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Anxiolytic effect of saponins from *Panax quinquefolium* in mice

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Abstract

The anxiolytic effect of the saponins from *Aniliaeea Panax quinquefolium* L. (PQS) was studied in male mice by using a number of experimental paradigms of anxiety and compared with that of the known anxiolytic compound diazepam. Use of the elevated plus-maze test revealed that PQS (50 mg/kg, p.o.) and diazepam (2.5 mg/kg, p.o.) increased the percentage of time and entries spent in open arms. In the light/dark test, PQS (50 and 100 mg/kg, p.o.) and diazepam (2.5 mg/kg, p.o.) prolonged the time spent in the light area. In the hole-board test, PQS (50 and 100 mg/kg, p.o.) and diazepam (2.5 mg/kg, p.o.) significantly increased both head-dip counts and head-dip duration. Both PQS (50 and 100 mg/kg, p.o.) and diazepam (2.5 mg/kg, p.o.) decreased the total fighting time in the isolation-induced aggressive test. Since PQS, in contrast to diazepam, had no effect on locomotion in these tests, its side-effect profile might be considered superior to the benzodiazepines. Thus, the present findings suggest that PQS might be a potential candidate for use as an anxiolytic drug.

Keywords: Anxiolytic; PQS; Elevated plus-maze test; Hole-board test; Light-dark test; Isolation-induced aggressive test

1. Introduction

Ginseng (*Panax ginseng* Meyer) has been used as one of the most valuable natural medicines in China for more than 2000 years. American ginseng (*Panax quinquefolium* L.), a plant native to North America, is now also cultivated and used in many countries. It belongs to the *Panax* genus of the Araliaceae. It has been shown that ginseng administration produces a variety of effects on the central nervous system. For example, ginseng causes behavioral changes in animals, and these changes appear to be related to the regulation of GABAergic transmission (Kimura et al., 1994). Chronic intake of ginseng stabilizes sleep and wakefulness in food-deprived rats (Lee et al., 1990) and the effects of ginseng extract on learning, memory, and physical capacities have also been reported (Petkov and Mosharov, 1987).

It has been shown that *Panax quinquefolium* exerts many beneficial effects similar to ginseng. For example, *Panax quinquefolium* has been widely used in folk medicine for its antioxidant, antilipid peroxidation, antihypoxia and antifatigue properties (Persons, 1986; Fu and Ji, 2003). In animal experiments, modulatory effects of *Panax quinquefolium* on the central nervous system have been observed by Bensky and Gamble (1993). An extract from the leaves and stems of *Panax quinquefolium* has recently been shown to have an anticonvulsant effect in several animal models of seizures (Lian et al., 2005). These effects are thought to be attribute to the major active ingredients, saponins, in *Panax quinquefolium* (PQS). Over the last decade, researchers have found that PQS can exert beneficial effects on the cardiovascular system via its antiischemic, antiarrhythmic, antihypertensive and antioxidative actions (Lu and Sui, 1996). Previous studies have demonstrated that PQS increases the plasma high-density lipoprotein content and decreases the lipid peroxide levels in hyperlipidemic rats (Li et al., 1993). In addition, PQS protects cultured rat cardiac myocytes from oxidative damage (Yang et al., 1992). It has been reported that PQS has a beneficial effect on stress-induced pathophysiological changes in the central nervous system and it has recently been reported that PQS can improve memory impaired by scopolamine, cycloheximide and sodium pentobarbital in the passive avoidance test (Gao et al., 1995). Ginsenoside Rb1 and pseudoginsenoside-F11, components of PQS, can prevent the memory deficits induced by scopolamine in rats (Benishin et al., 1991; Benishin, 1992; Li et al., 1999). These results

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strongly suggest that PQS might be a potential neuroactive principle.

According to the pharmacological profile of PQS, it is reasonable to assume that PQS might have some other neuroactive activities. Therefore, the present study was designed to investigate the anxiolytic effects of PQS by using several anxiety paradigms: the elevated plus-maze test, light/dark test, hole-board test and isolation-induced aggressive test.

2. Materials and methods

2.1. Plant material

The roots of *Panax quinquefolium* L. were collected from the Jilin province of China, and identified by Prof. Qi-Shi Sun (Shenyang Pharmaceutical University).

