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Haemodynamic effects of a selective adenosine A_{2A} receptor agonist, CGS 21680, in chronic heart failure in anaesthetized rats

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1 Recently we demonstrated that the administration of an A_{2A} adenosine receptor agonist, CGS 21680, to anaesthetized rats with acute heart failure (1 h post-coronary artery ligation) resulted in an increase in cardiac output. In the present investigation, the effects of CGS 21680 on cardiac output, vascular resistance, heart rate, blood pressure and mean circulatory filling pressure (Pmcf) were investigated in anaesthetized rats with chronic heart failure (8 weeks post-coronary artery ligation).

2 Experiments were conducted in five groups ($n=6$) of animals: sham-operated vehicle-treated (0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) animals in which the occluder was placed but not pulled to ligate the coronary artery; coronary artery-ligated vehicle-treated animals; and coronary artery-ligated CGS 21680-treated (0.1, 0.3 or 1.0 μg kg^{-1} min^{-1}) animals.

3 Baseline blood pressure, cardiac output and rate of rise in left ventricular pressure ($+dP/dt$) were significantly reduced in animals with coronary artery ligation when compared to sham-operated animals. Coronary artery ligation resulted in a significant increase in left ventricular end-diastolic pressure, Pmcf and venous resistance when compared to sham-operated animals.

4 Administration of CGS 21680 at 0.3 and 1.0 μg kg^{-1} min^{-1} significantly ($n=6$; $P<0.05$) increased cardiac output by $19\pm4\%$ and $39\pm5\%$, and heart rate by $14\pm2\%$ and $15\pm1\%$, respectively, when compared to vehicle treatment in coronary artery-ligated animals. Administration of CGS 21680 also significantly reduced blood pressure and arterial resistance when compared to coronary artery-ligated vehicle-treated animals. Infusion of CGS 21680 also significantly reduced venous resistance when compared to vehicle-treated coronary artery-ligated animals.

5 The results show that heart failure is characterized by reduced cardiac output, and increased left ventricular end-diastolic pressure, venous resistance and Pmcf. Acute treatment with CGS 21680 in animals with chronic heart failure decreased left ventricular end-diastolic pressure and increased cardiac output. This increase in cardiac output was the result of reduced arterial and venous resistances and increased heart rate.

Keywords: Chronic heart failure; adenosine receptors; cardiac output; mean circulatory filling pressure; vascular resistance

Introduction

Vasodilators are widely used clinically for the treatment of congestive heart failure (for review see Parmley, 1989; Bonarjee & Dickstein, 1996). The usefulness of vasodilators has been attributed to their ability to reduce peripheral vascular resistance and afterload, thereby, augmenting left ventricular function (for review see Bonarjee & Dickstein, 1996). Some of these drugs have also been found to produce venodilatation which results in an increase in venous capacitance, thereby, lowering preload (for review see Amsterdam *et al*., 1978). In addition, the increase in venous capacitance is accompanied by the relief of pulmonary congestion and dyspnea (for review see Amsterdam *et al*., 1978). Moreover, the reduction in preload can result in a reduced left ventricular filling volume, thus reducing cardiac work-load.

Adenosine and adenosine analogues have been shown to lower blood pressure (Fozard & Carruthers, 1993; Stella *et al*., 1995) and reduce systemic vascular resistance (Hernandez *et al*., 1995; Nekooeian & Tabrizchi, 1996). In addition, adenosine and its analogues have been reported to increase total venous capacitance (Glick *et al*., 1992) and reduce venous tone and venous resistance (Tabrizchi, 1997). Based on pharmacological evidence, at least four sub-types of adenosine receptors (A_1, A_{2A}, A_{2B} and A_3) have been reported to be responsible for regulating cardiovascular function (for review see Collis & Hourani, 1993; Linden, 1994; Olah & Stiles, 1995). Specifically, activation of A_2 and A_3 adenosine receptor subtypes produce hypotension while activation of A_1 receptors results in bradycardia.

The activation of A_2 receptors *in vivo* has been reported to result in a reduction of arterial pressure and peripheral resistance and an increase in cardiac output (Webb *et al*., 1991; Nekooeian & Tabrizchi, 1996; Tabrizchi, 1997). We previously demonstrated that cardiac output was impaired following an increase in peripheral resistance and that the administration of a selective A_{2A} adenosine receptor agonist, 2-*p*-(2-carboxyethyl)phenethylamino-5′-N-ethylcarboxamido-adenosine (CGS 21680) (Jarvis *et al*., 1989), attenuated the impairment (Nekooeian & Tabrizchi, 1996). Moreover, we also recently demonstrated that the administration of CGS 21680 to rats in a state of acute heart failure (1 h post-coronary artery ligation) resulted in an increase in cardiac output. This increase in cardiac output was predominantly due to reduced resistance to venous return and, thus, reduced preload (Nekooeian & Tabrizchi, 1998). In the present investigation, we have attempted to further examine the influence of CGS 21680 on cardiac output, vascular resistance (arterial & venous), heart rate, cardiac contractility and left ventricular end-diastolic pressure in animals with chronic heart failure (8

