Taylor & Francis
Taylor & Francis Group

RESEARCH ARTICLE

The effects of *Salvia przewalskii* total phenolic acid extract on immune complex glomerulonephritis

Yang Yang[a,b,c], Zhi-Peng Wang[b], Shou-Hong Gao[b], Hong-Qi Ren[c], Ren-Qian Zhong[a] and Wan-Sheng Chen[b]

[a]Department of Laboratory Diagnostics, Changzheng Hospital, Second Military Medical University of CPLA, Shanghai, China; [b]Department of Pharmacy, Changzheng Hospital, Second Military Medical University of CPLA, Shanghai, China; [c]Affiliated Huaihai Hospital of Xuzhou Medical University (The 97th Hospital of CPLA), Xuzhou, China

ABSTRACT

Context: *Salvia przewalskii* Maxim. (Lamiaceae) is a Chinese herbal medicine that has long been used for the treatment of cardiovascular disease.

Objective: The study investigated the therapeutic efficacy of *S. przewalskii* total phenolic acid extract (SPE) on immune complex glomerulonephritis (ICG) in rats.

Materials and methods: Sixty-two Wistar rats were randomized into six groups. ICG was induced in all groups except normal control group. SPE was administered intragastrically at 24 h intervals for 40 consecutive days. Urine protein (UP), total serum protein (TSP), serum albumin (SA), serum cholesterol (SC) and serum urea nitrogen (SUN) were measured one day before, on day 20 and 40 after SPE administration. On day 40 after SPE administration, the kidneys were removed and prepared into pathologic sections. In addition, kidney wet mass was measured for calculating the kidney wet mass coefficient (KWMC).

Results: UP excretion was reduced significantly on day 20 after SPE administration in all three SPE groups as compared with that in medium group, and this effect was observable continuously until 40 days after SPE administration. Compared with medium group, TSP and SA were increased in all three SPE groups after 40 days treatment, while SC and SUN were decreased. KWMC was decreased significantly in 100 mg/kg SPE group after 40 days treatment compared with that in medium group. Histopathologic analyses showed that renal inflammatory infiltration and kidney intumesce were alleviated in all three SPE groups.

Conclusions: SPE may be a potential therapeutic drug for glomerulonephritis.

Introduction

Salvia (Lamiaceae) is an important plant genus consisting of about 1000 species around the world (Huang and Sun 1977). More than 40 species of *Salvia* have been utilized as medical remedies in China (Li et al. 2008). *Salvia przewalskii* Maxim., called *Ganxishuweicao* in Chinese, is a herbaceous perennial. It mainly grows in Gansu, Sichuan, Yunnan and Tibet provinces of China, and has long been used as a traditional Chinese herbal remedy for the treatment of cardiovascular diseases by local inhabitants. The main chemical components of *S. przewalskii* are volatile oil (Liu et al. 2006), diterpenoids (Yang et al. 2008; Ohsaki et al. 2011; Jiang et al. 2013; Xue et al. 2014; Wang et al. 2015), triterpenoids (Wang et al. 1988; Yang et al. 2008), polyphenols, phenolic acids, protocatechuic acids and salvianolic acids (Lu et al. 1991; Wu et al. 1999; Qing et al. 2004; Xue et al. 2014). The pharmacological activities of *S. przewalskii* are similar to those of *Salvia miltiorrhiza* Bunge because of the similar chemical constituents of tanshinones and phenolics (Skała and Wysokińska 2005). *S. przewalskii* is used as a substitute for *S. miltiorrhiza*.

In our previous preliminary study on *S. przewalskii* total phenolic acid extract (SPE), we identified five new diterpenoids and one new monoterpenoid glycoside from SPE, finding that

SPE could reduce whole blood viscosity in Wistar rats and increase the urine excretion of water load Wistar rats (Chen et al. 2003; Yang et al. 2011, 2012, 2013, 2017). In addition, we found that SPE could reduce proteinuria and preserve the morphology and structure of podocytes by retaining the level of slit diaphragm proteins in a rat model of puromycin aminonucleoside-induced podocyte injury (Dai et al. 2015).

The aim of this study was to explore the efficacy of SPE more comprehensively in a rat model of immune complex glomerulonephritis (ICG) induced by bovine serum albumin (BSA) and complete Freund's adjuvant (CFA) in an attempt to explain the underlying mechanism, knowing that this animal model is closely relevant to human mesangial proliferative glomerulonephritis (MsPGN) (Fujita et al. 1991; Jia and Zou 1996).

Materials and methods

Plant material

The roots and rhizomes of *S. przewalskii* were collected from Wen County, Gansu Province of China in September 2009 and identified by Prof. ZHANG Hanming from Department of Pharmacognosy, School of Pharmacy, the Second Military

---

CONTACT Ren-Qian Zhong ✉ zhongrq@smmu.edu.cn ▣ Department of Laboratory Diagnostics, Changzheng Hospital, Second Military Medical University, No. 415 Fengyang Rd, Shanghai 200003, China; Wan-Sheng Chen ✉ chenwansheng@smmu.edu.cn ▣ Department of Pharmacy, Changzheng Hospital, Second Military Medical University, No. 415 Fengyang Rd, Shanghai 200003, China

Medical University (SMMU) of CPLA (Shanghai, China). A voucher specimen (Batch No. 200909-b15) was deposited in the herbarium of the said School of Pharmacy.

Preparation of SPE

Fresh roots and rhizomes of *S. przewalskii* were cautiously dried at room temperature of $25 \pm 2\,°C$ for three days, and then the raw medicinal material (5 kg) was cut and grounded to powder mechanically in a mill. The dried powder was macerated and percolated with 50 L 50% ethanol solution for 48 h. The solvent was evaporated at $60\,°C$ under reduced pressure to yield dried ethanol-soluble-extracts (0.6 kg). Then, the ethanol-soluble-extracts were chromatographed over the macroporous adsorptive resin AB-8 (0.8–1.2 mm) (Zhentiancheng Technology Co., Ltd., Tianjin, China) eluting with gradient mixtures of water and ethanol. The eluting solution of 50% ethanol was concentrated and turned to be SPE (90 g) (Yang et al. 2012).

