2002. Vol. 3. No. 1. 71-75
Korean Journal of Oriental Medicine

Original Article

The Effects of Water Extract of *Genus Panax* on Rat Blood Vessels

Ki-Deog Yu, Hyung-Hwan Kim, Ho-Kun Park[1)], Duk-Kyun Ahn, Ho-Young Choi

Department of Herbology, College of Oriental Medicine, KyungHee University, Korea Institute of Science and Technology[1)]

Abstract

Objective: We have examined the relaxational response to the water extract of genus Panax in rat thoracic aorta and mesenteric artery.

Methods: Segments of thoracic aorta and mesenteric artery obtained from rats immediately after delivery were mounted in organ baths superfused on a polygraph.

Results: We found that the thoracic aorta segments responded to the water extract of genus Panax with a dose-dependent vasorelaxation. At 10^{-5}M 5-hydroxytrptamine (5-HT), the maximal contraction force was 94.9% of the maximum KCl-response. At 10^{-5}M 5-HT-induced contraction, the contractile response of thoracic aortic rings was inhibited by 54.7%, 36.3% and 31.3% after addition of a high concentration (100 mg/ml) water extract of *Panax ginseng, Panax japonicus* and *Panax quinquefolium*. The contractile response of mesenteric arteries were inhibited by 88.3%, 87.7%, and 70.3% after addition of a high concentration (100 mg/ml) water extract of *Panax ginseng, Panax japonicus* and *Panax quinquefolium*.

Conclusions: Water extract of genus Panax induced relaxation in the isolated rat thoracic aorta and mesenteric artery composed of endothelium-independent relaxation and dose-dependent relaxation.

Korean Journal of Oriental Medicine 2002;3(1):71-75

Key Words: *Genus Panax*, 5-hydroxytryptamine, phenylephrine, relaxation

Introduction

The roots of genus Panax (*Panax ginseng, Panax quinquefolium and Panax japonicus*) have been used as agents for the regulation of blood pressure and prescribed in Korean and Chinese medicine as a tonic or prophylactic for chronic bronchitis, hypertension and ischemic disease. It is used to replenish the vital energy and to promote the secretion of body fluids for the treatment of shock, prostration, etc. It has been shown to possess the effects of stimulating the central nervous system, cardiotonic, antifatigue and stimulating the mechanism of blood formation[1-3)].

Panax ginseng has elicited relaxation of penile endothelial cells and ginsenosides, saponins extracted from *Panax ginseng*, relax pulmonary blood vessels and the basilar arteries[4,5)]. Nitric oxide plays a primary role in ginsenosides-enhanced relaxation within the cerebral vasculature[5)].

Panax japonicus and *Panax quinquefolium* as well as *Panax ginseng* have shown the inhibition of contractions[6,7)].

We sought to determine the relative effect of genus Panax on rat thoracic aorta and mesenteric artery.

· Correspondence to : Ho-Young Choi, College of Oriental Medicine, Kyung Hee University. Seoul, 130-701, Korea; Tel : 82-2-961-9372, E-mail : hychoi@khu.ac.kr

Materials and Methods

1. Animals

Male Sprague-Dawley rats weighing 300 ~ 350 g were used for all experiments. They were purchased from Semtaco, Animal Ltd. They were subjected to preliminary group-housing (3-4 per cage) under controlled conditions with food and water ad *libitum*.

2. Plant Material

Dried roots of genus Panax were purchased in June 2002, at Keumsan Market, Korea (*Panax ginseng*, 4 years), and CheongPyung Market, Kwangdong, China (*Panax quinquefolium*), and in August 2002, in Tokyo, Japan (*Panax japonicus*).

3. Preparation of genus Panax

A water extract of genus Panax was prepared from dried roots: they were cut into small pieces and mashed with a mortar and pestle. The roots (100 g) were extracted with boiling water (50 g/L); the total volume was 2L.

4. Materials

NaCl, KCl, NaH_2PO_4, $MgSO_4$, $CaCl_2$, $NaHCO_3$, glucose, 5-hydroxytryptamine, acetylcholine, N^G-monomethyl-L-arginine (L-NMMA) and 3-[3-(chloramidopropyl)-dimetylammonio]-1-propane sulfonate (CHAPS) were purchased from Sigma (Sigma, USA).

5. Preparation of isolated aortic rings

Male Sprague-Dawley rats (300-350 g) were killed with an overdose of chloral hydrate (400 mg/kg, i.p.) and the thoracic aorta and mesenteric artery were removed and cleaned of adherent tissue. The thoracic aorta and mesenteric artery were mounted on a length of scoured polythene tubing and placed in a petri dish containing modified physiological salt solution (PSS) of the following (mM) composition: NaCl 119.0, $NaHCO_3$ 25.0, KCl 4.7, KH_2PO_4 1.2, $MgSO_4$ 1.2, glucose 11.0, $CaCl_2$ 0.25. The aorta was cleared of surrounding adipose tissue and the endothelium was removed by gentle rubbing of the intimal surface with the polythene tube. Six to nine ring segments (2-3 mm length) were prepared from each aorta and were mounted between two stainless-steel wires in 10 ml organ baths, thermostatistically controlled at 37℃, containing modified PSS.