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weeks post-coronary artery ligation). Unlike acute heart failure, chronic heart failure results in morphological/structural changes and cardiovascular remodeling over time (for review see Zelis *et al*., 1981; Goldman & Raya, 1995). The many differences between animals with acute versus chronic heart failure are, in part, the result of changes in neurohormal factors (due to increased activity of the sympathetic nervous system and renin-angiotensin system) that ultimately result in cardiac hypertrophy, left ventricular dilatation, alteration of responses to vasoactive substances and pulmonary congestion (van Wijngaarden *et al*., 1991; Thuillez *et al*., 1995; Ganguly *et al*., 1997; Mulder *et al*., 1997). Therefore, the primary goals of our present investigation were (a) to examine the effects of CGS 21680 in an established chronic heart failure model (Pfeffer *et al*., 1979), and (b) to compare the haemodynamic effects of CGS 21680 in chronic heart failure to that of acute heart failure from our previous study (Nekooeian & Tabrizchi, 1998).

Methods

Coronary artery ligation

Male Sprague-Dawley rats (200–235 g) were anaesthetized with halothane (5% in 100% oxygen, induction; 1.5% in 100% oxygen, maintenance). A left thoracotomy was performed at the level of the fourth intercostal space and the heart was exposed. The left main coronary artery was ligated 2–4 mm from its origin using a 6-0 Proline suture. In the sham-operated group, the suture was passed around the coronary artery but it was not tightened. The chest wall and skin incisions were closed in layers. Before closure of the skin incision, local anesthetic (2.0% Bupivacain) and antibiotic powder (Cicratin) were applied to the wound. Each animal was allowed to recover from the anesthesia. Animals were then housed separately under 12 h light/dark cycles and maintained on standard rat chow with water *ad libitum*.

Eight weeks later, animals were anesthetized by an intraperitoneal injection of sodium pentobarbital (65 mg kg^{-1}). Catheters (polyethylene tubing; I.D. 0.58 mm, O.D. 0.965 mm) were inserted into the left and right iliac arteries and veins. The left venous catheter was advanced into the inferior vena cava and used for the measurement of central venous pressure. The left arterial and right venous catheters were used for the measurement of blood pressure and drug/vehicle administration, respectively, while the right arterial catheter was used for blood withdrawal of radiolabeled microspheres. An additional catheter was inserted into the left ventricle *via* the right carotid artery for the measurement of left ventricular end-diastolic pressure and injection of radiolabeled microspheres. A saline-filled balloon-tipped catheter was placed in the right atrium *via* the right external jugular vein for the purpose of transient circulatory stop as necessary for the measurement of mean circulatory filling pressure (Pmcf) (Tabrizchi *et al*., 1993). Pmcf is an index of the body's total venous tone, and it is independent of cardiac contractility, arterial resistance and heart rate since it is measured at a time when the circulation is stopped. Pmcf predominately reflects venous tone and is inversely dependent on venous compliance (Pang, 1994).

All catheters were filled with heparinized saline (25 iu mL^{-1}). Body temperature was maintained at 37°C *via* a rectal thermometer and a heating pad connected to a Thermistemp Instrument Controller (Model 71; Yellow Spring Instrument Co., OH, U.S.A.). Arterial blood pressure, as well as left ventricular end-diastolic and central venous pressures were recorded with a pressure transducer (Model PD23B; Gould Statham, CA, U.S.A.) connected to a polygraph (Model 79D; Grass Instruments Co., MA, U.S.A.). Heart rate was derived from the upstroke of the arterial pulse pressure by a tachograph (Model 7P4G; Grass Instruments Co., MA, U.S.A.). The rate of rise of left ventricular pressure (+dP/dt) was measured using an electronic differentiator (Model 7P20C; Grass Instruments Co., MA, U.S.A.). Cardiac output was measured using the reference sample microsphere method and Pmcf was measured after circulation was transiently stopped by inflating the balloon in the right atrium. Final arterial pressure and venous plateau pressure were recorded at 5–7 s after the circulatory stop (Pang & Tabrizchi, 1986).

Reference sample microsphere method

This technique has been described in detail elsewhere (Pang, 1983). Briefly, suspensions of microspheres (15 μm diameter; Du Pont Canada Inc.) labeled with ^{57}CO (25,000–30,000 in 150 μl) were injected into the left ventricle over a period of 10 s. Blood was withdrawn from the right femoral artery at the rate of 0.35 mL min^{-1} starting 15 s before microsphere injection using an infusion/withdrawal pump (Model 940; Harvard Apparatus Inc., MA, U.S.A.). The blood sample and syringes used for injection of microscopes or withdrawal of blood were counted for radioactivity at 80–160 Kev using a Searl 185 dual channel automatic gamma counter (Nuclear-Chicago, IL, U.S.A.). The withdrawn blood sample was slowly injected back into the animals immediately after counting of radioactivity.