SPE was dissolved in double-distilled water containing 0.8% sodium carboxymethyl cellulose (CMC-Na) and left at room temperature for 20 min to infuse. SPE solution was diluted at 5, 10 and 20 mg/mL with double-distilled water containing 0.8% CMC-Na. Then, 1 mL/kg (body mass) of each solution was administered orally to the rats (equivalent doses at 50, 100 and 200 mg/kg with respect to the mass of SPE). These solutions were freshly prepared just before administration.

HPLC analysis of SPE

The content of two main chemical constituents of SPE, rosmarinic acid and salvianolic acid B, was analysed by high performance liquid chromatography (HPLC). A methanol stock solution of rosmarinic acid and salvianolic acid B was prepared and diluted to proper concentrations to set up the calibration curves. An Agilent 1200 HPLC system (Agilent Technologies, Palo Alto, CA) comprising a quaternary solvent delivery system, an on-line degasser, an autosampler, a column temperature controller and photodiode array detector was used. All data were acquired and analysed in Agilent chemstation software. The chromatographic column was a Dikma Diamonsil RP C_{18} column (5 μm, 250 mm ×4.6 mm). The injection volume was 20 μL and the flow rate was 1.0 mL/min. The column temperature was set and maintained at $30\,°C$ and the detection wavelength was set at 288 nm. The mobile phase consisted of A (acetonitrile) and B (0.05% phosphoric acid aqueous solution). Gradient variations are shown in Table 1.

Reagents and chemicals

Tripterygium glycoside (TG) tablets (Batch No. 040901; Fudan Fuhua Pharmaceutical Co., Ltd., Shanghai, China) were dissolved in 0.8% CMC-Na double-distilled water to get a 1.5 mg/mL solution as the control medicine just before administration. BSA (Batch No. 0409A09; Shenggong Bioengineering Co., Ltd., Shanghai, China) was dissolved in sterilized 0.15 mol/L NaCl

solution to 3 mg/mL. CFA (Batch No. 122K8927) was purchased from Sigma Corporation (St. Louis, MO).

Animals and drug administration

A total of 62 Wistar rats of SPF grade (Slaccas Experimental Animal Co., Ltd., Shanghai, China; Certificate: SCXK2003-0003) weighing $120 \pm 15\,g$ were housed in the experimental animal center of the SMMU (Certificate: SYXK2002-0026). The rats were placed in the animal facility at $25 \pm 2\,°C$, $50 \pm 2\%$ humidity and 12 h light/dark cycle with free access to food and water for at least one week before the experiment. Rats were randomized into six groups: normal control (N) group ($n = 12$); medium (M) group ($n = 10$); TG control medicine group receiving intragastrical administration of 15 mg/kg TG ($n = 10$); and three SPE treatment groups receiving intragastrical administration of low (50 mg/kg), medium (100 mg/kg) and high (200 mg/kg) dose SPE as SPE1, SPE2 and SPE3 groups ($n = 10$ each). Then, the animals were housed individually in metabolic cages to obtain their urine.

All experimental procedures were carried out according to the protocols approved by the Animal Care Committee of the Animal Center at the Chinese Academy of Sciences, and performed in accordance with the recommendations and policies of the SMMU for the protection of animals used for experimental and other scientific purposes.

Experimental protocol and treatment

ICG was induced as previously described (Jia and Zou 1996). Briefly, except for N group, one kidney was resected from each of the animals in the other five groups after 10% chloralization before initiation of the experiment (Cheng et al. 1995). BAS (3 mg) and CFA (0.1 mL) were injected in rats at a foot pad on day 1, and again at the end of the first week. At the end of the second week, the animals received a series of intraperitoneal (i.p.) injections of 0.5, 1, 1.5 and 3 mg BSA at 1 h intervals. During the third week, 2 mg BSA was injected i.p. on day 1, followed by 3 mg BSA on day 3, 5 and 7. Then, 100 μg lipopolysaccharide (LPS) of *Escherichia coli* was injected i.p.

SPE and TG were administered intragastrically (i.g.) at the designated doses in 24 h intervals for 40 consecutive days. Urine protein (UP) and blood biochemical indexes were measured one day before, on day 20 and 40 after drug administration. On day 40 after SPE administration, the animals were sacrificed to obtain the kidneys for the preparation of pathologic sections. In addition, kidney wet mass was measured. The kidney histopathologic sections were observed under a light microscope. UP was quantified by the biuret method. TSP, SA, SC and SUN were detected by routine laboratory tests.

Assessment of kidney function

Measurement of total serum protein (TSP), serum albumin (SA), serum cholesterol (SC) and serum urea nitrogen (SUN) was carried out at three time points, one day before, on day 20 and 40 after drug administration. The kidney specimens were weighed to calculate the ratio of kidney wet mass and rat body mass, which is known as kidney wet mass coefficient (KWMC).

Table 1. Gradient composition of the mobile phase for HPLC analysis of SPE.

Time (min)	A (%)	B (%)
0–20	12 → 20	88 → 80
20–60	20 → 30	80 → 70
60–65	30 → 12	70 → 88

Histopathological examination

Kidney specimens were examined by light microscopy to evaluate the severity and extent of glomerular lesions. Briefly, the renal tissues were fixed in 10% formalin solution, paraffin embedded, sliced to 4 μm sections, stained with haematoxylin–eosin (HE) and periodic acid Schiff, and finally observed under a microscope by a pathologist who was blind to the experimental protocol.

Statistical analysis

All results were expressed as mean ± standard deviation, except for histopathological examinations. Statistical analysis was performed using one-way analysis of variance followed by Fisher's LSD multiple comparison test (SPSS software Version 16.0, SPSS Inc., Chicago, IL). Values of $p < 0.05$ were considered statistically significant.

Results

HPLC analysis of SPE

The retention time of rosmarinic acid and salvianolic acid B was 48.554 min and 37.264 min, respectively. The calibration curves of rosmarinic acid and salvianolic acid B were obtained by the ratio of a series of peaks area (Y) to corresponding concentrations (X, μg/mL), and were used to calculate their content in SPE. The content of rosmarinic acid and salvianolic acid B in SPE was 31.58 and 5.52%, respectively. The results of HPLC analysis are shown in Figure 1.