2.2. Preparation of extracts

The procedures for extraction and isolation of the saponins from the roots of *Panax quinquefolium* L. were as follow. Briefly, the dried roots of the plant (5000 g) were powdered and then extracted with 70% EtOH (50 L) three times (2 h for each time) under reflux. After filtration, excess solvent was removed under reduced pressure. The EtOH extract was suspended in water and defatted with ether followed by partitioning with *n*-BuOH. The combined *n*-BuOH layers were concentrated to dryness. The dried extract was subjected to HPD100 resin column chromatography, washed with water, and eluted with EtOH to afford a total saponins fraction (1645 g).

2.3. Animals

Male Swiss mice (Experimental Animal Center of Shenyang Pharmaceutical University) weighing 18–20 g were used. The mice were housed in groups of five in cages of 28 cm × 20 cm × 16 cm. Food and water were freely available and the room temperature was controlled at 22 ± 2 °C. Each animal was used once in the behavior tests. All animal use procedures were in accordance with the Regulations of Experimental Animal Administration issued by State Committee of Science and Technology of the People's Republic of China on 14 November 1988.

2.4. Drugs

PQS was purified by the Department of Chemistry for Natural Drugs of Shenyang Pharmaceutical University (purity > 95%) and dissolved in distilled water. Diazepam (DZ) was used as a positive control and purchased from Hubei Pharmaceutical Factory (Hubei, China). Diazepam was ultrasonically dispersed in distilled water. Test drugs were orally administered 30 min before the experiments in a volume of 10 mL/kg. Blank control animals were given the corresponding vehicles. All drugs were freshly prepared before each experiment.

2.5. Procedures

2.5.1. Elevated plus-maze test

The elevated plus-maze comprised two open (30 cm × 5 cm × 0.25 cm) and two enclosed (30 cm × 5 cm × 15 cm) arms that radiated from a central platform (5 cm × 5 cm) to form a plus sign. The maze was constructed of black painted wood. A slight raised edge on the open arms (0.25 cm) provided additional grip for the animals. The plus-maze was elevated to a height of 40 cm above floor level by a single central support. Four 25 W red fluorescent lights arranged as a cross at 100 cm above the maze were used as the source of illumination (Chen et al., 2003). The experiment was conducted during the dark phase of the light cycle (9:00–14:00 h). The trial was started by placing an animal on the central platform of the maze facing an open arm. The number of entries into, and the time spent in, each of the two types of arm, were counted during a 5 min test period. The percentage open arm entries and percentage open arm time were used as indices of anxiety. A mouse was considered to have entered an arm when all four paws were on the arm. The apparatus was cleaned thoroughly between trials with damp and dry towels. All behavioral recordings were carried out with the observer unaware of the treatment the mice had received.

2.5.2. Light/dark test

The apparatus consisted of two 20 cm × 10 cm × 14 cm plastic boxes: one was dark and the other was transparent. The mice were allowed to move from one box to the other through an open door between the two boxes. A 100 W bulb placed 30 cm above the floor of the transparent box was the only light source in the room. A mouse was put into the light box facing the hole. The transitions between the light and the dark box and time spent in the light box were recorded for 5 min immediately after the mouse stepped into the dark box (Lepicard et al., 2000; Guo et al., 2004). The apparatus was cleaned thoroughly between trials. All behavioral recordings were carried out with the observer unaware of the treatment the mice had received.

2.5.3. The hole-board test

The apparatus was composed of a gray wooden box (50 cm × 50 cm × 50 cm) with four equidistant holes 3 cm in diameter in the floor (Moraira et al., 2000). The centre of each hole was 10 cm from the nearest wall of the box. The floor of the box was positioned 15 cm above the ground and divided into squares of 10 cm × 10 cm with a water-resistant marker. An animal was placed in the center of the hole-board and allowed to freely explore the apparatus for 5 min. The total locomotor activity (numbers of squares crossed), and the number and duration of head-dippings were recorded. A head dip was scored if both eyes disappeared into the hole.