The solution was bubbled with a gas mixture consisting of 95% O_2 and 5% CO_2 in order to keep a pH in the bath of around 7.35-7.38. Experiments were carried out after the vessel had equilibrated, usually within 1-2 h of mounting. The tension was recorded isometrically with a Grass FT03C force-displacement transducer and registered on a Grass Model 7 Polygraph. The vessels were given an initial passive load of about 1.5-2 g and allowed to equilibrate for at least 30 min prior to the experiments. After the equilibrating period, vessels were stimulated with KCl (100 mM) in order to obtain a reference contraction. This contraction was defined as the maximal contraction to KCl. Vessels that did not respond or responded abnormally were not tested further. Genus Panax and other substances were dissolved in 0.9% NaCl and given to the baths in volumes of 10 ml. The response to the added substances (contraction, relaxation) was expressed as a percentage of the maximal KCl-induced contraction exhibited by each ring.

6. Removal of endothelium

To preclude the possible role of endothelium in the vasodilation of genus Panax, the tests were

conducted in endothelium-denuded preparation. The endothelium was removed by gentle rubbing against the teeth of a pair of forceps. Success of the removal of the endothelium was characterized using the failure of 10^{-6}M acetylcholine to relax the rings precontracted with 10^{-5}M 5-HT.

7. Measurement of Vasodilation

After the resting tension became stabilized, 10^{-5}M 5-HT was administrated into a bathing buffer to induce a rapid increase of vascular tone followed by the stable vasoconstriction.

Treatments groups were administered genus Panax from concentrations of 0.01 to 100 mg/ml to observe vasodilation (the decrease of tonic contraction). Concentration-relaxation curves were generated in a cumulative fashion.

8. Statistical analysis

Analysis of data from two groups was performed using Student's t-test. Data from several groups were examined using analysis of variance (ANOVA), using the computer program GraphPad Prism, GraphPad Software, SanDiego, CA. Significance levels were set as follows: P = 0.05(*), P=0.01(**), P=0.001(***).

Results

1. Dose-dependent contraction of 5-HT in thoracic aorta

5-HT (0.001 ~ 10 μM) produced a concentration-dependent contraction of the thoracic aorta. Thoracic aorta segments responded to 5-HT with a dose-dependent vasocontriction. At 10^{-5}M 5-HT, the maximal contractile response was 94.9% of the maximum KCl-response (Fig. 1).

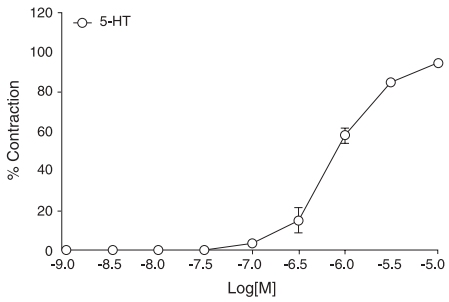

Fig. 1. Contractional response of thoracic aorta to 5-HT at concentration ranging from 10-3 to 10 μM. Values represent mean ± SEM on the maximal contractional response. On the X-axis: M; on the Y-axis: % Contraction. n=3.

2. Effects of genus Panax on 5-HT-induced relaxation of thoracic aortas

Thoracic aorta segments responded to 5-HT (0.001 ~ 10 μM) with a dose-dependent vasoconstriction. At 10-5M 5-HT, the contractile response was 94.9% of the maximum KCl-response. At 100 mg/ml *Panax ginseng*, *Panax japonicus* and *Panax quinquefolium*, the relaxational response at thoracic aorta were 53.5%, 36.3% and 31.3% of the maximum 5-HT induced contraction (Fig. 2).

3. Effects of genus Panax on 5-HT-induced relaxation of mesenteric arteries

Mesenteric artery segments responded to 5-HT (0.001 ~ 10 μM) with a dose-dependent vasocontriction. At 10^{-5}M 5-HT the contractile response was 94.9% of the maximum KCl-response. At 100 mg/ml *Panax ginseng, Panax japonicus* and *Panax quinquefolium*, the relaxational response at mesenteric artery were 88.3%, 87.7% and 70.3% of the maximum 5-HT induced contraction (Fig. 3).

