydroponic culture experiment that used soil-free systems, and although they also thought that this positive plant growth response might have been linked to P nutrition. Since As can be substituted for P in plants, but it is unable to carry out the role of P in energy transfer, the plant reacts as if there is a P deficiency [29]. Thus, as plant As increases, the plant reacts by increasing P uptake [30]. In contrast to the above findings, the stimulation of growth by As additions (as arsenate) has not been observed in other pot experiments of rice [20,31,32], wheat and maize [30], cucumber, kidney bean and cabbage [33], maize (*Zea mays*), rapeseed (*Brassica napus*) and sunflower (*Helianthus annuus*) [34]. Further studies should be conducted to determine the necessary conditions and reasons for the stimulation of growth by As additions (as arsenate).

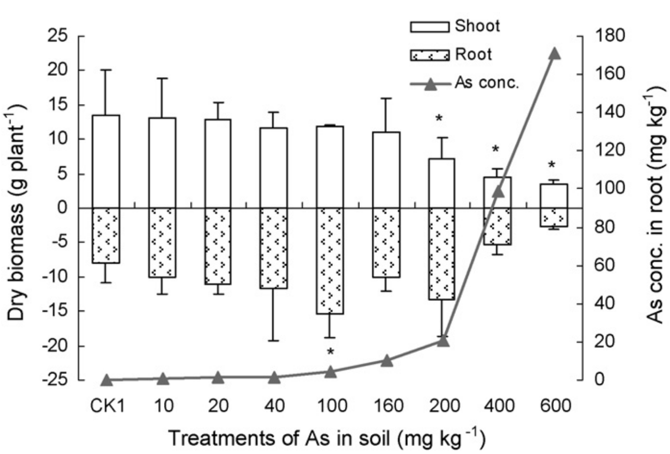

Fig. 1. The effects of As stress on As concentration in the roots and on dry biomass of *S. baicalensis* Georgi. *Note*: The data are reported as means ± S.D. ($n = 3$). One-way analysis of variance was carried out to determine if a significant difference was observed between treatments and the control (CK1), "*" means $P < 0.05$ (the LSD test).

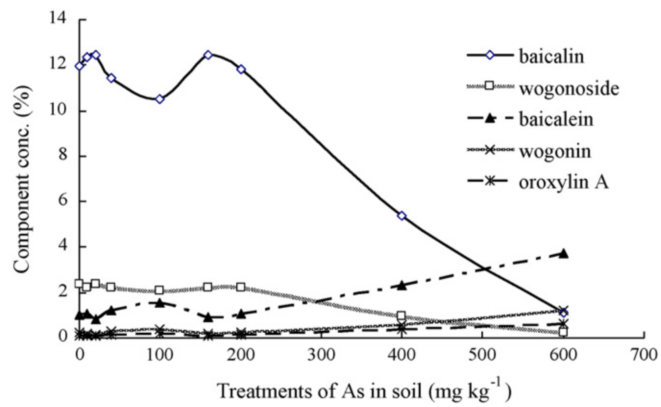

Fig. 2. Effect of As stress on the contents of five flavone components in roots.

3.2. Response of plant pharmaceutical components to different treatments of As

Fig. 2 shows the effects of soil As treatments on the content of pharmaceutical components in *S. baicalensis* Georgi. The results reveal that there was no significant effect on the contents of five flavone components examined at low concentrations of As addition ($\leq$200 mg kg^{-1}) to soils. However, it was observed that high levels of As caused a reduction in the accumulation of baicalin and wogonoside, but these As concentrations resulted in an increase in the contents of baicalein, wogonin and oroxylin A in *S. baicalensis* Georgi. It is interesting that with the increase in the treatment concentration of As, baicalein changed in a trend opposite to that of baicalin, and the change in wogonin was opposite to that of wogonoside. The results of correlation analysis show that the concentration of baicalin in the roots of *S. baicalensis* Georgi has a significant negative correlation with that of baicalein ($P < 0.01$), and the concentration of wogonin in the roots has a significant negative correlation with that of wogonoside ($P < 0.01$) (Table 1). The concentrations of baicalin and wogonoside have negative correlations with the As treatment concentrations ($P < 0.01$), and the concentrations of baicalein, wogonin and oroxylin A have positive correlations with the As treatment concentrations ($P < 0.01$). Chinese Pharmacopoeia [10] stipulates that baicalin in *S. baicalensis* Georgi cannot be lower than 9.0%. In our experiment, when the As treatment concentration was lower than 200 mg kg^{-1}, the baicalin content met this requirement.

The mechanisms for the change in the concentrations of flavone components in plants under As stress have been discussed. Abi-

Table 1
Correlations between soil As treatment concentrations and the contents of five flavone components in *S. baicalensis* Georgi.