2.5.4. Isolation-induced aggressive test

Isolated mice were prepared as described by Guo et al. (2004). Each mouse was isolated in cages of 28 cm × 20 cm × 16 cm for 6 weeks. Isolated mice were prescreened for aggressive behavior prior to the experiment. An intruder mouse was introduced

Table 1
Effects of PQS in the elevated plus-maze test in mice

Drug	Dose (mg/kg)	Open arm entries	Close arm entries	Total arm entries
Veh	–	4.10 ± 1.12	18.80 ± 1.72	32.90 ± 3.53
PQS	25	6.90 ± 1.59	17.20 ± 1.30	24.10 ± 1.79
	50	8.80 ± 0.71	18.70 ± 1.64	27.50 ± 1.85
	100	8.90 ± 2.29	18.30 ± 1.34	27.00 ± 1.25
DZ	2.5	13.00 ± 1.32**	19.70 ± 2.88	22.90 ± 2.27*

Values represent means ± S.E.M. ($n = 10$). *$P < 0.05$, **$P < 0.01$ compared with vehicle.

Table 2
Effects of PQS on exploratory behavior in mice on the hole-board test

Drug	Dose	Head-dip counts	Head-dip duration (s)	locomotion
Veh	–	8.20 ± 1.05	13.50 ± 1.29	64.50 ± 6.66
PQS	25	11.0 ± 0.79	14.20 ± 1.27	58.00 ± 7.10
	50	17.40 ± 1.80**	18.10 ± 2.15	57.60 ± 5.89
	100	18.20 ± 1.70**	21.20 ± 1.91*	64.50 ± 4.53
DZ	2.5	17.10 ± 1.29**	20.30 ± 2.32*	46.40 ± 6.39

Results are expressed as means ± S.E.M. ($n = 12$). *$P < 0.05$, **$P < 0.01$, compared with vehicle.

into the isolated mouse's cage for 3 min, and the isolated mice exhibiting bite marks for more than 20 s were used for the test experiments on the following day. Two isolated mice that were pretreated with drugs were placed in a neutral cage, which was the same size as their home cages as previously reported. An assessment of the aggressive behavior (biting attacks, wrestling, lateral threats and tail switching) of two isolated mice was conducted for 5 min as the attack duration as previously reported (Abramov et al., 2004) after minor modification.

2.6. Statistical analysis

All analyses were performed using SPSS V11.5 software for Windows. All the data were given as means ± S.E.M. Data were analyzed by one-way ANOVA. Whenever ANOVA was significant, further comparisons between vehicle- and drug-treatment groups were performed using the Dunnett's *t*-test. The level of statistical significance adopted was $P < 0.05$.

3. Results

3.1. Elevated plus-maze test

The results are shown in Fig. 1 and Table 1. Both PQS (50 mg/kg) and DZ (2.5 mg/kg) resulted in a significant increase in the percentage of time and entries into open arms ($P < 0.05$ and $P < 0.01$). However, some differences were observed between the two drugs, e.g., DZ increased open arm entries and total arm entries at the same time while PQS had no such effect.

3.2. Light/dark test

The results of the light/dark test are shown in Fig. 2. PQS at the doses of 50 and 100 mg/kg and DZ (2.5 mg/kg) induced a

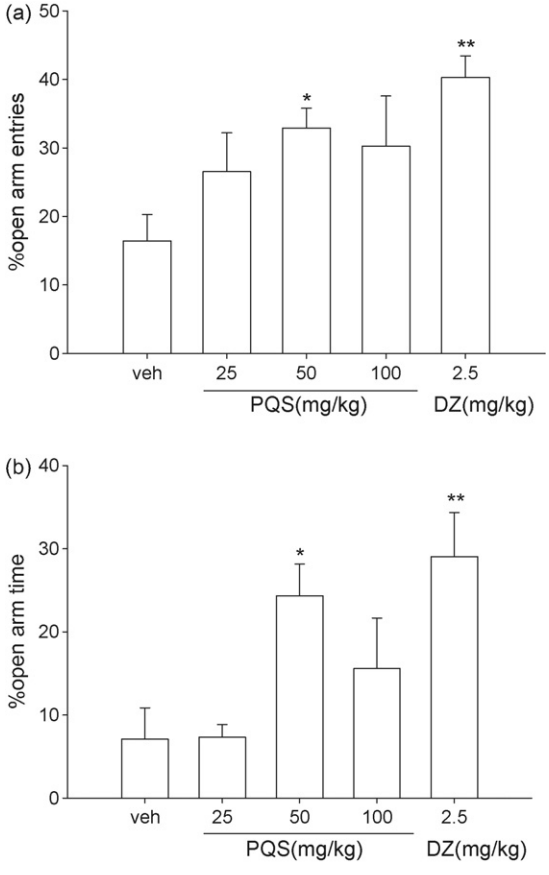

Fig. 1. Effects of PQS in the elevated plus-maze test in male mice. Results are expressed as means ± S.E.M. ($n = 10$). The following parameters are shown: %open arm entries [percentage of entries into open arms with respect to total entries into the arms (a)]; %open arm time [percentage of time spent in open arms with respect to total time spent in the arms (b)]. *$P < 0.05$, **$P < 0.01$, compared with vehicle-treated animals.