The effect of SPE on UP excretion

UP excretion in all groups of model animals was significantly higher than that in M group one day before drug administration, indicating that the animal model was established successfully.

On day 20 after administration of TG and SPE, UP excretion in TG group (66.3 ± 77.7 mg/24 h), and three SPE groups (59.4 ± 49.9, 58.0 ± 39.3, 54.3 ± 43.1 mg/24 h) in particular, was significantly lower than that in M group (148.3 ± 132.3 mg/24 h) (all $p < 0.01$). UP excretion kept on descending on day 40 after SPE administration in all three SPE dose groups (54.9 ± 47.8, 54.1 ± 50.7, 50.2 ± 45.5 mg/24 h) in a positive dose-effect relationship, and was significantly lower than that in M group (120.1 ± 82.1 mg/24 h). The differences in the value of UP

excretion between N, M, TG, SPE1, SPE2 and SPE3 groups are shown in Figure 2.

The effect of SPE on KWMC

As shown in Figure 3, there was a significant difference of KWMC in between SPE2 group and M group (6.1 ± 1.0 g/kg versus 7.5 ± 2.1 g/kg; $p < 0.05$) after 40 days treatment. KWMC in SPE1 and SPE3 groups (6.5 ± 1.0 and 6.6 ± 1.8 g/kg) was also lower than that in M group, while the difference was insignificant compared with M group.

The effect of SPE on TSP, SA, SC and SUN

As shown in Figure 4, the levels of TSP, SA, SC and SUN were ameliorated in all three SPE groups.

The level of TSP in SPE2 and SPE3 groups rose significantly after 20 days SPE treatment as compared with that in M group (60.1 ± 2.3 and 61.9 ± 3.2 g/L versus 56.8 ± 2.0 g/L; both $p < 0.01$). Compared with M group, TSP was increased in all three SPE groups after 40 days administration (57.9 ± 2.2, 58.9 ± 1.3 and 58.6 ± 2.3 g/L versus 57.4 ± 2.5 g/L) (Figure 4(a)). The level of SA was increased in all three SPE groups after 20 days treatment as compared with that in M group (40.9 ± 1.2, 40.7 ± 2.4 and 40.6 ± 2.8 g/L versus 38.6 ± 2.1 g/L). This effect remained almost the same after 40 days treatment (39.2 ± 1.5, 40.1 ± 1.5 and 38.8 ± 1.9 g/L versus 37.6 ± 3.2 g/L), and was more pronounced in SPE2 group ($p < 0.01$) (Figure 4(b)).

Both SC and SUN levels were decreased in all three SPE groups after 20 days treatment, and this effect was more pronounced in SPE1 and SPE3 groups as compared with that in M group (1.62 ± 0.5, 2.00 ± 0.4 mmol/L versus 2.56 ± 0.6 mmol/L for SC; 7.61 ± 0.8, 7.98 ± 0.9 mmol/L versus 9.31 ± 1.6 mmol/L for SUN; all $p < 0.05$). Compared with M group, both SC and SUN levels continued to decrease in the three SPE groups after 40 days treatment (2.24 ± 0.3, 2.05 ± 0.50 and 1.94 ± 0.5 mmol/L versus 2.66 ± 0.9 mmol/L for SC; 8.52 ± 0.8, 8.05 ± 1.1 and 7.94 ± 0.6 mmol/L versus 9.88 ± 2.4 mmol/L for SUN), especially in SPE2 and SPE3 groups (both $p < 0.01$) (Figure 4(c,d)).

Histopathological findings

Light microscopic examination of the renal tissues showed severe glomerular engorgement, apomorphosis of the renal tubular epithelium, interstitial inflammatory infiltration and hypercellularity
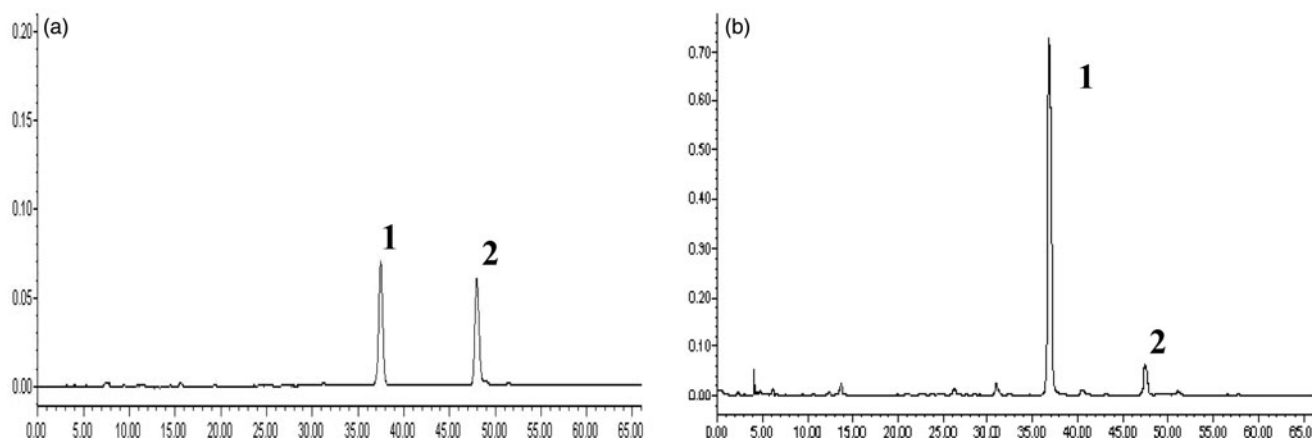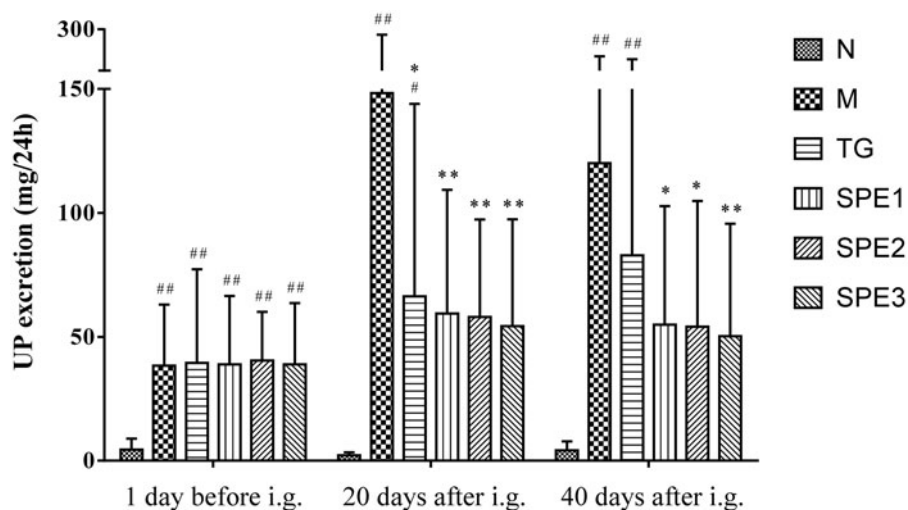

