EXPERIMENTAL RESEARCH

Effects of Corocalm (Shuguan Capsule, 疏冠胶囊) on Acute Myocardial Ischemia in Anesthetized Dogs

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ABSTRACT

Objective: To investigate the effects of Corocalm (Shuguan Capsule, 疏冠胶囊) on acute myocardial ischemia in anesthetized dogs and its possible therapeutic mechanism. Methods: The acute ischemia model was established by ligating the left anterior descending (LAD) artery. Twenty-five dogs were randomly divided into 5 groups (5 dogs in each group): the control group (treated with normal saline 3 mL/kg), the refined Guanxin Capsule group (精制冠心胶囊, GXC 200 mg/kg), high and low dose Corocalm groups (48.5 mg/kg for low dose group and 194.0 mg/kg for high dose group) and the Diltiazem group (5 mg/kg). The animals were treated via a single duodenal administration after the model was established. The experiments used epicardial electrocardiogram (EECG) to measure the scope and degree of myocardial ischemia. Simultaneously, the coronary blood flow (CBF) and serum activity levels of creatine phosphokinase (CK) and lactate dehydrogenase (LDH) were measured by electromagnetic flow meter and automatic biochemical analyzer respectively. The plasma endothelin (ET) content was quantified by radioimmunoassay. Results: Corocalm (48.5 mg/kg and 194.0 mg/kg) significantly decreased the degree and scope of myocardial ischemia, reduced the infarct area, markedly increased the CBF, and inhibited the increase of CK and LDH activities and ET levels induced by myocardial ischemia/infarction. Conclusion: Corocalm could improve the state of acute myocardial ischemia and infarction in dogs. The mechanism of action might be correlated to increasing CBF, inhibiting CK and LDH activities and preventing ET release.

KEY WORDS Corocalm (Shuguan Capsule), acute myocardial ischemia, coronary blood flow, endothelin, mongrel dog

In recent years, the incidence of cardiovascular diseases is gradually increasing. Myocardial ischemia/infarction has become one of the leading causes of death in the aged and even in the young. Corocalm (Shuguan Capsule, 疏冠胶囊) is a candidate anti-ischemic drug consisting of Radix Salvia miltiorrhiza Bge., Radix Ligusticum chuanxiong Hort., Flos Carthamus tinctorius L., and Radix Paeonia lactiflora pall. "Shu Guan" in Chinese means "dredging the blockage of coronary vessel". Corocalm is the commercial name, which means "calming the coronary heart disease". It promotes blood circulation, prevents blood stasis and clears the channels or blood vessels. It may, therefore, be applied for the treatment of acute heart attack, chronic myocardial ischemia and congestive heart failure. However, the effects of Corocalm on acute myocardial ischemia and the underlying mechanism remain unclear. In this study, three standard indices associated with the cardiovascular function were measured to investigate effects of Corocalm on myocardial ischemia after the ligation of the left anterior descending (LAD) coronary artery in anesthetized dogs.

METHODS

Drugs and Reagents

Corocalm, consisting of Radix Salvia miltiorrhiza Bge 5 000 g, Radix Ligusticum chinences Hort 2 500 g, Flos Carthamus tinctorius L. 2 500 g, and Radix Paeonia lactiflora pall. 2 500 g per 1 000 capsules. One gram of crude drug equals 24.2 mg of dry extract. The dose used in this study was calculated by the weight of the dry extract which was supplied by the Institute of Chinese Materia Medica, China Academy of Chinese Medical Sciences (batch No. 981026). Refined Guanxin Capsule (精制冠心胶囊, GXC, 0.35 g drug powder/capsule) was produced by Tonghua Dongbao Pharmaceutical Co., Ltd. (batch No. 960801). Diltiazem Tablet (diltiazem...
hydrochloride controlled release tablet, 30 mg/tablet) was produced by Yatai Pharmaceutical Factory, Zhejiang (batch No. 960404). Creatine phosphokinase (CK) and lactate dehydrogenase (LDH) assay kits were purchased from Beijing Zhongsheng Bio-Tech Co., Ltd. (batch No. 9812). Radioimmunoassay (RIA) kit for endothelin (ET) in plasma was purchased from Beijing Furui Bioengineering Co., Ltd. (batch No. 9812). Nitroblue tetrazolium (N-BT) was supplied by Sigma Co.