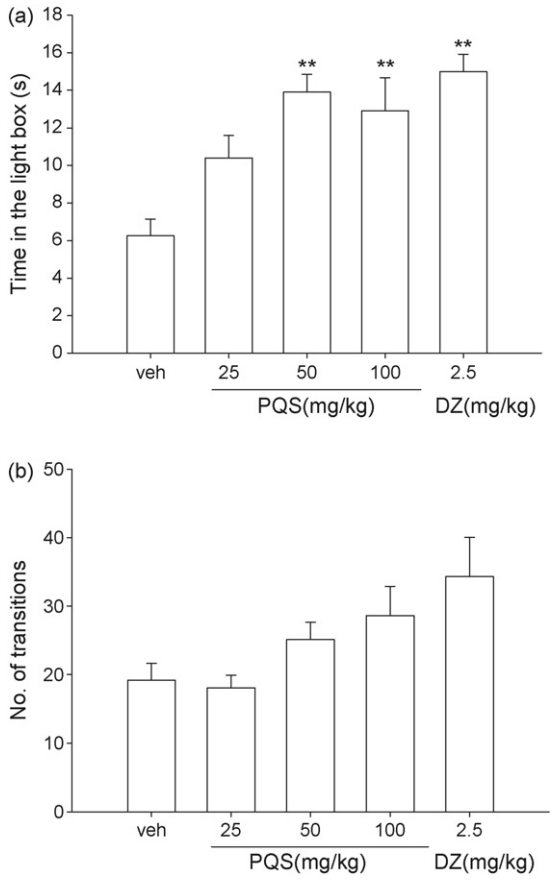

Fig. 2. Effects of PQS in the light/dark test in male mice. (a) Time in the light box (s); (b) No. of transitions. Results are expressed as means ± S.E.M. ($n = 10$). $^{**}P < 0.01$, compared with vehicle-treated animals.

significant increase in the time spent in the light box ($P < 0.01$) without affecting other parameters.

3.3. The hole-board test

The data are summarized in Table 2. PQS (50 and 100 mg/kg) significantly increased head-dip counts ($P < 0.01$) and PQS (100 mg/kg) increased head-dip duration ($P < 0.05$), which was similar to DZ (2.5 mg/kg). Both PQS and diazepam had no effect on locomotion.

3.4. Isolation-induced aggressive test

The results are represented in Fig. 3. Compared with the control group, PQS (50 and 100 mg/kg) produced a dose-dependent decrease in total fighting time. DZ, at the dose of 2.5 mg/kg, also produced a significant decrease in total fighting time.

4. Discussion and conclusions

Asian ginseng (*Panax ginseng* Meyer) and American ginseng (*Panax quinquefolium* L.) have become universally popular in recent years. The major active ingredients of both ginseng species have been demonstrated to be saponins (Nagai et al., 1972) and they contribute to their multiple medicinal properties

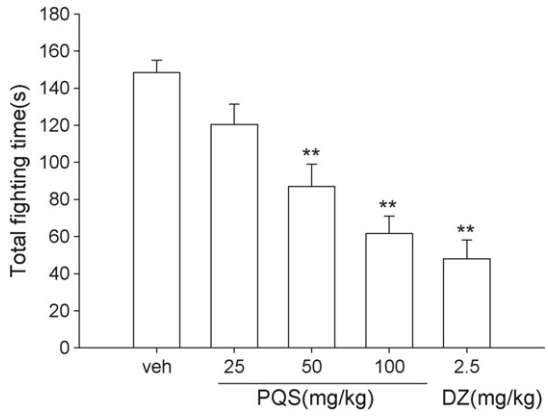

Fig. 3. Effects of PQS in the isolation-induced aggressive test. Results are expressed as means ± S.E.M. ($n = 14$). $^{**}P < 0.01$, compared with vehicle-treated mice.