Experimental design and protocol

Animals were randomly assigned to five groups ($n=6$ in each group). Sham-operated vehicle-treated animals (group I), in which the occluder was placed but not pulled to ligate the coronary artery, and coronary artery-ligated vehicle-treated animals (group II), received normal saline (0.037 mL kg^{-1} min^{-1}). Animals in groups III, IV and V were coronary artery-ligated and received a single dose of CGS 21680 at 0.1, 0.3 or 1.0 μg kg^{-1} min^{-1}, respectively. After the completion of surgery, each animal was allowed to stabilize for a period of 1 h while blood pressure, heart rate, +dP/dt, and left ventricular end-diastolic pressure were continuously monitored. After the 1 h stabilization period, a control measurement of cardiac output and Pmcf were made. Subsequently, animals received vehicle (0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) or CGS 21680 (0.1, 0.3 or 1.0 μg kg^{-1} min^{-1}) and the second measurements of cardiac output and Pmcf were made 15 min after the start of infusion. At the end of each experiment, animals were sacrificed and the heart and lungs were removed and weighed. The infarct area of the left ventricle was determined using the technique described by Chien *et al*. (1988). Briefly, after cutting away the atria, the ventricles were cleaned of blood and a saline-filled balloon was inserted into the left ventricle. The balloon was inflated and sealed, following which the heart was placed in 100% formalin. After 24 h, the right ventricle was trimmed away and an incision was made in the left ventricle so the tissue could be pressed flat. The circumferences of the left ventricle and the infarct area were outlined on a clear plastic sheet for both the endocardial and epicardial surfaces. The difference in weight between the two marked areas was used to determine the size of the infarct area. This was estimated as a percentage of left ventricular surface area.

Drugs

CGS 21680 was dissolved in normal saline (0.9% NaCl). It was purchased from Research Biochemical International (Natick, MA, U.S.A.).

Calculations and statistical analysis

Blood pressure (mmHg) is reported as diastolic pressure plus one third of the difference between systolic and diastolic pressures. Cardiac output (mL min^{-1}) was calculated as the rate of withdrawal of blood multiplied by total injected c.p.m. divided by c.p.m. in withdrawn blood. Arterial resistance (mmHg min mL^{-1}) was obtained by dividing blood pressure by cardiac output, and venous resistance (mmHg min mL^{-1}) was calculated as the difference of Pmcf and central venous pressure divided by cardiac output (Wang *et al*., 1995).

The data were analysed by one-way analysis of variance with repeated measure for comparison. Newman-Keuls multiple range test was used for comparison between means. A difference of $P<0.05$ was considered to be significant.

Results

Ligation of the left main coronary artery produced a mortality rate of 40% within 48 h. Thereafter, the mortality rate was 5% per month. There were no significant differences in body weight and ventricular weight indexed for body weight between sham-operated and coronary artery-ligated animals. However, lung weight indexed for body weight was significantly higher after coronary artery ligation when compared to sham-operated animals. We found that the infarct areas were not different among the various groups of animals (Table 1).

Baseline blood pressure, cardiac output and rate of rise in left ventricular pressure ($+dP/dt$) were significantly reduced in animals 8 weeks post-coronary artery ligation when compared to sham-operated animals. The range of this reduction was 28–32% for blood pressure, 30–32% for cardiac output, and 27–36% for $+dP/dt$ among the different groups of animals 8 weeks after coronary artery ligation (Table 2). Moreover, coronary artery ligation also resulted in a significant increase in Pmcf and venous resistance when compared to sham-operated animals. The magnitude of this increase ranged between 37–44% for Pmcf, and 80–113% for venous resistance. Left ventricular end-diastolic pressure was significantly elevated after coronary artery ligation when compared to sham-operated animals (Table 2).

The infusion of CGS 21680 at 0.1 μg kg^{-1} min^{-1} did not significantly affect any haemodynamic parameters in coronary artery-ligated animals when compared to vehicle treatment (Figure 1–4). However, the infusion of CGS 21680 at the two higher dose levels of 0.3 and 1.0 μg kg^{-1} min^{-1} to coronary artery-ligated animals significantly increased cardiac output by $19\pm4\%$ and $39\pm5\%$, and heart rate by $14\pm2\%$ and $15\pm1\%$, respectively, when compared to vehicle-treated coronary artery-ligated animals (Figure 1A,B). In addition, administration of CGS 21680 at both 0.3 and 1.0 μg kg^{-1} min^{-1} significantly reduced blood pressure by $12\pm2\%$ and $31\pm6\%$, and arterial resistance by $25\pm3\%$ and $50\pm4\%$, respectively, when compared to vehicle administration in coronary artery-ligated animals (Figure 2A,B). Treatment with the highest dose of CGS 21680 was able to significantly reduce Pmcf ($14\pm2\%$) (Figure 3A). However, administration of CGS 21680 both at 0.3 and 1.0 μg kg^{-1} min^{-1} significantly reduced venous

