The Hemodynamic Characteristics of Right Ventricle After Gradient Pulmonary Artery Banding in Rats

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Research article

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Abstract

**Background:** Right ventricular (RV) failure induced by sustained pressure overload is a major contributor to morbidity and mortality in several cardiopulmonary disorders. Reliable and reproducible animal models of RV failure are important in order to investigate disease mechanisms and effects of potential therapeutic strategies. To establish a rat model of RV failure perfectly, we observed the right ventricle and carotid artery hemodynamics characteristics in different degrees of pulmonary artery banding of rats of different body weights.

**Methods:** Rats were subjected to 6 groups—control (0%, n=5), PAB (1-30%, n=4), PAB (1-30%, n=6), PAB (61-70%, n=5), PAB (71-80%, n=4), PAB (100%, n=3). We measured the right ventricular pressure (RVP) by right heart catheterization when the pulmonary arterial was ligated.

**Results:** The RVP gradually increased with increasing degree of banding, but when occlusion level exceeding 70%, high pressure state can be only maintained for a few minutes or seconds, and then the RVP drops rapidly until it falls below the normal pressure, which in Group F particularly evident.

**Conclusions:** RVP have different reactions when the occlusion level is not the same, and the extent of more than 70% ligation is a successful model of acute right heart failure. These results may have important consequences for therapeutic strategies to prevent acute right heart failure.

**Background**

Heart failure is a common, costly, disabling and deadly disease [1]. Heart failure reduces physical and mental health of patients significantly, resulting in a markedly decreased quality of life [2, 3]. In developing and developed countries around 2% of adults suffer from heart failure, but in those over the age of 65, this increases to 6–10%[4–7]. Left heart failure has been well studied and a number of research hypotheses have been developed. However, right heart failure is characterized by increasing rates of morbidity and mortality, and the exact pathological mechanisms of right heart failure is largely unknown. Right ventricular (RV) dysfunction has been shown to be the main outcome of pulmonary hypertension and congenital heart disease.

Pulmonary arterial hypertension (PAH) affects all ages from newborns to adults. PAH leads to dysfunctions of both pulmonary vasculature and the heart. Acute PAH results in significant right ventricular failure. RV adaptation and ventricular remodeling occurs after changes in pulmonary vasculature. Right ventricle hypertrophy (RVH) follows PAH because of compensatory mechanisms to the increased afterload. However, persistent overload results in RV dysfunction and failure. Ventricular pressure-volume relationships, changes in wall thickness and geometry are included in RV remodeling. Recently, there is a variety of preclinical right heart failure models have emerged, just like these acute model (pulmonary trunk banding)[8, 9] and chronic model (monocrotaline(MCT)-induced right heart
failure) [10, 11] have contributed to an increased research for right heart failure. Unfortunately, the MCT model was major to offers several pathological mechanisms key aspects of human PAH, including vascular remodeling, proliferation of smooth muscle cells, endothelial dysfunction, and up regulation of inflammatory cytokines rather than the right ventricle failure[12]. Accordingly, it is not possible to conclude if an improvement in RV function after an intervention is secondary to afterload reducing pulmonary vascular effects or if it is caused by direct effects on the RV. At present, the better model of acute right heart failure is pulmonary arterial trunk banding which model was created by prolone suture tied tightly around a needle alongside the main pulmonary arterial, after subsequent rapid removal of the needle, a fixed constricted opening was created in the lumen equal to the diameter of the needle[9]. But there is a big problem in this way. Firstly, the diameter of the banding is not exactly the same as the outer diameter of the needle as the ligature is tied around both the needle and the pulmonary trunk. Secondly, there may be significant variation to how tightly the knot is tied making it difficult to reproduce a certain degree of banding. Thirdly, the diameters of main pulmonary trunk of rats of different body weights were significantly different according to our pre-experiment. The affection of different degree pulmonary trunk banding to right ventricle and artery hemodynamics characteristic is indefinite for this reason, whether this model can be used as an acute model of right heart failure is also unknown.

In this situation, we observed the right ventricle and carotid artery hemodynamics characteristics in different degrees of pulmonary artery banding of rats of different body weights to establish a rat model of RV failure perfectly.

**Methods**

Thirty-two 8-week-old male Sprague Dawley rats were purchased from the Animal Core Facility of Nanjing Medical University. The rats were housed in a temperature-and humidity-conditioned facility (25 ± 1 °C with the relative humidity of 40–70%) and fed ad lib with food and water. The illumination was under 12 h light-dark cycles. All experimental procedures conformed to the Guide for the Care and Use of Laboratory Animals published by the US National Institutes of Health. The study was approved by the Ethical Committee of Gansu Provincial Hospital.