	As in soil	Baicalin	Wogonoside	Baicalein	Wogonin	Oroxylin A
As in soil	1					
Baicalin	−0.939**	1				
Wogonoside	−0.956**	0.994**	1			
Baicalein	0.923**	−0.989**	−0.981**	1		
Wogonin	0.903**	−0.975**	−0.963**	0.993**	1	
Oroxylin A	0.948**	−0.995**	−0.990**	0.996**	0.985**	1

** Denotes correlations significant at $P = 0.01$ (2-tailed).

otic stresses, such as drought, salinity, extreme temperatures and chemical toxicity, lead to secondary osmotic or oxidative stresses to plants [35]. Early in 1932, Miwa [36] found the existence of a β-glucuronidase (GUS) called baicalinase in *S. baicalensis* Georgi. In response to an elicitor, the cells immediately initiate the hydrolysis of baicalein 7-O-b-D-glucuronide (baicalin) by β-glucuronidase, and the released baicalein is then quickly oxidized to 6,7-dehydrobaicalein by the cell wall peroxidases. Hydrogen peroxide is effectively consumed during the peroxidase reaction [23]. Therefore, the hydrolysis of baicalin into baicalein has been thought to be related to oxidative stress. In our study, when the soil As stress exceeded 400 mg kg^{-1}, baicalin decreased rapidly, while baicalein increased correspondingly, but not to the extent of the decrease in baicalin. This result could be explained by the metabolic mechanism of H_2O_2 by β-glucuronidase identified by Morimotos' work [23], but this proposed mechanism still needs experimental verification. The changes in the concentrations of wogoninside and wogonin in response to the increase of As stress in our study imply that the turnover of wogoninside to wogonin may occur by the same mechanism.

3.3. The uptake of As and its partitioning in S. baicalensis Georgi

Fig. 1 shows the As concentration in the roots under As stress. Table 2 shows the amount and distribution of As after uptake and accumulation in the whole plant of *S. baicalensis* Georgi. The concentration of As in each part of the plant was proportional to the As treatment concentration. When soil As treatment concentrations were no more than 200 mg kg^{-1}, the concentrations of As in both the shoots and roots increased gradually with the incremental increase in the treatment level of As in the soil. When As treatment concentrations surpassed 400 mg kg^{-1}, however, the concentrations of As in both the shoots and the roots increased abruptly by a large amount. From observations of plant growth, we know that above this As treatment concentration of 400 mg kg^{-1} the plants exhibited a response to poisoning, and they died. This result was in accordance with the report of Richardson that within a certain threshold the absorption of As by plants had a positive correlation to the As content in the environment, and beyond its tolerance limit the restraining mechanism collapsed and the plant absorbed large quantities of As and showed the symptoms of poisoning, and even death [37].

Table 2
The uptake and partitioning of As in *S. baicalensis* Georgi under As stress.

As treatment (mg kg^{-1})	As concentration (mg kg^{-1})		As distribution factors	
	Root	Shoot	BF	TF
CK1	0.34	0.15	0.03	0.44
10	1.01	0.40	0.07	0.40
20	1.28	0.65	0.05	0.51
40	1.44	1.86	0.04	1.29
100	4.74	2.36	0.06	0.50
160	10.1	6.04	0.12	0.60
200	20.4	14.5	0.19	0.71
400	99.0	572	0.28	5.78
600	171	819	0.26	4.79

Note: The bioaccumulation factor (BF) is defined as the ratio of the As concentration in the shoots to that in the soil; and the transfer factor (TF) is defined as the ratio of the As concentration in the shoots to that in the roots.

The bioaccumulation factor (BF), defined as the ratio of the As concentration in the shoots to that in the soil, reflects the capacity of a plant to absorb and accumulate As from soil. The transfer factor (TF), defined as the ratio of the As concentration in the shoots to that in the roots, measures the effectiveness of a plant in transferring As from roots to shoots. When the As treatment concentration in soil exceeded 400 mg kg^{-1}, the tolerance limit, the BF was larger than 0.25 and the TF was larger than 4.79; this means that the roots absorbed much more As from the soil, the plant transferred most of the As from the roots to the shoots, and the shoots accumulated As. When the As treatment concentration in the soil was lower than 200 mg kg^{-1}, however, the BF was in the range of 0.03–0.19 and the TF was mostly in the range of 0.44–0.71. The concentrations of As in the roots were, in most of these As treatments, higher than those in the shoots; this means that the ability of *S. baicalensis* Georgi to uptake and translocate As was weak, more than half (57.3–72.4% in weight) of As taken up by the whole plant was retained in the roots. The above results show that *S. baicalensis* Georgi has no specific uptake and accumulation capacity for As. In other words, *S. baicalensis* Georgi is not a hyper-accumulator of As.