Figure 1. HPLC plots of standard solution (a) and SPE (b). The peaks are identified as: rosmarinic acid (1), salvianolic acid B (2).

Figure 2. The effect of SPE administration on UP excretion. N: normal control group; M: model group; TG: control medicine group (15 mg/kg TG i.g.); SPE1: low dose treatment group (50 mg/kg SPE i.g.); SPE2: medium dose treatment group (100 mg/kg SPE i.g.); SPE3: high dose treatment group (200 mg/kg SPE i.g.). #$p < 0.05$, ##$p < 0.01$, compared with N group; *$p < 0.05$, **$p < 0.01$, compared with M group.
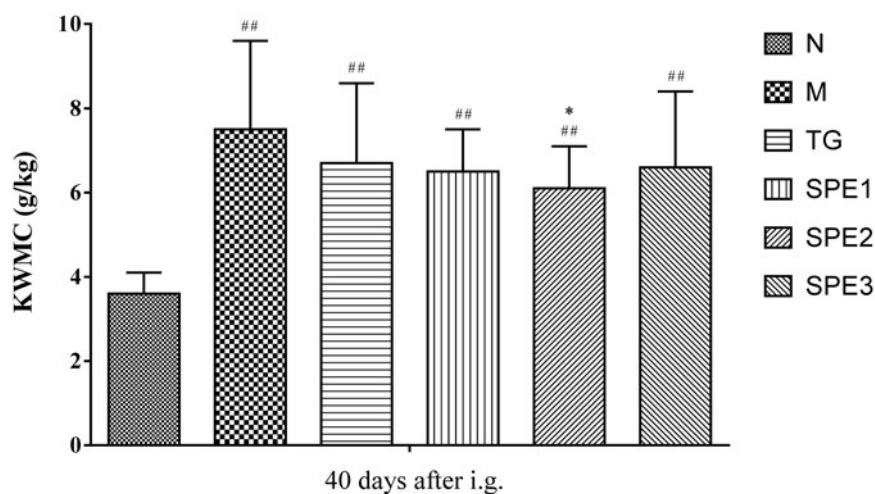
Figure 3. The effect of SPE administration on KWMC. N: normal control group; M: model group; TG: control medicine group (15 mg/kg TG i.g.); SPE1: low dose treatment group (50 mg/kg SPE i.g.); SPE2: medium dose treatment group (100 mg/kg SPE i.g.); SPE3: high dose treatment group (200 mg/kg SPE i.g.). ##$p < 0.01$, compared with N group; *$p < 0.05$, compared with M group.

in all groups except N group. Severe glomerular fibrosis, necrosis and calcification of the renal tubules were observed in M group. Compared with M group, glomerular engorgement and apomorphosis of the renal tubular epithelium were ameliorated significantly in TG, SPE1, SPE2 and SPE3 groups after 40 days treatment. In addition, renal inflammatory infiltration and intumesce were attenuated in all three SPE groups, which were consistent with effect of SPE on KWMC. The changes are shown in Figure 5.

Discussion

It is common knowledge that human nephritis is mainly due to glomerular deposition and/or immune complex formation (Kasap et al. 2008). These immune complexes, which act as a trigger to deliver cytokines and locally yield more transforming growth factor-β (TGF-β) and platelet-derived growth factor, lead to mesangial cell proliferation and glomerulosclerosis (Isaka et al. 1993; Gómez-Guerrero et al. 2000). MsPGN is one of the most common pathological types of chronic glomerulonephritis in Chinese

populations (Li 1989; Cheng et al. 2009). It is defined as a glomerulonephritis with an essentially uniform increase in mesangial cells in all or nearly all glomeruli, of which the specific biopsy findings are diffuse or present focal increase in glomerular mesangial cells and matrix with or without mesangial deposits of immunoglobulins and/or complement (Kasap et al. 2008). Generally, deposition of immune complexes in the mesangial area stimulates mesangial cell proliferation and leads to MsPGN.

Proteinuria is the most common clinical manifestation of kidney disease and the most marked signal of renal damage (Dai et al. 2015). Urine protein excretion level shows an intimate connection with the renal function. Most MsPGN cases are associated with persistent proteinuria or with nephrotic syndrome (Danilewicz and Wągrowska-Danilewicz 2001). Nephrotic syndrome is characterized by proteinuria, hypercholesterolaemia, hypoalbuminaemia and oedema (Maxie and Newman 2007). So, the reduction of proteinuria and the amelioration of blood biochemical indexes are two main targets in assessing the therapeutic efficacy of traditional Chinese medicines.