A rat model of the different degrees pulmonary artery banding (PAB) was established to measure right ventricle and carotid artery pressure. Eight-week-old male Sprague-Dawley rats (200–300 g) were anesthetized with intraperitoneal pentobarbital (50 mg/kg body weight) and ventilated with air by using a volume controlled respirator (4 mL, 70 cycles/min). Rats were subjected to 6 groups according to the degree of pulmonary artery banding(Fig. 2): control(0%, n = 5)(pulmonary artery banding 0%), PAB(1–30%, n = 4)( pulmonary artery banding 1–30%), PAB(31–60%, n = 6)(pulmonary artery banding 31–60%),PAB(61–70%, n = 5)(pulmonary artery banding 61–70%),PAB(71–80%,n = 4)(pulmonary artery banding 71–80%), PAB(100%, n = 3)(pulmonary artery banding 100%).

The pulmonary arterial trunk was carefully exposed and the right ventricle was isolated through a middle thoracotomy. Right ventricular pressure was measured by Power Lab 8/35.The carotid artery was isolated
through a median line incision at the neck, then, measured the pressure by Power Lab 8/35. Five of them died of intraoperative bleeding. The procedural surgery success rate was 84.4%. The pulmonary arterial trunk was clipped by a small arterial clip (Fig. 1) when the main pulmonary arterial and carotid artery was absolute exposed and measured. The degree of pulmonary artery banding was represented as stenosis rate of cross sectional area.

Histopatholgical analysis: At the end of the study, all rats were anesthetized by an intraperitoneal injection of 50 mg/kg pentobarbital sodium, and then sacrificed by cervical dislocation. Digital recordings were obtained by a micro tip catheter installed in the RV and carotid artery pressures before euthanasia. The hearts were quickly removed, and the fresh ventricular tissues were immediately blotted dry and weighted to clarify the relationship between heart weight and pulmonary artery diameter. Tissue specimens for pathological analysis were obtained from whole hearts in cross sections, cut into 5 μm thick sections, and stained with hematoxylin and eosin for morphologic analysis, including measurement of the cross sectional area of RV and LV by Image J software.

Statistical Analysis. All data were expressed as mean ± SEM. Two-way ANOVA of Linear regression was used to compare group means. Unpaired t-test was used to item between the control and PAB groups. Probability of 0.05 or less was considered to best statistically significant.

**Results**

**Baseline characteristics of the rats included in this study**

The basal characteristics of patients and procedural features are shown in Table 1.

|                  | Control | PAB(1–30%) | PAB(31–60%) | PAB(61–70%) | PAB(71–80%) | PAB(100%) |
|------------------|---------|------------|-------------|-------------|-------------|-----------|
| N                | 5       | 4          | 6           | 5           | 4           | 3         |
| weight(g)        | 242.7 ± 22.27 | 220.48 ± 12.77 | 234.23 ± 23.4 | 225.17 ± 7.5 | 234.7 ± 12.76 | 240.77 ± 13.41 |
| Heart(g)         | 0.77 ± 0.08 | 0.70 ± 0.06 | 0.74 ± 0.06 | 0.71 ± 0.07 | 0.70 ± 0.07 | 0.73 ± 0.05 |
| PA diameter (mm) | 3.50 ± 0.52 | 3.26 ± 0.33 | 3.39 ± 0.20 | 3.52 ± 0.32 | 3.49 ± 0.49 | 3.18 ± 0.29 |

**The change of right ventricle area after gradient pulmonary artery banding**

Gross morphology is change obvious that the right ventricular cavity increased after pulmonary artery banding, just like the Fig. 2 showed. The right ventricle area was increased with the increase of ligation degree, especially in the PAB(61–70%) group, PAB(71–80%) group and PAB(100%) group, the RV/LV CSA and RV/(RV + LV) CSA was significantly higher than control (Fig. 3) (P < 0.01).
The hemodynamic characteristics of right ventricle after gradient pulmonary artery banding

As Fig. 4 showed, the RV pressure was no difference between PAB(1–30%) group and control group, but PAB(31–60%) and PAB(61–70%) group was significantly greater than control group, however the pressure of RV increased rapidly and dropped down quickly in 2 seconds when the degree of ligation was 71–100%, just like PAB(71–80%) and PAB(100%) group. While the min pressure of right ventricle was no difference between PAB(1–30%), PAB(31–60%), PAB(61–70%) and control group, but PAB(71–80%) and PAB(100%) group was significantly greater than control group (P < 0.01).