A regression analysis was conducted based on the data for six of the soil As treatments, excluding the two that exceeded the tolerance limit of 400 mg kg^{-1}. The corresponding relationships for the roots and shoots can be expressed using the following quadratic regression equations:

$$Y_{\text{root}} = 0.0006X^2 - 0.0324X + 1.2064 \quad (R^2 = 0.98,\ P < 0.01) \tag{1}$$

and

$$Y_{\text{shoot}} = 0.0005X^2 - 0.0374X + 1.0047 \quad (R^2 = 0.95,\ P < 0.01), \tag{2}$$

where Y_{root} and Y_{shoot} are the As concentrations in the roots and shoots, respectively, and X is the soil As treatment concentration.

3.4. The influence of P in soil on As accumulation in S. baicalensis Georgi

Fig. 3 shows the effects of P addition to soil on the growth of *S. baicalensis* Georgi. It should be noted that the same amount of As (40 mg kg^{-1}) has been added to the soil for each P treatment. There were no significant effects of soil P on the dry biomass of either shoots or roots under low levels of P addition (<400 mg kg^{-1}). When the concentration of additional P exceeded 400 mg kg^{-1}, however, the dry biomass of both shoots and roots increased, but these increases were not statistically significant compared to the CK2. Thus, it was concluded that P could stimulate the growth of *S. baicalensis* Georgi, but growth was only elevated by the addition of a high concentration of P. Fig. 4 shows that no significant effects

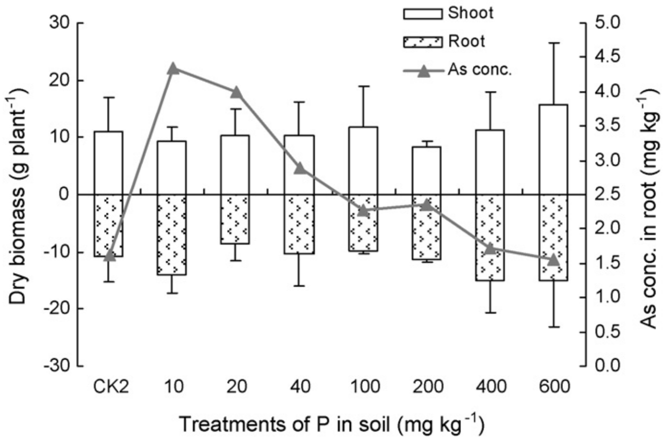

Fig. 3. The effects of P addition on As concentration in the roots and on dry biomass of *S. baicalensis* Georgi. *Note*: The same amount of As (40 mg kg^{-1}) was added to the soil for each P treatment. The data are reported as means ± S.D. (n = 3). One-way analysis of variance was carried out, no significant difference (P < 0.05, the LSD test), however, was observed between any treatment and the control (CK2).

of P addition were observed on the contents of the five flavone components examined.

Fig. 3 also shows that the As concentrations in the roots of the plants under most P treatment levels are higher than those of the CK2, except for under the highest level of P addition, 600 mg kg^{-1}. The As concentrations in the roots of *S. baicalensis* Georgi had the trend of decreasing with the incremental increase in the P treatment concentration from 10 to 600 mg kg^{-1}. This means that low levels of the application of fertilizer containing P promoted the absorption of As, but this synergistic action became weaker and weaker with the incremental increase in the level of P addition.

Some studies have been conducted to investigate the influence of P addition on the growth of and As accumulation in *Pteris vittata* L. A field experiment by Liao et al. [38] showed that increasing the amount of added P enhanced yields of *P. vittata*. The application of phosphate fertilizer enhanced the As concentration in the plant, and the As concentration in the above ground part of *P. vittata* was decreased by excessive P addition. In their sand culture experiment, Huang et al. [39] found that the higher the concentrations of phosphate, the lower the accumulation of arsenate (As(V)) and arsenite (As(III)) in the fronds of *P. vittata*., and the authors speculated that it may be in part due to competition between P and As(V) during transport. Finally, Tu and Ma [19] reported that P inhibited As uptake at all concentrations in the hydroponic experiment. In summary, it has been concluded that the effect of P on the uptake and accumulation of As in plants may be related to culture conditions and may be different from plant to plant, and that more experiments are necessary to clarify the joint effects of As and P.