Animals and Grouping

Twenty-five mongrel dogs of both sexes were provided by Beijing Experimental Animal Reproduction and Regulation Center (Certificate No. Jing-030). These dogs had an average weight of 13.4 ± 2.3 kg, and were divided into 5 groups (5 in each group): a vehicle control group (treated with normal saline at 3 mL/kg), a GXC (200.0 mg/kg) group, a Corocalm low dosage (48.5 mg/kg) group and high dosage (194.0 mg/kg) group and a Diltiazem (5.0 mg/kg) group. All drugs were prepared before the experiment with normal saline to an isovolumetric solution and were given only once via the duodenum at 3 mL/kg after model establishment.

Procedure of Myocardial Ischemic Model

The dogs were anesthetized with pentobarbital (30 mg/kg) by intravenous injection. Tracheal intubation was performed and an artificial respirator (SC-3, Shanghai Medical Equipment Factory) was connected. The ischemic model in dogs was prepared as described previously: The heart was exposed by opening the chest. The multi-point epicardial electrodes were sutured on the ventricular surface and a physiological polygraph (RM-6100, Nihon Kohden, Japan) was connected to record a 30-point epicardial electrocardiogram (EECG). EECG was recorded as pre-treatment control at 15 min after the ligation of the LAD. At the same time, drugs or saline were administrated via the duodenum. Thirty points of EECG were recorded at 15, 30, 45, 60, 90, 120 and 180 min after the medication. An ST segment elevation of more than 2 mV was regarded as the ischemic criteria to calculate the degree of myocardial ischemia (total mV of ST segment elevating, Σ-ST) and myocardial ischemic scope (total point number of ST segment elevating, N-ST).

Measurement of Coronary Blood Flow in the Myocardium

The left circumflex branch of coronary artery was isolated and the probe of an electromagnetic flow meter (MF-1100, Nihon Kohden, Japan) was placed on the vessel origin to determine the coronary blood flow (CBF) before and after coronary ligation.

Determination of Serum CK, LDH Activities and Plasma ET Content

Blood samples were collected from the coronary venous sinus before coronary ligation at 0 min (15 min after LAD ligation), 30, 60, 90, 120, and 180 min after medication. Serum CK and LDH activities at different time points were determined by automatic biochemical analyzer (RA-1000, TECHNICON, USA). Blood samples for ET determination were anticoagulated with 3.8% sodium citrate (V/V is 1/9) and centrifuged at 150 × g for 15 min. The plasma ET content was quantified by radioimmunoassay (RIA) with γ-counter (FT-630G, Nuclear Devices Factory, Beijing, China).

Calculation of Myocardial Infarction Area

The EECG record was ended at 180 min. The heart was taken out, washed with normal saline and weighed immediately. Under the ligation line, the ventricle was transversely divided into five pieces with equal thickness. The tissues were then infiltrated with N-BT staining solution at 25 °C for 15 min. Both infarct areas (N-BT non-stained area) and non-infarct areas (N-BT stained area) were determined by multimedia color pathological image analytical system (MPIAS-500, Beijing Konghai Company, China). The infarction percentage of the entire heart or ventricle was calculated.

Statistical Analysis

To eliminate the individual differences, the percent of pre-administration control (0 min) was proposed as 100%. The percent change of each parameter was calculated as: post-administration value/pre-administration value × 100%. All data are expressed as ±s. Statistical analysis was performed using t-test. P<0.05 was considered significant.

RESULTS

Effect of Corocalm on the Degree of Myocardial Ischemia

Dogs treated with Corocalm at 48.5 mg/kg and 194.0 mg/kg all showed significant alleviation to some degree. At 60 min after administration, the Σ-ST of dogs treated with Corocalm was reduced from 294 ± 11 mV and 296 ± 15 mV to 268 ± 22 mV (8.8%) and 203 ± 78 mV (32.1%), respectively, showing a significant difference when compared with that of the vehicle control.