Table 1 Body weight (BW; g), wet organ weight: ventricle (VW; g kg^{-1} body weight), lungs (LW; g kg^{-1} body weight), and infarct area (IA; % of left ventricular area) of the sham-operated vehicle-treated (SV; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}), coronary artery-ligated vehicle-treated (L-V; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) and CGS 21680-treated (0.1, 0.3 and 1.0 μg kg^{-1} min^{-1}) groups

Groups	*I* *SV*	*II* *L-V*	*III* *L-CGS 21680* (0.1)	*IV* *L-CGS 21680* (0.3)	*V* *L-CGS 21680* (1.0)
BW	482±2	473±9	501±2	493±9	486±7
VW	2.8±0.1	3.1±0.1	3.0±0.1	3.3±0.2	3.0±0.1
LW	3.4±0.1	4.6±0.2*	4.5±0.2*	5.1±0.2*	4.5±0.1*
IA	–	31.7±2.0	33.6±2.2	34.4±1.8	34.1±1.5

Each value represents means±s.e.mean of six experiments. *Significantly differnt from sham-operated vehicle-treated group $P<0.05$.

Table 2 Baseline blood pressure (BP; mmHg), cardiac output (CO; mL min^{-1}), heart rate (HR; beats min^{-1}), left ventricular end-diastolic pressure (LVEDP; mmHg), mean circulatory filling pressure (Pmcf; mmHg), rate of rise of left ventricular pressure ($+dP/dt$; mmHg sec^{-1}), arterial resistance (A_R; mmHg min mL^{-1}) and venous resistance (V_R; mmHg min mL^{-1}) of the sham-operated vehicle-treated (SV; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}), coronary artery-ligated vehicle-treated (L-V; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) and CGS 21680-treated (0.1, 0.3 and 1.0 μg kg^{-1} min^{-1}) groups 8 weeks post-surgery prior to administration of vehicle or drug

Groups	*I* *SV*	*II* *L-V*	*III* *L-CGS 21680* (0.1)	*IV* *L-CGS 21680* (0.3)	*V* *L-CGS 21680* (1.0)
BP	120±5.0	82±3.0*	83±6.0*	82±5.0*	86±4.0*
CO	97±2.3	68±3.5*	65.7±3.2*	67±3.0*	68.2±3.0*
HR	385±15	382±7	370±7	368±8	358±15
[1]LVEDP	−3.5±0.8	8.2±0.9*	6.1±0.5*	6.6±0.5*	7.5±1.4*
Pmcf	5.3±0.1	7.6±0.6*	7.3±0.4*	7.3±0.4*	7.3±0.4*
$+dP/dt$	4875±56	3104±215*	3395±267*	3166±201*	3562±101*
A_R	1.25±0.07	1.21±0.05	1.27±0.07	1.21±0.05	1.26±0.06
V_R	0.030±0.001	0.054±0.005*	0.057±0.005*	0.055±0.003*	0.064±0.004*

Each value represents means±s.e.mean of six experiments. [1]Relative to baseline at atmospheric pressure. *Significantly different from sham-operated vehicle-treated group $P<0.05$.