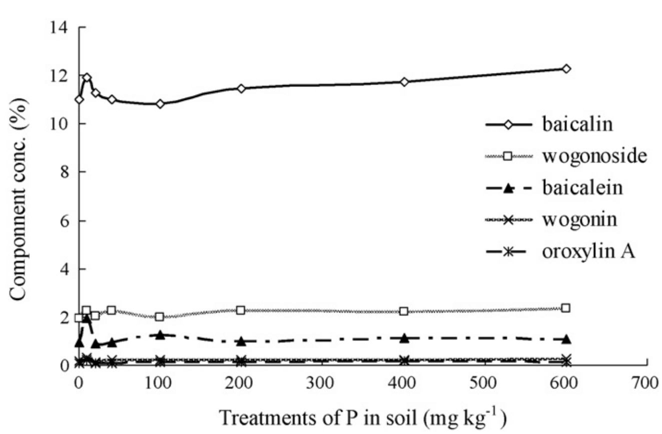

Fig. 4. Effects of P addition on the contents of five flavone components in the roots. *Note*: The same amount of As (40 mg kg^{-1}) was added to the soil for each P treatment.

3.5. The suggested maximum safety limit of As in S. baicalensis Georgi and in cultivated soil

The part of *ScutellariaS. baicalensis* Georgi that is used as a medicine is the roots. The suggested maximum safety limit of As in *S. baicalensis* Georgi was determined based on two considerations: (1) that the As concentration in the roots of *S. baicalensis* Georgi does not lead to harmful health impacts to human beings who ingest the usual dosage of medical preparations containing *S. baicalensis* Georgi and (2) by referring to relevant existing standards. The "Green trade standards of importing & exporting medicinal plants & preparations" [40] sets the maximum statutory limit of As in medicinal plants and preparations as no more than 2.0 mg kg^{-1}, and the maximum limit of As was stipulated as 1 mg kg^{-1} for only Fucus and Inulin in British Pharmacopoeia [8]. Our previous study [41] determined a maximum limit for As of 2.0 mg kg^{-1} in *S. baicalensis* Georgi, based on human health risk assessments of ingesting medical preparations containing *S. baicalensis* Georgi. In this previous study, the World Health Organization (WHO)'s tolerable daily intake (TDI) of 2.1 μg kg^{-1} d^{-1} was used, and the As that came from the ingestion of medicine was assumed to be no more than 1% of total As intake [42]. In summary, the suggested maximum safety limit of As in *S. baicalensis* Georgi is 2.0 mg kg^{-1}.

The suggested maximum safety limit of As in cultivated soil was determined based on three considerations: (1) the As in soil will not affect the growth of medicinal plants much; (2) the As in soil will not affect the accumulation of pharmaceutical components in medicinal plants much; (3) most importantly, the As level in soil will not lead to As concentrations in medicinal plants that exceed the maximum safety limit. *S. baicalensis* Georgi was not a hyper-accumulator of As, and the As concentration in *S. baicalensis* Georgi increased gradually with the incremental increase in the As treatment level in soil. Thus, controlling the As concentration in cultivated soils could effectively decrease As uptake and accumulation in *S. baicalensis* Georgi and ensure the safety of its usage as an herbal medicine. From the results of regression analysis of the relationship between soil As treatment concentrations and As concentrations in the roots of plants, it can be estimated that when the As treatment concentration in cultivated soil is less than 72 mg kg^{-1} the As concentrations in the roots of *S. baicalensis* Georgi are below the maximum statutory limit of 2.0 mg kg^{-1} under our experimental conditions. When the As treatment level was lower than 100 mg kg^{-1}, there was no reduction in the dry biomass of *S. baicalensis* Georgi. Chinese Pharmacopoeia stipulates that baicalin in *S. baicalensis* Georgi cannot be lower than 9.0%. In our experiment, when the As treatment concentration in soil was lower than 200 mg kg^{-1}, the baicalin content in roots of the plant was larger than 10.0%. In summary, the suggested maximum safety limit of As in cultivated soil is 70 mg kg^{-1}.

4. Conclusions

S. baicalensis Georgi has no specific uptake and accumulation capacity for As. Within a certain threshold, the accumulation of As in plants had a positive correlation to the As content in the soil. Beyond the tolerance limit of 400 mg kg^{-1}, the restraining mechanism collapsed, the plant absorbed a large quantity of As and showed symptoms of poisoning, they died. Thus, controlling the As concentration in cultivated soils could effectively decrease As uptake and accumulation in *S. baicalensis* Georgi and ensure the safety of its usage as an herbal medicine. Soil As treatment concentrations below 200 mg kg^{-1} had no significant adverse effects on either contents of five flavone components or dry biomass of the roots. These results enable us to formulate or revise the safety standards for this herbal medicine scientifically, and also allow us to direct the cultivation of this herbal medicine in terms of both the selection of plantation location and the appropriate agricultural management practices in order to provide people with a safe, high-quality herbal medicine.

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