(Court et al., 1996). The present study demonstrated for the first time that PQS has an anxiolytic effect.

The elevated plus-maze is a well-established animal model for testing anxiolytic drugs (Dawson and Tricklebank, 1995; Kulkarni and Reddy, 1996). In this test, the percentages of entries into opens arms and of time spent in open arms have generally been used as indices of anxiety. In the present study, PQS (50 mg/kg) increased the percentages of open arm entries and open arm time, indicating an anxiolytic effect. However, unlike DZ (2.5 mg/kg), a typical anxiolytic agent, PQS did not alter total arm entries, which suggested that PQS did not affect locomotor activity.

The light/dark box is also widely used for rodents as a model for screening anxiolytic or anxiogenic drugs, based on the innate aversion of rodents to brightly illuminated areas and on the spontaneous exploratory behavior of rodents in response to mild stressors, that is, a novel environment and light. It has been reported that simply the measurement of the time spent in the light area, but not the number of transfers, is the most consistent and useful parameter for assessing an anxiolytic action (Young and Johnson, 1991). The present study showed that PQS (50 and 100 mg/kg) could increase the time in the light area, suggesting again that PQS possesses anxiolytic properties.

The hole-board test provides a simple method for measuring the response of an animal to an unfamiliar environment and is widely used to assess emotionality, anxiety and/or responses to stress in animals. Takeda et al. (1998) showed that head-dipping behavior was sensitive to changes in the emotional state of the animal, and suggested that the expression of an anxiolytic state in animals may be reflected by an increase in head-dipping behavior. In the present study, PQS (50 and 100 mg/kg) increased head-dip counts and PQS (100 mg/kg) increased head-dip duration without changing locomotion. These results indicate that PQS has a significant anxiolytic effect in this paradigm.

The inhibition of isolation-induced aggressive behavior in male mice has been proposed as an animal model for assessing anxiolytic activity (White et al., 1991). In the present study, PQS (50 and 100 mg/kg) produced a significant decrease in total fighting time in comparison with vehicle, suggesting that PQS has an antiaggressive effect, which further supports its anxiolytic role.

The dose–response curve of PQS was bell-shaped in the elevated plus-maze, which is different from the other three anxiety paradigms in our test. It has been reported that some non-classic anxiolytic compounds, such as 5-HT_3 receptor antagonists and 5-HT_4 receptor antagonists (Vasar et al., 1993; Silvestre et al., 1996), also have a bell-shaped dose–response curve in the elevated plus-maze test. These phenomena suggest that (1) anxiety may be a nervous disorder mediated by multiple neuronal pathways in the central nervous system, and (2) different models of anxiety test predominately represent activities of certain neuronal systems. Therefore, under certain circumstances, some drugs cannot express their dose-dependent effects in anxiolytic studies.

Ginseng has also been used traditionally for the treatment of psychiatric disorders, such as anxiety and depression. It has been reported that ginseng produces anxiolytic effects and the saponin fraction plays an important role in the plus-maze model in mice (Cha et al., 2005). Among the three types of pure ginsenoside (Rb1, Rg1, and Ro), only ginsenoside Rb1 significantly increased both the frequency and duration of open arm entries, indicating that ginsenoside Rb1 is one of the active anxiolytic components of ginseng root (Carr et al., 2006). It is interesting to note that ginsenoside Rb1 has also been demonstrated to be effective in suppressing intermale aggression (Yoshimura et al., 1988a) and maternal aggression (Yoshimura et al., 1988b).

Although *Panax quinquefolium* contains many saponins in common with *Panax ginseng* and other *Panax* species, it has its own characteristic saponin profile that includes substantial amounts of the acetylated saponin quinquenoside R1, making it chemically different from other *Panax* species. Accordingly, the anxiolytic activity and the possible mechanisms may be different between saponins of *Panax ginseng* and *Panax quinquefolium*.

The pharmacological mechanism that might account for the anxiolytic effect of PQS has not been clearly identified. It has been reported that PQS can interact with $GABA_A$ receptors, suggesting that the GABAergic system is at least partly involved in the pharmacological activity of PQS (Yuan et al., 1998). The results showing that ginsenoside Rb1 and pseudoginsenoside-F11 (the components of PQS) antagonize scopolamine-induced memory impairment suggest that their effects may be associated with the cholinergic system in the central nervous system.