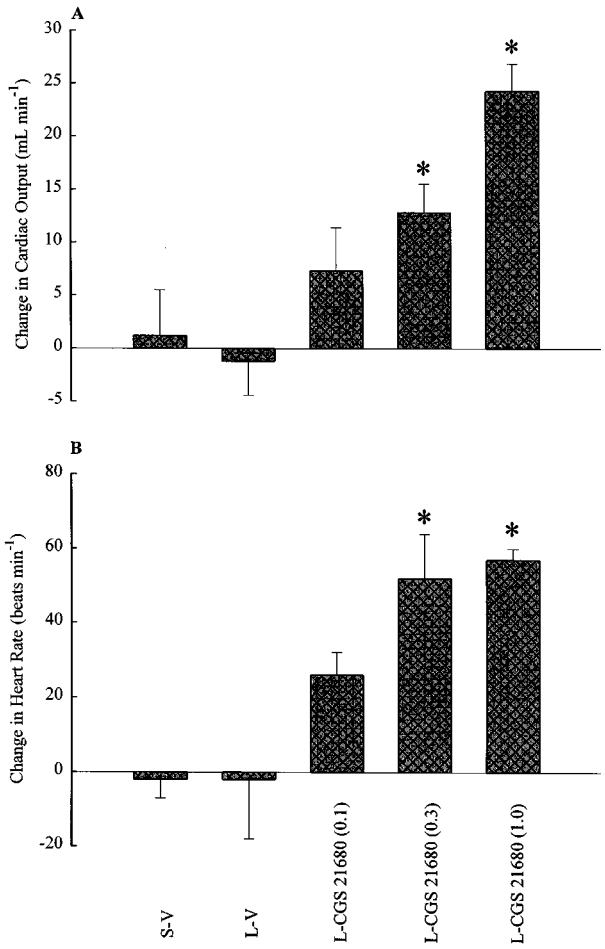

Figure 1 Changes from pre-treatment values of cardiac output (A) and heart rate (B) of the sham-operated vehicle-treated (SV; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}), coronary artery-ligated vehicle-treated (L-V; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) and CGS 21680-treated (L-CGS 21680; 0.1, 0.3 and 1.0 μg kg^{-1} min^{-1}) rats. *Significantly different from L-V group, $P<0.05$.

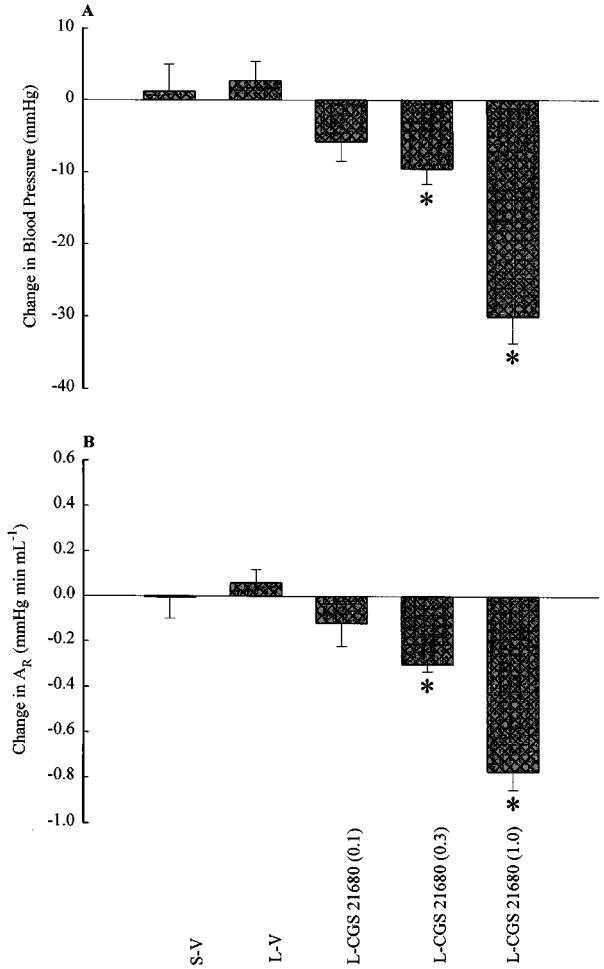

Figure 2 Changes from pre-treatment values of arterial pressure (A) and arterial resistance (A_R) (B) of the sham-operated vehicle-treated (SV; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}), coronary artery-ligated vehicle-treated (L-V; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) and CGS 21680-treated (L-CGS 21680; 0.1, 0.3 and 1.0 μg kg^{-1} min^{-1}) rats. *Significantly different from L-V group, $P<0.05$.

resistance (14±4% and 46±4%) when compared to vehicle-treated coronary artery-ligated animals (Figure 3B). The infusion of CGS 21680 both at 0.3 and 1.0 μg kg^{-1} min^{-1} was able to significantly reduce left ventricular end-diastolic pressure when compared to vehicle treatment in animals with chronic heart failure (Figure 4A). Treatment of animals with CGS 21680 did not significantly influence +dP/dt when compared to vehicle treatment (Figure 4B).

Discussion

In the present investigation, we found that ligation of the coronary artery resulted in chronic heart failure (Pfeffer *et al.*, 1979; Mulder *et al.*, 1997). Animals in which the coronary artery was ligated had lower cardiac output and blood pressure. In animals with a low cardiac output, venous tone, as measured by Pmcf, was elevated and resistance to venous return had increased. In addition, left ventricular end-diastolic pressure had increased in the coronary artery-ligated animals. In animals with chronic heart failure, infusion of CGS 21680 at the two highest doses resulted in a significant increase in cardiac output which was the result of a reduction in preload and afterload. This observation contrasts with our previous findings in animals with acute heart failure. Our previous findings indicated that CGS 21680 was only able to increase cardiac output (28%) following the administration of the highest dose.