Tripterygium wilfordii Hook. f. (Celastraceae) is a shrub plant. TG is an extract from this plant and has been utilized for 30 years
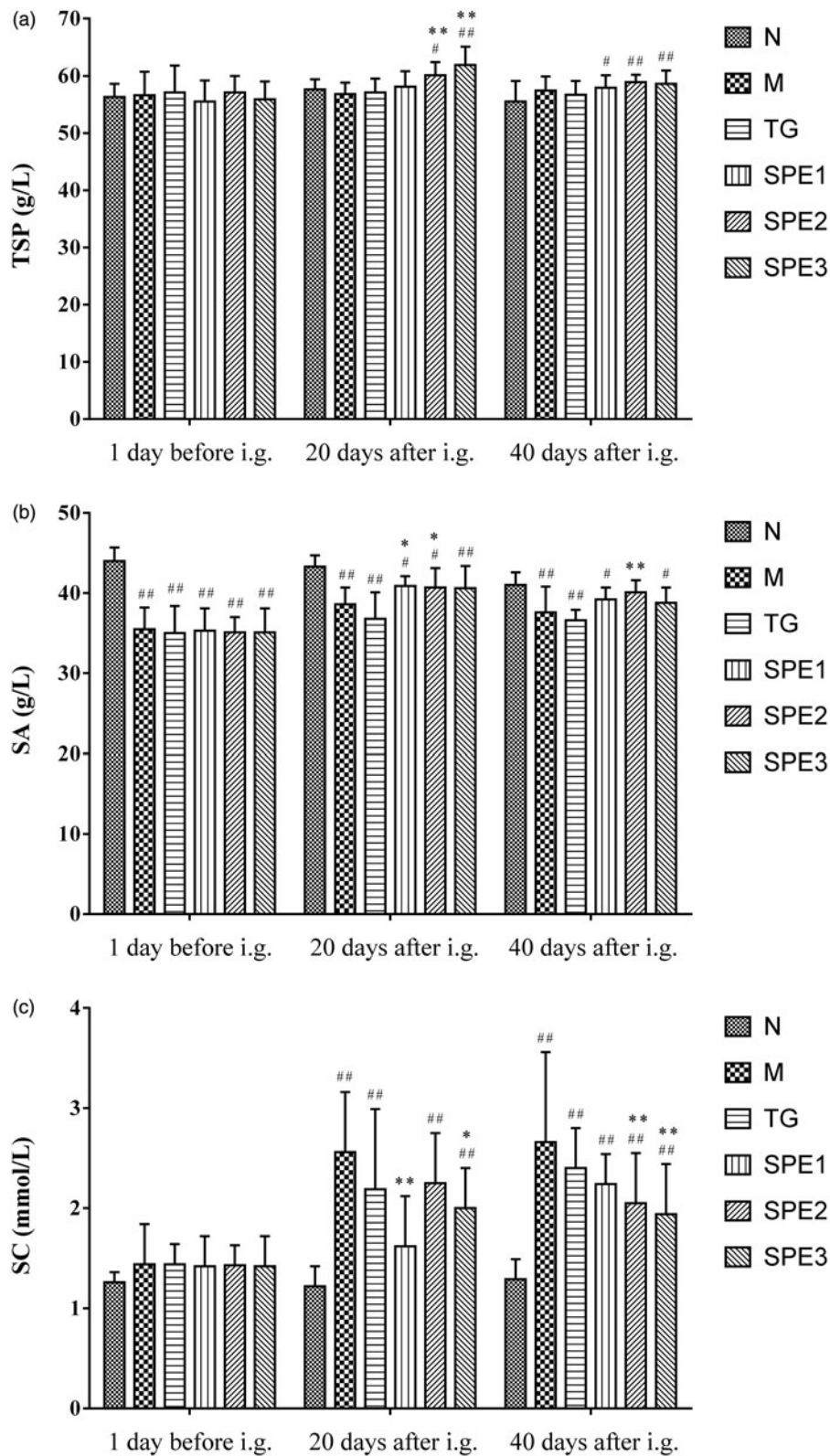
Figure 4. The effect of SPE administration on TSP (a), SA (b), SC (c) and SUN (d). N: normal control group; M: model group; TG: control medicine group (15 mg/kg TG i.g.); SPE1: low dose treatment group (50 mg/kg SPE i.g.); SPE2: medium dose treatment group (100 mg/kg SPE i.g.); SPE3: high dose treatment group (200 mg/kg SPE i.g.). #$p < 0.05$, ##$p < 0.01$, compared with N group; *$p < 0.05$, **$p < 0.01$, compared with M group.

in the treatment of various immune and inflammatory diseases in China, especially rheumatoid arthritis, glomerulonephritis and proliferative glomerulonephritis (Wan et al. 2011). Recent studies have demonstrated the remarkable therapeutic efficacy of TG on proteinuria. But as an immunosuppressive agent, the toxicity and adverse effects of TG are also obvious (Li and Liu 2003; Wan et al. 2011), which limit the more extensive use of TG in clinical practice.