It is reported that pseudoginsenoside-F11 can also antagonize the behavior effects of morphine (Li et al., 2000) and the reduction in dopamine levels induced by methamphetamine in the mouse brain (Wu et al., 2003). Thus, it is plausible to assume that the possible mechanism of PQS on anxiety might be related to its effects on certain neuronal systems. However, which neuronal system is predominately involved requires further investigation.

References

Abramov, U., Raud, S., Koks, S., Innos, J., Kurrikoff, K., Matsui, T., Vasar, E., 2004. Targeted mutation of CCK(2) receptor gene antagonises behavioural changes induced by social isolation in female, but not in male mice. Behavioural Brain Research 155, 1–11.

Benishin, C.G., 1992. Actions of ginsenoside Rb1 on choline uptake in central cholinergic nerve endings. Neurochemistry International 21, 1–5.

Benishin, C.G., Lee, R., Wang, L.C.H., Liu, H.J., 1991. Effects of ginsenoside Rb1 on central cholinergic metabolism. Pharmacology 42, 223–229.

Bensky, D., Gamble, A., 1993. Chinese Herbal Medicine: Materia Medica. Eastland Press, Seattle.

Carr, M.N., Bekku, N., Yoshimura, H., 2006. Identification of anxiolytic ingredients in ginseng root using the elevated plus-maze test in mice. European Journal of Pharmacology 531, 160–165.

Cha, H.Y., Park, J.H., Hong, J.T., Yoo, H.S., Song, S., Hwang, B.Y., Eun, J.S., Oh, K.W., 2005. Anxiolytic-like effects of ginsenosides on the elevated plus-maze model in mice. Biological and Pharmaceutical Bulletin 28, 1621–1625.

Chen, S.W., Xin, Q., Kong, W.X., Min, L., Li, J.F., 2003. Anxiolytic-like effect of succinic acid in mice. Life Sciences 73, 3257–3264.

Court, W.A., Hendel, J.G., Elmi, J., 1996. Reversed-phase high-performance liquid chromatographic determination of ginsenosides of *Panax quinquefolium*. Journal of Chromatography A 755, 11–17.

Dawson, G.R., Tricklebank, M.D., 1995. Use of the elevated plus-maze in the search for novel anxiolytic agents. Trends in Pharmacological Sciences 16, 33–36.

Fu, Y., Ji, L.L., 2003. Chronic ginseng consumption attenuates age-associated oxidative stress in rats. Journal of Nutrition 133, 3603–3609.

Gao, N.N., Yu, S.R., Lu, R.M., 1995. The facilitative effect of *Panax quinquefolium* saponin on learning and memory in mice. Pharmacology and Clinics of Chinese Materia Medica 5, 12–14.

Guo, M., Wu, C.F., Liu, W., Yang, J.Y., Chen, D., 2004. Sex difference in psychological behavior changes induced by long-term social isolation in mice. Progress in Neuro-Psychopharmacology and Biological Psychiatry 28, 115–121.

Kimura, T., Saunders, P.A., Kim, H.S., Rheu, H.M., Oh, K.W., Ho, I.K., 1994. Interactions of ginsenosides with ligand-bindings of GABAA and GABAB receptors. General Pharmacology 25, 193–199.

Kulkarni, S.K., Reddy, D.S., 1996. Animal behavioral models for testing antianxiety agents. Methods and Findings in Experimental and Clinical Pharmacology 18, 219–230.

Lee, S.P., Honda, K., Rhee, Y.H., Inoue, S., 1990. Chronic intake of *Panax ginseng* extract stabilizes sleep and wakefulness in food-deprived rats. Neuroscience Letters 111, 217–221.

Lepicard, E.M., Joubert, C., Hagneau, I., Perez-Diaz, F., Chapouthier, G., 2000. Differences in anxiety-related behavior and response to diazepam in BALB/cByJ and C57BL/6J strains of mice. Pharmacology, Biochemistry and Behavior 67, 739–748.

Li, J., Lu, Z.Z., Lu, Y.F., 1993. Lipoprotein–cholesterol metabolism and antioxidative effects of *Panax quinquefolium* saponins on experimental hyperlipidemic rat. Chung Kuo Yao Hsueh Tsa Chih 28, 355–357.