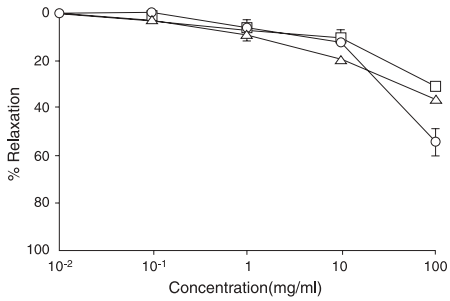

Fig. 2. Genus Panax (Panax ginseng (○); Panax japonicus (△); Panax quinquefolium (□)) caused concentration-dependent relaxation in the 5-HT-induced contraction of rat thoracic aorta without endothelium. Data represent mean ± SEM. n=3.

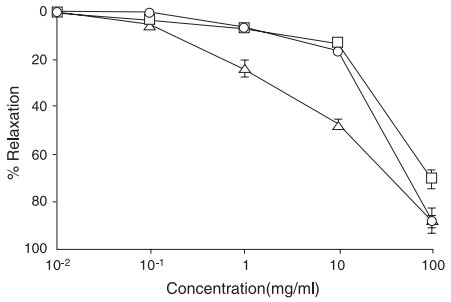

Fig. 3. Genus Panax (Panax ginseng (○); Panax japonicus (△); Panax quinquefolium (□)) caused concentration-dependent relaxation in the 5-HT-induced contraction of rat mesenteric artery without endothelium. Data represent mean ± SEM. n=3.

Discussion

Panax ginseng C.A. Mey. (Araliaceae) is one of the oldest tonics; the source of earliest record of this drug is 『Sinnong Boncho Kyung』. It is used to replenish the vital energy and to promote the secretion of bodily fluids for the treatment of shock, prostration, etc. It has been shown to possess the effects of stimulating the central nervous system, cardiotonic, antifatigue and stimulating the mechanism of blood formation[1-3].

5-HT produced a concentration- and dose-dependent contraction of the Sprague-Dawley rat thoracic aorta. Thoracic aorta segments responded to 5-HT with a dose-dependent vasoconstriction.

Previously, Panax ginseng elicited relaxation of penile endothelial cells and ginsenosides, saponins extracted from *Panax ginseng* relax pulmonary blood vessels and the basilar arteries[4,5]. Nitric oxide plays a primary role in ginsenosides-enhanced relaxation within the cerebral vasculature[5].

It is reported that *Panax quinquefolium* and *Panax japonicus* as well as *Panax ginseng* showed the inhibition of intracellular and extracellular Ca^{2+}-dependent contraction and action activating the blood circulation[6,7].

In our results, at high concentration, the relaxational responses of *Panax ginseng, Panax japonicus* and *Panax quinquefolium* at thoracic aorta were 53.5%, 36.3%, and 31.3% of the maximum 5-HT induced contraction. The responses were significantly enhanced by genus Panax (*Panax ginseng, Panax japonicus* and *Panax quinquefolium*; 10^{-2}, 10^{-1}, 1, 10 and 100 mg/ml); the relaxational responses at mesenteric arteries were more potent than thoracic aortas.

In conclusion, in the present study the water extracts from three kinds of Panax were found to show dose-dependent relaxation response to thoracic aorta and mesenteric artery, and the effects of mesenteric arteries were more sensitive than thoracic aorta. In addition, our results may support their potential as therapeutic strategies in hypertension, cerebral ischemia and other related vascular dysfunctions.

References

1. Liu Chang-Xiao, Xiao Pei-Gen and Li Da-Peng. Modern Research and Application of Chinese Medicinal Plants. Hong Kong: Hong Kong Medical Publisher. 2000:501-17.
2. Cui M, Song F, Liu Z, Liu S. Metal ion adducts in the structural analysis of ginsenosides by electrospray ionization with multi-stage mass spectrometry. Rapid Commun Mass Spectrom. 2001; 15(8):586-95.
3. Cui M, Song F, Zhou Y, Liu Z, Liu S. Rapid identification of saponins in plant extracts by electrospray ionization multi-stage tandem mass spectrometry and liquid chromatography/tandem mass spectrometry. Rapid Commun Mass Spectrom. 2000; 14(14):1280-6.
4. Chen X, Salwinski S and Lee TJF. Extracts of ginkgo biloba and ginsenosides exert cerebral vasorelaxation via nitric oxide pathway. Clinical and Experimental Pharmacology and Physiology. 1997; 24:958-959.
5. Xiu Chen and Lee, TJF. Ginsenosides-induced nitric oxide-mediated relaxation of the rabbit corpus cavernosum. British Journal of Pharmacology. 1995; 115:15-18.
6. Guan L, Yi X, Feng Q, Yang L. Effects of saponins from stems and leaves of *Panax quinquefolium* L. on the contraction of rabbit aortic strips. Zhongguo Zhong Yao Za Zhi. 1996 Jul; 21(7):431-4.
7. Wu JX, Chen JX. Depressant actions of Panax notoginseng saponins on vascular smooth muscles. Zhongguo Yao Li Xue Bao. 1988 Mar; 9(2):147-52.