In the present study, we demonstrated the therapeutic efficacy of SPE *in vivo* in improving urinary protein excretion in a rat
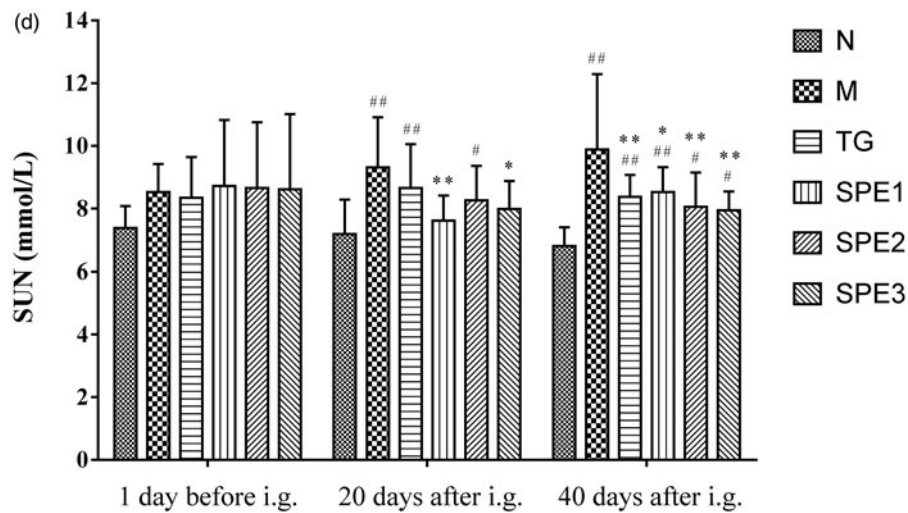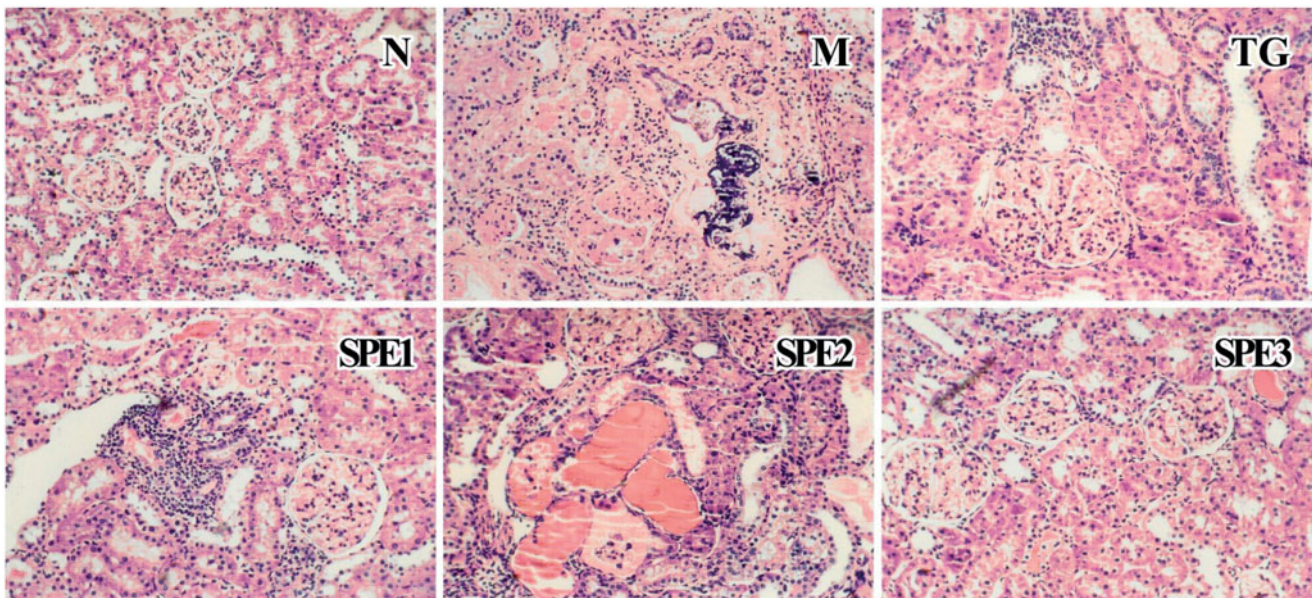
Figure 4. Continued.

Figure 5. Light micrographic changes of the kidney specimens (stained with HE, 10×20). N: normal control group; M: model group; TG: control medicine group (15 mg/kg TG i.g.); SPE1: low dose treatment group (50 mg/kg SPE i.g.); SPE2: medium dose treatment group (100 mg/kg SPE i.g.); SPE3: high dose treatment group (200 mg/kg SPE i.g.).

ICG model, knowing that this nominal model mimics human MsPGN. The anti-proteinuria effect of SPE in rats was observed on day 20 after SPE administration. The efficacy of SPE in decreasing proteinuria was even better than that of TG, especially after 40 days treatment. More importantly, increased TSP and SA values together with decreased SC and SUN values showed that SPE could maintain liver function and renal function effectively. Inflammatory response is the characteristic feature of glomerular disease in immune glomerular injury due to the infiltration of leukocytes and the proliferation of glomerular cells. SPE treatment reduced the KWMC and rectified the histological abnormalities of the kidney. Observably, SPE inhibited inflammatory infiltration of glomerular cells and dropsy of the kidney as shown on light microscopy. Oedema could be treated with diuretic agents such as hydrochlorothiazide in some types of glomerulonephritis (Beitollahi et al. 2015). In addition, as a diuretic agent,

SPE is also a possible contributor to reduce oedema and adjust renal function.

The major active components of SPE are rosmarinic acid (content 31.58%) and salvianolic acid B (content 5.52%). Rosmarinic acid, an ester of caffeic acid and 3,4-dihydroxyphenyl lactic acid, is an active polyphenolic phytoconstituent. It has strong anti-oxidative (Lamien-Meda et al. 2010), anti-inflammatory (Parnham and Kesselring 1985; Jiang et al. 2009), anti-allergic (Lee et al. 2008), anti-ultraviolet and anti-radiation activities (Sánchez-Campillo et al. 2009). *In vitro* studies showed that rosmarinic acid inhibited cytokine-induced murine mesangial cell proliferation (Makino et al. 2000). *In vivo* studies showed that oral administration of rosmarinic acid suppressed the proliferation of mesangial cells and glomerular matrix expansion by its antifibrotic effect and anti-oxidative activity on rat MsPGN established by intravenous injection of rabbit anti-rat thymocyte

serum to rats (Makino et al. 2002). Salvianolic acid B is a tetramer of caffeic acid, as well as a potent inhibitor of TGF-β1-induced epithelial-mesenchymal transition for the treatment of tubulointerstitial fibrosis (Yao et al. 2009). It showed a strong protective effect on rats with induced diabetic nephropathy (Lee et al. 2003; Kang et al. 2008).

Given the water-soluble polyphenolic compounds in SPE, its efficacy on ICG is closely related to the pharmacological action of rosmarinic acid and salvianolic acid B. The possible pathways are thought to depend on two mechanisms. One is possibly the anti-complementary pathway, knowing that rosmarinic acid possesses the activity to reduce immunohaemolysis by inhibiting the C3-convertase of the classical complement pathway (Englberger et al. 1988). As compounds with polyhydroxylated phenyl rings are highly reactive with the thioester bond in nascent C3b, rosmarinic acid and salvianolic acid B block complement activation by preventing attachment of C3b to the activating surface (Sahu et al. 1999). Improved serum complement level and reduced proteinuria were reported to have a beneficial effect on hypocomplementaemia by inhibiting complement activation in patients with increased serum C3 levels (Fujita et al. 1993). The other possible pathway is the anti-inflammatory and antioxidative pathway. Rosmarinic acid has the inhibitory effect against inflammatory response and the scavenging effect against reactive oxygen radicals (Sánchez-Campillo et al. 2009). It owns an inhibitory activity on the formation of malondialdehyde in human platelets (Gracza et al. 1985). It was reported that malondialdehyde, a kind of lipid peroxidation product, was eliminated by means of suppressing markedly the formation of hydroxyl free radicals and lipid peroxidation reaction by *S. przewalskii* (Cheng and Yang 2003). So, the therapeutic efficacy of SPE may be achieved by the above-mentioned pathways. Furthermore, rosmarinic acid showed very low toxicity with a LD_{50} in mice of 561 mg/kg after intravenous administration (Petersen and Simmonds 2003). So, the safety of utilizing SPE to treat MsPGN is reliable.

