Basic Science and Experimental Studies

Gender Differences in Cardiac Dysfunction and Remodeling due to Volume Overload

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ABSTRACT

Background: This study examined the sex differences for hemodynamic and echocardiographic changes in hypertrophied and failing hearts induced by arteriovenous (AV) shunt.

Methods and Results: Echocardiographic and hemodynamic alterations were determined in male and female rats at 4 and 16 weeks after AV shunt. Ovariectomized females treated with estrogen for 16 weeks post-AV shunt were also used. Both genders developed cardiac hypertrophy at 4 and 16 weeks post-AV shunt; however, the increase in cardiac muscle mass was greater in females than males at 16 weeks. At 4 weeks post-AV shunt, increases in ventricular dimensions and left ventricular end-diastolic pressure (LVEDP) as well as a decrease in fractional shortening occurred in males only. Unlike the females, the rates of pressure development (+dP/dt) and decay (-dP/dt) were depressed and LVEDP increased in male rats at 16 weeks post-AV shunt. An increase in cardiac output was seen in both genders, but this was more marked in the males at 4 and 16 weeks post-AV shunt. Although mRNA levels for ACE were increased in both male and female rats at 4 and 16 weeks, mRNA levels for angiotensin II type 1 receptor were increased in males at 16 weeks only. Furthermore, increases in plasma catecholamines were elevated in males but were decreased or unchanged in females at 16 weeks of AV shunt. LV internal diameters as well as depressed fractional shortening occurred in males whereas increases in posterior wall thickness were seen in the female rats at 16 weeks of AV shunt. Ovariectomy resulted in depressed +dP/dt, -dP/dt, and fractional shortening, whereas a marked increase in cardiac output as well as increased LVEDP and LV internal diameters were observed at 16 weeks post-AV shunt. Although treatment with 17-β estradiol normalized ±dP/dt, LVEDP remained elevated.

Conclusion: Gender differences in cardiac function may be due to differences in the type of cardiac remodeling as a consequence of AV shunt. Furthermore, estrogen appears to play an important role in preventing cardiac dysfunction and adverse ventricular remodeling in female rats. (*J Cardiac Fail 2010;16:439–449*)

Key Words: Arteriovenous shunt, sex differences, ventricular remodeling, heart dysfunction, estrogen therapy.

Cardiac hypertrophy is an adaptive response of the heart to hemodynamic overload, during which terminally differentiated cardiomyocytes increase in size without undergoing cell division.[1] Although the hypertrophic response may serve to maintain cardiac function for a certain period, prolonged hypertrophy becomes detrimental, resulting in cardiac dysfunction and congestive heart failure (CHF).[1,2] Various epidemiological investigations have suggested sex differences with respect to the incidence of hypertension[3,4] coronary artery disease,[5] CHF,[6–9] and apoptosis.[10] Different experimental studies have also indicated gender differences in the development of cardiac hypertrophy from pressure overload[11,12] and myocardial infarction (MI).[13] Although cardiac remodeling in chronic volume overload has been shown to occur in the male rat,[14–22] there is

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very limited information available in the literature regarding the gender differences in the pattern of ventricular remodeling and contractile function in response to chronic volume overload. In this regard, some investigators[23] have reported that mortality in female rats is 10-fold less than in male rats after 8 weeks of volume overload. Both sexes had increases in left ventricle and right ventricle weights relative to controls, whereas lung weight was increased in males only. Although male rats had marked dilatation and increased compliance, female rats had no significant change in left ventricular (LV) dilatation or compliance. In contrast to males, female rats had no indications of CHF; however, in this study cardiac function was assessed by in vitro techniques.[23]