Li, Z., Guo, Y.Y., Wu, C.F., Li, X., Wang, J.H., 1999. Protective effects of pseudoginsenoside-F11 on scopolamine-induced memory impairment in mice and rats. Journal of Pharmacy and Pharmacology 51, 435–440.

Li, Z., Wu, C.F., Pei, G., Guo, Y.Y., Li, X., 2000. Antagonistic effect of pseudoginsenoside-F11 on the behavioral actions of morphine in mice. Pharmacology, Biochemistry and Behavior 66, 595–601.

Lian, X.Y., Zhang, Z., Stringer, J.L., 2005. Anticonvulsant activity of ginseng on seizures induced by chemical convulsants. Epilepsia 46, 15–22.

Lu, Z.Z., Sui, D.Y., 1996. Recent progress on the effect of *Panax quinquefolium* saponins on the pharmacology of cardiovascular disease. Chinese Pharmacological Bulletin 12, 395–396.

Moraira, E.G., Nascimento, N., Rogero, J.R., 2000. Gabaergic-benzodiazepine system is involved in the crotoxin-induced anxiogenic effect. Pharmacology, Biochemistry and Behavior 65, 7–13.

Nagai, M., Ando, S., Tanaka, N., Tanaka, O., Shibata, S., 1972. Chemical studies on the oriental plant drugs. XXVIII. Saponins and sapogenins of ginseng: stereochemistry of the sapogenin of ginsenosides-Rb1, -Rb2 and -Rc. Chemical and Pharmaceutical Bulletin 20, 1212–1216.

Persons, W.S., 1986. American Ginseng: Green Gold. Bright Mountain Books, Asheville.

Petkov, V.D., Mosharov, A.H., 1987. Effects of standardized ginseng extract on learning, memory and physical capabilities. American Journal of Chinese Medicine 15, 19–29.

Silvestre, J.S., Fernandez, A.G., Palacios, J.M., 1996. Effects of 5-HT4 receptor antagonists on rat behavior in the elevated plus-maze test. European Journal of Pharmacology 309, 219–222.

Takeda, H., Tsuji, M., Matsumiya, T., 1998. Changes in head-dipping behavior in the hole-board test reflect the anxiogenic and/or anxiolytic state in mice. European Journal of Pharmacology 350, 21–29.

Vasar, E., Peuranen, E., Oopik, T., Harro, J., Mannisto, P.T., 1993. Ondansetron, an antagonist of 5-HT3 receptor, antagonizes the antiexploratory effect of caerulein, an agonist of CCK receptors, in the elevated plus-maze. Psychopharmacology 110, 213–218.

White, S.M., Kucharik, R.F., Moyer, J.A., 1991. Effects of serotonergic agents on isolation-induced aggression. Pharmacology, Biochemistry and Behavior 39, 729–736.

Wu, C.F., Liu, Y.L., Song, M., Liu, W., Wang, J.H., Li, X., Yang, J.Y., 2003. Protective effects of pseudoginsenoside-F11 on methamphetamine-induced neurotoxicity in mice. Pharmacology, Biochemistry and Behavior 76, 103–109.

Yang, S., Qu, J., Zhong, G., Zhang, W., 1992. Protective effects of *Panax quinquefolium* saponin on oxidative damage of cultured rat cardiac cells. Chung Kuo Chung Yao Tsa Chih 17, 555–557.

Yoshimura, H., Watanabe, K., Ogawa, N., 1988a. Psychotropic effects of ginseng saponins on agonistic behavior between resident and intruder mice. European Journal of Pharmacology 146, 291–297.

Yoshimura, H., Watanabe, K., Ogawa, N., 1988b. Acute and chronic effects of ginseng saponins on maternal aggression in mice. European Journal of Pharmacology 150, 319–324.

Young, R., Johnson, D.N., 1991. A fully automated light/dark apparatus useful for comparing anxiolytic agents. Pharmacology, Biochemistry and Behavior 40, 739–743.

Yuan, C.S., Attele, A.S., Wu, J.A., Liu, D., 1998. Modulation of American ginseng on brainstem GABAergic effects in rats. Journal of Ethnopharmacology 62, 215–222.