Conclusions

In summary, our present study demonstrated that SPE could reduce proteinuria, regulate protein and lipid metabolisms, attenuate renal inflammatory cell infiltration, and delay the progression of glomerular lesions in a rat ICG model. The results of the present research provide evidences that SPE has a potentiality to become a therapeutic drug for glomerulonephritis. To confirm our conclusion, more studies are required to improve quality control of SPE and clarify its action mechanism against glomerulonephritis for clinical application.

Disclosure statement

The authors declare that they have no conflicts of interest.

Funding

References

Beitollahi H, Hamzavi M, Torkzadeh-Mahani M. 2015. Electrochemical determination of hydrochlorothiazide and folic acid in real samples using a modified graphene oxide sheet paste electrode. Mater Sci Eng C Mater Biol Appl. 52:297–305.

Chen WS, Tao ZY, Zhang WD, Sun LN. 2003. A new compound from the root of *Salvia przewalskii* Maxim. Chin Chem Lett. 14:711–712.

Cheng QL, Orikasa M, Morioka T, Kawachi H, Chen XM, Oite T, Shimizu F. 1995. Progressive renal lesions induced by administration of monoclonal antibody 1-22-3 to unilaterally nephrectomized rats. Clin Exp Immunol. 102:181–185.

Cheng TJ, Yang ZY. 2003. Comparison between *S. przewalskii* Maxim injection and *S. miltiorrhiza* Bunge injection on the protection on acute focal cerebral ischemia of rats and the effect against lipid peroxidation. Chin J Clin Pharmacol Ther. 8:23–26.

Cheng Y, Zhang JB, Hou WP, Wang DH, Li FR, Zhang YQ, Yuan FH. 2009. Immunoregulatory effects of sinomenine on the T-bet/GATA-3 ratio and Th1/Th2 cytokine balance in the treatment of mesangial proliferative nephritis. Int Immunopharmacol. 9:894–899.

Dai DS, Liu X, Yang Y, Luo XM, Tang RX, Yin ZC, Ren HQ. 2015. Protective effect of *Salvia przewalskii* extract on puromycin-induced podocyte injury. Am J Nephrol. 42:216–227.

Danilewicz M, Wągrowska-Danilewicz M. 2001. Morphometric comparison of the quantity of the mesangial deposits in rebiopsied patients with idiopathic mesangial proliferative glomerulonephritis. Pathol Res Pract. 197:545–550.

Englberger W, Hadding U, Etschenberg E, Graf E, Leyck S, Winkelmann J, Parnham MJ. 1988. Rosmarinic acid: a new inhibitor of complement C3-convertase with anti-inflammatory activity. Int J Immunopharmacol. 10:729–737.

Fujita M, Iida H, Asaka M, Izumino K, Takata M, Sasayama S. 1991. Effect of the immunosuppressive agent, ciclosporin, on experimental immune complex glomerulonephritis in rats. Nephron. 57:201–205.

Fujita Y, Inoue I, Inagi R, Miyata T, Shinzato T, Sugiyama S, Miyama A, Maeda K. 1993. Inhibitory effect of FUT-175 on complement activation and its application for glomerulonephritis with hypocomplementemia. Nihon Jinzo Gakkai Shi. 35:393–397.

Gómez-Guerrero C, Duque N, Casado MT, Pastor C, Blanco J, Mampaso F, Vivanco F, Egido J. 2000. Administration of IgG Fc fragments prevents glomerular injury in experimental immune complex nephritis. J Immunol. 164:2092–2101.

Gracza L, Koch H, Löffler E. 1985. Über biochemisch-pharmakologische untersuchungen pflanzlicher arzneistoffe, 1. mitt. isolierung von rosmarinsäure aus *Symphytum officinale* und ihre anti-inflammatorische wirksamkeit in einem in-vitro-modell. Arch Pharm Pharm Med Chem. 318:1090–1095.

Huang YQ, Sun XC. 1977. Salvia Linn. In: Wu ZY, Li XW, editors. *Flora Reipublicae Popularis Sinicae*, vol. 66. Beijing: Science Press; p. 70–79, 86–89.

Isaka Y, Fujiwara Y, Ueda N, Kaneda Y, Kamada T, Imai E. 1993. Glomerulosclerosis induced by *in vivo* transfection of transforming growth factor-beta or platelet-derived growth factor gene into the rat kidney. J Clin Invest. 92:2597–2601.

Jia H, Zou WZ. 1996. An improved animal model for mesangioproliferative glomerulonephritis in rat. Chin J Nephrol Dialy Transplant. 5:21–25.

Jiang HL, Wang XZ, Xiao J, Luo XH, Yao XJ, Zhao YY, Chen YJ, Crews P, Wu QX. 2013. New abietane diterpenoids from the roots of *Salvia przewalskii*. Tetrahedron. 69:6687–6692.

Jiang WL, Chen XG, Qu GW, Yue XD, Zhu HB, Tian JW, Fu FH. 2009. Rosmarinic acid protects against experimental sepsis by inhibiting proinflammatory factor release and ameliorating hemodynamics. Shock. 32:608–613.

Kang ES, Lee GT, Kim BS, Kim CH, Seo GH, Han SJ, Hur KY, Ahn CW, Ha H, Jung M, et al. 2008. Lithospermic acid B ameliorates the development of diabetic nephropathy in OLETF rats. Eur J Pharmacol. 579:418–425.

Kasap B, Türkmen M, Sarıoğlu S, Sis B, Soylu A, Kavukçu S. 2008. The relation of IgM deposition to clinical parameters and histomorphometry in childhood mesangial proliferative glomerulonephritis. Pathol Res Pract. 204:149–153.