Unlike our previous observations in anaesthetized rats in a state of acute heart failure in which the administration of CGS 21680, only at the highest dose, significantly reduced venous resistance and left ventricular end-diastolic pressure (Nekooeian & Tabrizchi, 1998), in the present investigation, administration of CGS 21680 significantly decreased venous resistance and left ventricular end-diastolic pressure at the two higher dose levels. It is recognized that a decrease in Pmcf and an increase in venous capacitance can result in a reduction of cardiac loading. This could, in part, be responsible for the decrease in left ventricular end-diastolic pressure. Certainly, this idea is supported by evidence presented in a study conducted by Raya and colleagues (1989) who demonstrated that captopril decreased Pmcf and left ventricular end-diastolic pressure in a similar model of chronic heart failure. Hydralazine, in contrast, failed to reduce either Pmcf or left ventricular end-diastolic pressure (Raya *et al.*, 1989). This lack of effect on left ventricular end-diastolic pressure is attributed to the absence of a venodilator effect by hydralazine (Raya *et al.*, 1989; D'Oyley *et al.*, 1989). Furthermore, a beneficial effect resulting from a decrease in left ventricular end-diastolic

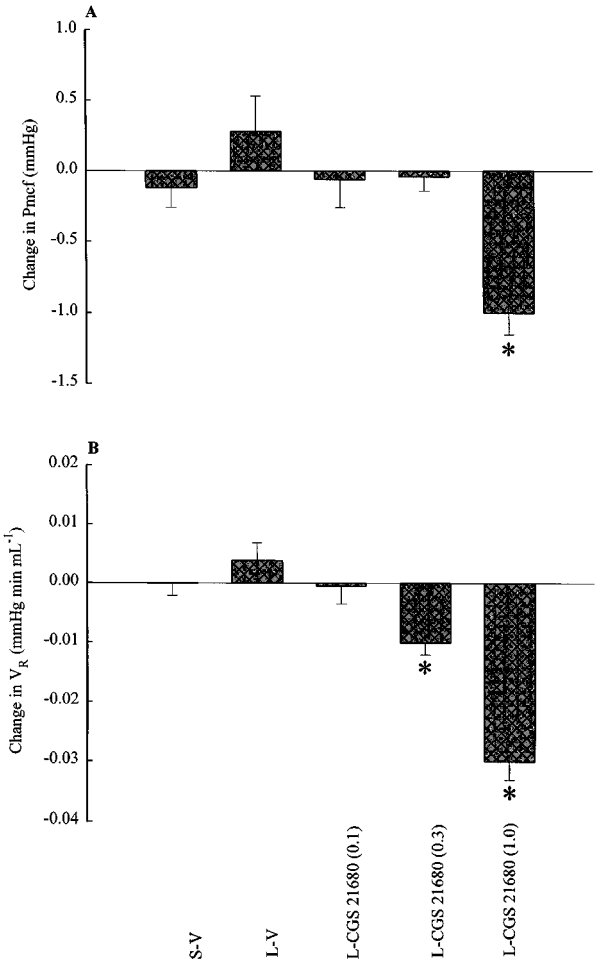

Figure 3 Changes from pre-treatment values of mean circulatory filling pressure (Pmcf) (A) and venous resistance (V_R) (B) of the sham-operated vehicle-treated (SV; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}), coronary artery-ligated vehicle-treated (L-V; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) and CGS 21680-treated (L-CGS 21680; 0.1, 0.3 and 1.0 μg kg^{-1} min^{-1}) rats. *Significantly different from L-V group, $P<0.05$.

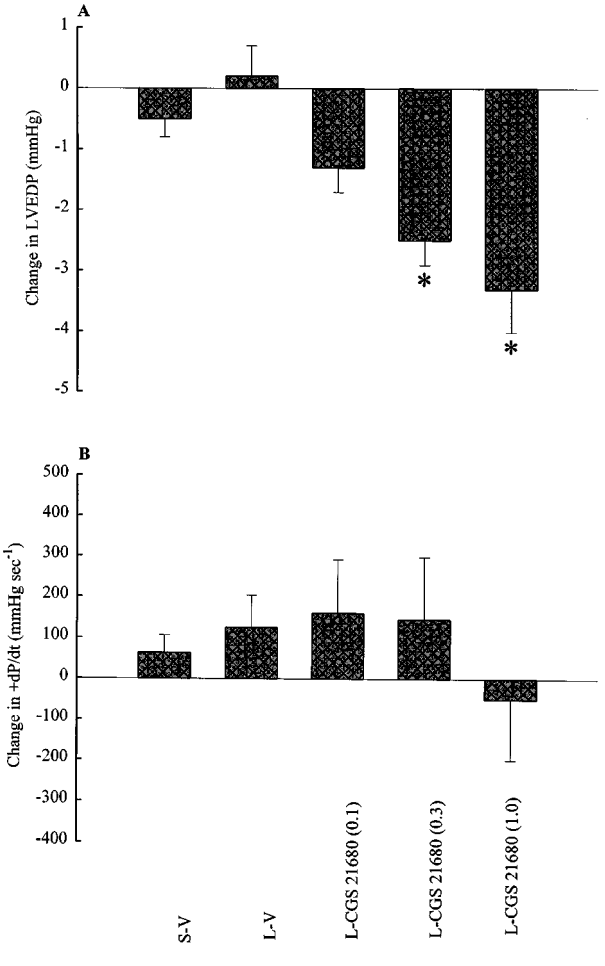

Figure 4 Changes from pre-treatment values of left ventricular end-diastolic pressure (LVEDP) (A) and rate of rise of left ventricular pressure (+dP/dt) (B), of the sham-operated vehicle-treated (SV; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}), coronary artery-ligated vehicle-treated (L-V; 0.9% NaCl; 0.037 mL kg^{-1} min^{-1}) and coronary artery-ligated CGS 21680-treated (L-CGS 21680; 0.1, 0.3 and 1.0 μg kg^{-1} min^{-1}) rats. *Significantly different from L-V group, $P<0.05$.