The contribution of ovarian hormones in the prevention of adverse ventricular remodeling because of volume overload in female animals was examined by using ovariectomized female rats.[24] It was observed that ovariectomized rats developed more extensive ventricular remodeling and a substantial increase in LV dilatation than control female rats at 21 weeks post- arteriovenous (AV) shunt. In addition, there was a marked increase in lung weight and decrease in LV function of ovariectomized rats, indicating that circulating ovarian hormones may have an influence on the pattern of myocardial remodeling in response to chronic volume overload.[24] In this study, cardiac function was also assessed by in vitro techniques and the effects of estrogen therapy on ventricular remodeling were not assessed.[24] However, later studies by this group found that estrogen treatment of ovariectomized female rats subjected to volume overload prevented pulmonary edema and attenuated LV hypertrophy and dilation, but did not maintain contractile function.[25] Moreover, these animals were studied at 8 weeks post-AV shunt and systolic and diastolic cardiac functions were assessed by using a blood-perfused, isolated heart preparation. Although these studies suggested gender-specific differences for cardiac remodeling post-fistula, echocardiographic or extensive hemodynamic assessment was not performed to characterize ventricular remodeling as well as contractile function in response to chronic volume overload. Accordingly, this investigation was undertaken to determine the pattern of ventricular remodeling and contractile function in cardiac hypertrophy at 4 weeks and CHF at 16 weeks post-AV shunt in both male and female rats. Cardiac mRNA levels for angiotensin-converting enzyme (ACE) and angiotensin II type 1 receptor (AT_1R) were also measured at 4 and 16 weeks post-AV shunt in both male and female rats to assess the status of renin-angiotensin system (RAS). In addition, the contribution of estrogen (17-β estradiol) for the prevention of adverse ventricular remodeling and cardiac dysfunction from volume overload induced by AV shunt was investigated in female rats by employing M-mode echocardiography as well as the in vivo catheterization technique for measuring the left ventricular function. Alterations in the sympathetic nervous system (SNS) from an AV shunt for 16 weeks was also monitored by measuring plasma catecholamines in male and female as well as in ovariectomized rats with or without estrogen treatment.

Materials and Methods

Experimental Model

All experimental protocols for animal studies were approved by the Animal Care Committee of the University of Manitoba following the guidelines established by the Canadian Council on Animal Care. An AV shunt was performed in male and female Sprague-Dawley rats (weighing 150 to 200 g) as previously described.[22,26] Briefly, the animals were anesthetized with 5% isoflurane with a 2 L/min flow rate of oxygen, and the abdominal laparotomy was performed. After exposure of the abdominal aorta and inferior vena cava between the renal arteries and ileac bifurcation, the descending aorta and the ileac bifurcation was temporarily occluded for 25 seconds to 1 minute proximal to the intended puncture site. An 18-gauge needle was inserted and withdrawn across the medial wall of the descending aorta three times to ensure the size and presence of the shunt and finally withdrawn. The puncture site was then sealed immediately with a drop of isocyanate (Krazy glue). The creation of the shunt was visualized by the pulsatile flow of oxygenated blood into the vena cava from the abdominal aorta. Throughout the operative procedure, which was completed within 10 minutes, rats were maintained on 2.5% isoflurane in 2 L/min of oxygen. Age-matched, sham-operated animals served as controls and were treated similarly, except that the puncture into the descending aorta was not performed. Both male and female animals were allowed to recover and maintained on food and water ad libitum for 4 or 16 weeks before use. There was no mortality in the control group, whereas the mortality of the AV shunt animals was less than 4% during the first 6 hours after surgery, but thereafter no mortality was seen in either group from the surgical procedure. In another set of experiments, ovariectomized females were purchased from the Charles River. The female Sprague-Dawley rats were ovariectomized by the supplier in 6-week-old (150 to 200 g) animals and were shipped on the fifth day after surgery. Half of these animals were operated for AV shunt and the remaining half were sham-operated; these animals in each group, received an implantable 90-day slow release 17-β estradiol pellet (1.5 mg) that delivered a daily dose of 17 μg/day to each female rat as described elsewhere.[27] All these female rats were allowed to recover and maintained on food and water ad libitum for 16 weeks. After 60 days, a second implantable 17- β estradiol pellet was inserted such that estrogen was administered for the full 16-week period.

Echocardiography of the Heart

An ultrasound imaging system (SONOS 5500, Agilent Technologies, Mississauga, ON, Canada) was used for the measurement of cardiac output, heart rate, LV wall thickness and internal diameters during systole and diastole, as well as the fractional shortening of hearts of experimental animals as previously described.[26] Echocardiography measurements were conducted in rats anaesthetized with 2.5% isoflurane in 2 L/min oxygen. Briefly, the transthoracic short axis measurements were performed using a 12 MHz annular array ultrasound transducer. The heart was first imaged in the 2-dimensional mode in the parasternal long-axis view. From this view, an M-mode cursor was positioned perpendicular to the interventricular septum and posterior wall of the LV at the level of the papillary muscle and M-mode images were obtained for wall thickness and chamber dimensions.[28] Images were stored in digital format on a magnetic optical disk for review and analysis. Several parameters such as cardiac output, fractional