Lamien-Meda A, Nell M, Lohwasser U, Börner A, Franz C, Novak J. 2010. Investigation of antioxidant and rosmarinic acid variation in the sage collection of the genebank in Gatersleben. J Agric Food Chem. 58:3813–3819.

Lee GT, Ha H, Jung M, Li H, Hong SW, Cha BS, Lee HC, Cho YD. 2003. Delayed treatment with lithospermate B attenuates experimental diabetic renal injury. J Am Soc Nephrol. 14:709–720.

Lee J, Jung E, Koh J, Kim YS, Park D. 2008. Effect of rosmarinic acid on atopic dermatitis. J Dermatol. 35:768–771.

Li LS, Liu ZH. 2003. Experiences in nephritis treated by *Tripterygium wilfordii* Hook. f. in the 25 years. Chin J Nephrol Dialy Transplant. 12:246–247.

Li LS. 1989. An overview of nephrology in China. Chin Med J. 102:488–495.

Li MH, Chen JM, Peng Y, Xiao PG. 2008. Distribution of phenolic acids in Chinese *Salvia* plants. World Sci Technol – Mod Tradit Chin Med Mater Med. 10:46–52.

Liu JM, Nan P, Tsering Q, Tsering T, Bai ZK, Wang L, Liu ZJ, Zhong Y. 2006. Volatile constituents of the leaves and flowers of *Salvia przewalskii* Maxim. from Tibet. Flavour Frag J. 21:435–438.

Lu XZ, Xu WH, Shen JX, Naoki H. 1991. Przewalskinic acid A, a new phenolic acid from *Salvia przewalskii* Maxim. Chin Chem Lett. 2:301–302.

Makino T, Ono T, Liu N, Nakamura T, Muso E, Honda G. 2002. Suppressive effects of rosmarinic acid on mesangioproliferative glomerulonephritis in rats. Nephron. 92:898–904.

Makino T, Ono T, Muso E, Yoshida H, Honda G, Sasayama S. 2000. Inhibitory effects of rosmarinic acid on the proliferation of cultured murine mesangial cells. Nephrol Dial Transplant. 15:1140–1145.

Maxie MG, Newman SJ. 2007. Urinary system. In: Maxie MG, editor. Jubb, Kennedy, and Palmer's pathology of domestic animals, vol. 2, 5th ed. New York, Edinburgh: Elsevier Saunders; p. 451–466.

Ohsaki A, Kawamata S, Ozawa M, Kishida A, Gong X, Kuroda C. 2011. Salviskinone A, a diterpene with a new skeleton from *Salvia przewalskii*. Tetrahedron Lett. 52:1375–1377.

Parnham MJ, Kesselring K. 1985. Rosmarinic acid. Drugs Fut. 10:756–757.

Petersen M, Simmonds MSJ. 2003. Rosmarinic acid. Phytochemistry. 62:121–125.

Qing DH, Chen HS, Peng ZG, Guo ZM. 2004. A new compound from *Salvia przewalskii* and its anti-HIV effect. Chin Tradit Herb Drugs. 35:725–728.

Sahu A, Rawal N, Pangburn MK. 1999. Inhibition of complement by covalent attachment of rosmarinic acid to activated C3b. Biochem Pharmacol. 57:1439–1446.

Sánchez-Campillo M, Gabaldon JA, Castillo J, Benavente-García O, Del Baño MJ, Alcaraz M, Vicente V, Alvarez N, Lozano JA. 2009. Rosmarinic acid, a photo-protective agent against UV and other ionizing radiations. Food Chem Toxicol. 47:386–392.

Skała E, Wysokińska H. 2005. Tanshinone production in roots of micropropagated *Salvia przewalskii* Maxim. Z Naturforsch C. 60:583–586.

Wan YG, Sun W, Zhen YJ, Che XY, Pu HP, Wang Y, Li M, Ruan JG, Yan QJ. 2011. Multi-glycoside of *Tripterygium wilfordii* Hook. f. reduces proteinuria through improving podocyte slit diaphragm dysfunction in anti-Thy1.1 glomerulonephritis. J Ethnopharmacol. 136:322–333.

Wang HQ, Yang LX, Chen XY, Yang PF, Chen RY. 2015. Chemical constituents from *Salvia przewalskii* root. J Chin Med Mater. 38:1197–1201.

Wang N, Niwa M, Luo HW. 1988. Triterpenoids from *Salvia przewalskii*. Phytochemistry. 27:299–301.

Wu ZJ, Ouyang MA, Yang CR. 1999. Polyphenolic constituents of Salvia przewalskii. Acta Bot Yunnanica. 21:512–516.

Xue YB, Wu Y, Zhu HC, Li XN, Qian JF, Lai YJ, Chen CM, Yao GM, Luo ZW, Li Y, et al. 2014. Salviprzols A and B, C_{21}- and C_{22}-terpenoids from the roots of *Salvia przewalskii* Maxim. Fitoterapia. 99:204–210.

Yang Y, Bing Z, Sun LN, Wu ZJ, Chen WS. 2013. Chemical constituents of *Salvia przewalskii* Maxim. Asian J Chem. 25:1747–1748.

Yang Y, Lu WQ, Wu ZJ, Chen WS. 2017. A new diterpenoid from Salvia przewalskii. Rec Nat Prod. 11:416–420.

Yang Y, Wu ZJ, Yang YB, Lai W, Sun LN, Chen WS. 2011. Three new terpenoids from *Salvia przewalskii* Maxim. Chem J Chin Univ. 32:1318–1322.

Yang Y, Zhang F, Cai F, Sun LN, Chen WS. 2008. Advances in studies on chemical constituents and pharmacological effects of *Salvia przewalskii* Maxim. J Chin Med Mater. 31:787–790.

Yang Y, Zhu B, Wu ZJ, Chen WS, Sun LN. 2012. Effects of *Salvia przewalskii* Maxim. extract on whole blood viscosity in normal rats and its diuresis in water-loaded rat. Chin J Hosp Pharm. 32:751–754.

Yao G, Xu LZ, Wu XC, Xu LL, Yang JW, Chen HM. 2009. Preventive effects of salvianolic acid B on transforming growth factor-beta1-induced epithelial-to-mesenchymal transition of human kidney cells. Biol Pharm Bull. 32:882–886.