Vasorelaxant effect of Salvia miltiorrhiza Radix extract on isolated rat aorta

Kim Hyun-Young¹ · Kim Young Kyun¹ *

I. Introduction

Salvia miltiorrhiza Radix (丹蔘, SR), commonly known as Dansam or Danshen, is from the root and rhizome of Salvia miltiorrhiza Bunge that belongs to the family of Labiatae, and it is a commonly and widely used traditional herbal medicine for the treatment of cardiovascular diseases such as stroke, angina pectoris and myocardial infarction. The cardio-protective efficacy of SR has been studied in animal ischemia/reperfusion experiments¹-⁵) and regional cerebral blood flow6). And it's mechanism may involve the ability of SR to enhance antioxidant activities to decrease or abolish the production of free radicals⁷).

SR has also been shown to attenuate the increase in intracellular calcium induced by anoxia/reoxygenation in isolated ventricular myocytes, which would decrease the transformation of xanthine oxidase from xanthine dehydrogenase to reduce the production of oxygen free radicals⁸). In addition, SR lowered the viscosity of whole blood, accelerated electrophoresis of red blood cells, and improved peripheral circulation⁹). The vasodilator and
hypotensive actions of SR probably contributed to these effects\textsuperscript{10–12}.

In the present experiments, therefore, we examined the vasorelaxations induced by SR extract, and investigated the relaxing mechanisms in comparison with the effect of each constituent using rat aorta ring strip.

II. Materials and Methods

1. Plant extracts
Dried root of Salvia miltiorrhiza (Salvia miltiorrhizae Radix, SR) was obtained from a local market and ground using a commercial food mixer. This powder was extracted consecutively under reflux with water for 1 h. The resulting water extract was evaporated under reduced pressure at 37–40°C of temperature and lyophilized (SREx). This solid extract was stored at −20°C until use. A solution was prepared with distilled water at a concentration of 100–300 mg/ml on the day of the experiment. Water extract of SR were extracted again three times with n-hexane in the sonicator. The suspension was filtered and evaporated under reduced pressure at low temperature and lyophilized. The residue of hexane extract was obtained. The remaining was extracted again with chloroform and methanol sequentially to yield chloroform and methanol extract fractions.

2. Artery ring preparation
Male Sprague-Dawley rats (200–250 g each) were sacrificed by stunning and bleeding. The descending thoracic aorta was dissected free from surrounding connective tissues and cut into rings of 2–3 mm in length. Rings were then transferred into 4 ml horizontal type organ chambers, and were bathed in physiological salt solution (PSS) at 37°C containing (mmol/l): NaCl, 136.9; KCl, 5.4; CaCl2, 1.5; MgCl2, 1.0; NaHCO3, 23.8; glucose, 5.5, and EDTA 0.01 (pH 7.4); and gassed with 95% O2 and 5% CO2. Rings were mounted on stainless steel hooks connected to a force-displacement transducer (FT03, Grass, Rhode Island, USA) connected to a polygraph system (RPS212, Grass, Rhode Island, USA) and a computer analyzer (Power Lab 400, MacLab System, Castle Hill, Australia) were used. A basal tension of 1 g was applied. Some segments were mechanically denuded of endothelium by gentle rubbing with a moistened cotton swab.

3. Experimental protocols
All rings were equilibrated for 60 min under a resting tension of 1 g and then exposed repeatedly to 72 mmol/l KCl PSS until responses became stable. Control contraction was produced using 300 mmol/l norepinephrine (NE). After sustained tension (60% or 80% of the maximal contraction to 72 mmol/l KCl PSS in endothelium-intact or -denuded rings) was obtained, SREx or vehicles were added sequentially to the bath solution. The high-potassium solution was prepared by replacing NaCl of PSS with equimolar KCl. In experiments where specific inhibitors were used, they were added 20 min before precontraction. The inhibitors tested were NG-nitro-L-arginine (LNNA, 10 mmol/l), NO-nitro-L-arginine methyl ester (NAME, 10 mmol/l) or NG-methyl-L-arginine (NMMA, 10 mmol/l) as an inhibitor of NO synthesis, methylene blue (1 mmol/l) or 1H-[1,2,3] oxadiazole

---

\textsuperscript{10–12}