pressure is a decrease in wall tension development in systole and a subsequent decline in oxygen consumption by the heart (Sonnenblick & LeJemtel, 1989). In the present study, administration of CGS 21680 reduced venous resistance, and this reduction in venous resistance was responsible for the reduction in preload. A reduction in preload can certainly result in improved cardiac output in a failing heart (Vanhoutte, 1983).

It is recognized that stimulation of adenosine receptors in veins produces relaxation. Adenosine has been shown to relax noradrenaline-precontracted rings of saphenous vein of dog (Verhaeghe, 1977), portal vein of rat (Sjöberg & Wahlström, 1975), hind limb vein of dog (Cotterrell & Karim, 1982), and dorsal hand vein of human (Ford *et al*., 1992). Moreover, administration of adenosine in dogs has also been shown to result in an increase in adipose tissue volume suggesting the occurrence of dilatation of capacitance blood vessels in the adipose tissue (Sollevi & Fredholme, 1981). In addition, adenosine has been reported to reduce the body's total venous tone as assessed by measurement of Pmcf in rats (Glick *et al*., 1992; Tabrizchi, 1997). Repetitive and reproducible measurements of Pmcf, which reflect the body's total venous tone, have been made using this technique (Pang, 1994). It is evident from our present observations that CGS 21680 has venodilating properties since it decreased Pmcf and venous resistance. Furthermore, Abiru and colleagues (1995) had previously demonstrated that CGS 21680 could also produce relaxation in rat isolated femoral vein. In addition, we recently examined the effects of CGS 21680 on Pmcf and venous resistance in ganglion-blocked or intact pentobarbital-anaesthetized rats. In ganglion-blocked animals, infusion of CGS 21680 reduced Pmcf and venous resistance. In animals not treated with ganglion-blockers, CGS 21680 administration caused arterial dilatation but not venodilatation due to hypotension-induced sympathetic activation (Tabrizchi, 1997). It is evident that the influence of CGS 21680 on venous circulation in normal animals not treated with ganglion-blockers is in contrast to our observations in animals with either acute or chronic heart failure. It seems that under pathophysiological situations, such as acute heart failure, in which Pmcf and venous resistance are elevated due to activation of the sympathetic nervous system, administration of CGS 21680 reduces both Pmcf and venous resistance (Nekooeian & Tabrizchi, 1998). Moreover, in the present investigation, administration of CGS 21680 to animals with established chronic heart failure resulted in reduced Pmcf and venous resistance. Obviously, the difference between the

effects of CGS 21680 on venous circulation in normal rats to those in a state of acute or chronic heart failure relates to changes that occur within the circulation following the dysfunction of the myocardium (i.e. pump). Furthermore, the impact of acute heart failure on venous circulation is quite different to that observed following chronic heart failure. Certainly, in acute failure, there is over-activation of the sympathetic nervous system, which will increase Pmcf. This is in contrast to chronic heart failure, in which over-activation of the sympathetic nervous system and the renin-angiotensin system will result in a reduction in vascular capacitance (i.e. volume loading) with time (for review see Zelis *et al*., 1981). This has been suggested to be responsible for cardiac hypertrophy and left ventricular dilatation which occurs following chronic heart failure (for review see Zelis *et al*., 1981). Progressive cardiac failure eventually results in venous congestion.

In the present investigation, we found that CGS 21680 was able to increase cardiac output without significantly affecting +dP/dt. Therefore, it can be suggested that CGS 21680 did not appear to exert positive inotropic effects on the myocardium to increase cardiac output. We have also previously reported that administration of CGS 21680 in acute heart failure did not affect +dP/dt but resulted in an increased cardiac output (Nekooeian & Tabrizchi, 1998). Furthermore, since CGS 21680 has the ability to increase coronary artery conductance and blood flow (Nekooeian & Tabrizchi, 1996), this effect would also have been responsible for the improvement of cardiac function in chronic heart failure.

A decrease in arterial resistance can also result in increased cardiac output. A reduction in arterial resistance helps reduce impedance to left ventricular ejection and reduce afterload. Such an effect augments left ventricular function and, thus, cardiac output (Amsterdam *et al*., 1978). It is recognized that CGS 21680 is a powerful arterial dilator as evidence in the current literature indicates that CGS 21680 is capable of producing arterial smooth muscle relaxation (Webb *et al*., 1991; Abebe *et al*., 1995; Conti *et al*., 1997). In the present study, we found that administration of CGS 21680 in animals with chronic heart failure resulted in a reduction of arterial resistance. This reduction in arterial resistance would have resulted in a reduction in afterload and this may have contributed to the increase in cardiac output. We have previously reported that administration of CGS 21680 can result in a reduction in arterial resistance and improve cardiac output under conditions where arterial impedance was elevated by continuous infusion with phenylephrine (Nekooeian & Tabrizchi, 1996). However, a reduction in afterload *per se* does not simply result in an increase in cardiac output. In our previous investigation, in animals with acute heart failure, infusion of CGS 21680 at both 0.3 and 1.0 μg kg^{-1} min^{-1} resulted in a significant reduction in peripheral resistance, however, it produced a significant increase in cardiac output only at the highest dose (Nekooeian & Tabrizchi, 1998). In the present investigation, the ability of CGS 21680 to significantly increase cardiac output at 0.3 and 1.0 μg kg^{-1} min^{-1} could be related to morphological/structural changes that occur within the cardiovascular system in chronic heart failure (van Wijngaarden *et al*., 1991; Thuillez *et al*., 1995; Ganguly *et al*., 1997; Mulder *et al*., 1997).

The increase in cardiac output by administration of CGS 21680 was, in part, due to an increase in heart rate. Certainly, an increase in heart rate can be due to a direct effect on the heart, or a reflex-mediated activation of the sympathetic nervous system as a result of arterial dilatation and the lowering of blood pressure. Evidence from our laboratory (Nekooeian & Tabrizchi, 1996; Tabrizchi, 1997) and the laboratories of other investigators (Hutchison *et al*., 1989; Webb *et al*., 1991; Fozard & Carruthers, 1993; Hernandez *et al*., 1994) support the view that CGS 21680-induced tachycardia is the result of reflex-mediated activation of the sympathetic nervous system rather than a direct effect on the myocardium. However, we believe that the increase in heart rate by CGS 21680 was not the sole mechanism responsible for the increase in cardiac output during the administration of CGS 21680 in animals with chronic heart failure. Our explanation for the latter view is supported by the observation in the present study, that administration of CGS 21680 at 0.3 and 1.0 μg kg^{-1} min^{-1} increased heart rate by $14 \pm 2\%$ and $15 \pm 1\%$, respectively. However, infusion of CGS 21680 at 0.3 and 1.0 μg kg^{-1} min^{-1} increased cardiac output by $19 \pm 4\%$ and $39 \pm 5\%$, respectively. While the highest dose of CGS 21680 was able to increase cardiac output by 2 fold in comparison to the lower dose, the changes in heart rate were of similar magnitude following the administration of each dose. Therefore, we interpret this observation as indicating that the increase in heart rate was not the primary mechanism by which CGS 21680 was able to improve cardiac output in chronic heart failure.

It is apparent that an infusion of CGS 21680 at 1.0 μg kg^{-1} min^{-1} had a greater impact on cardiac output than 0.3 μg kg^{-1} min^{-1}. Our results from the present investigation indicate that only the highest administered dose of CGS 21680 was able to significantly reduce Pmcf. This reduction in Pmcf manifested itself in a substantial reduction in venous resistance. In addition, a greater reduction in arterial resistance occurred during the administration of the highest dose of CGS 21680 when compared to the second dose. We believe that this difference in the increase in cardiac output following administration of 1.0 μg kg^{-1} min^{-1} in comparison to 0.3 μg kg^{-1} min^{-1} was mainly due to a considerable reduction in impedance to flow, as well as, a reduction in resistance to venous return.

It is obvious that arterial and venous dilatation can result in increased cardiac output in conditions of cardiac failure (Vanhoutte, 1983). Arterial dilatation can result in reduced arterial resistance and thus afterload, whereas venodilatation produces a decrease in venous resistance and, therefore, a reduction in preload. More importantly, a combination of arterial and venous dilatation has been found to prolong life in patients with moderate heart failure in the VAC trial (Cohn *et al*., 1986). It is evident that CGS 21680 is capable of reducing both arterial and venous resistances in animals with chronic heart failure. The fact that this compound can reduce both arterial and venous resistances and improve cardiac output may indicate that it could be of value in the treatment of chronic heart failure in the clinical setting.

In summary, the results of the present study suggest that chronic heart failure results from occlusion of the coronary artery in the rat. Chronic heart failure is characterized by reduced arterial pressure and cardiac output, and increased left ventricular end-diastolic pressure, venous resistance and Pmcf. The administration of CGS 21680 to animals with chronic heart failure resulted in an increased cardiac output which was due to reduced venous resistance and arterial resistance, as well as, increased heart rate.

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