The carotid artery blood pressure was no difference between PAB(1–30%), PAB(31–60%), PAB(61–70%) and control group, however the pressure of the arterial systolic blood decreased rapidly in 2 seconds when the degree of ligation was 71–100%, just like PAB(71–80%) and PAB(100%) group(Fig. 5A) (P < 0.01). The carotid arterial diastolic blood pressure has the same performance(Fig. 5B). The mean heart rate decreased rapidly in 2 seconds when the degree of ligation was 71–100%, however, there was no difference between PAB(1–30%), PAB(31–60%), PAB(61–70%) and control(0%) group, as Fig. 5C showed.

The positive rate of rise in RV pressure (+ dp/dt) was no difference between PAB(1–30%), PAB(31–60%), PAB(61–70%) and control group after pulmonary artery banding, however the + dp/dt increased rapidly and dropped down quickly in 2 seconds when the degree of ligation was 71–100%(Fig. 6A) (P < 0.01). In the same time, The negative rate of rise in RV pressure (-dp/dt) was no difference between PAB(1–30%), PAB(31–60%), PAB(61–70%) and control group, but PAB(71–80%) and PAB(100%) group was significantly greater than control group(Fig. 6B) (P < 0.01).

Discussion

In animal models, right heart failure are difficult to be made and definition, because there were neither objective cut off values of cardiac or ventricular dysfunction nor changes in pressure, dimension, or volume that could be reliably used to identified. In our study, we observed the right ventricle and carotid artery hemodynamics characteristics in different degree of pulmonary artery banding of rats to provide the basis for the future research.

Based on our data, we successful mimicked the acute right heart failure when the pulmonary artery banding was 71–80% (right ventricular pressure rise and then rapidly decline).Data from the past studies [9, 13]suggests that the pulmonary hypertension model were made by pulmonary artery banding with a fixation degree of ligation in a certain body weight, just like the Fujimoto, Y’s study. In our experience, the diameters of main pulmonary arterial of rats of different body weights were significantly different, and the diameter of this pulmonary artery will significantly affect the degree of ligation. This approach is likely to result in non-repeatable.

In another way, a rat model of monocrotaline(MCT)-induced right ventricular failure based upon a single MCT injection (usually 60 mg/kg) applied intraperitoneally, resulting in the development of PAH after 3-4weeks[10, 14, 15]. MCT-induced right ventricular failure is similar to human PAH in terms of
hemodynamic and histopathological severity, and high mortality[16]. But there is some difference from human PAH by the presentation of an initial permeability lung edema, with early loss of the endothelial barrier and prominent inflammatory adventitial proliferation[17]. Nevertheless, MCT can cause injury in other organs such as liver and kidney[18], this model is not the perfect model to expression the progress of the right heart failure which induced by PAH.

In this paper, we demonstrated right heart failure by objective recording the hemodynamic characteristics of right ventricle in different degree of pulmonary artery banding of rats of different body weights. There were no significant difference between PAB(1–30%),PAB(31–60%) group and control group in right ventricle pressure, in contrast, the pressure of right ventricle increased rapidly and drop down quickly in 2 seconds when the degree of ligation was 71–100%. It is worth noting that, the right ventricular pressure can be stably maintained at a high level when the degree of ligation is maintained at 61–70%( Fig. 4).In human, the pressure of right ventricle increased first and then decreased when the right ventricular pressure overload[19]. The above results suggest that if we want to design a chronic right heart failure model, can not to use the PAB(1–30%) and PAB(31–60%) group, can only use the PAB(61–70%) group, if we want to design an acute right heart failure model, maybe the PAB(71–80%) group was the best choice.

**Conclusions**

The hemodynamics characteristics is different according with the different degree of pulmonary artery banding of rats of different body weights. This suggests that we should choose the appropriate of pulmonary artery banding when the model is different. In chronic model, we should use the pulmonary arterial banding61-70% group. In acute model, we should use the pulmonary arterial banding71-80% group.

**Abbreviations**

RV
Right ventricular; LV:Lift ventricular; PAB:Pulmonary arterial banding; RVP:Right ventricular pressure; PAH:Pulmonary arterial hypertension; RVH:Right ventricle hypertrophy; MCT:Monocrotaline.

**Declarations**

**Ethics approval and consent to participate**

Animal experiments were approved by Gansu Provincial Hospital. The national and institutional guidelines for the standard care of animals were followed.

**Consent for publication**

Not applicable.
Availability of data and materials

The analyzed data sets generated during the study are available from the corresponding author on reasonable request.

Competing interests

The authors declare they have no competing interest.

Funding

Not applicable.

Authors’ contributions

All authors have read and approved the final manuscript. WN and CYS designed the study; SJY, WY and SST have contributed to the data collection and analysis, manuscript preparation. SJY wrote the initial paper; WN and CYS revised the paper; CYS had primary responsibility for final content.

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Figures
Figure 1

The degree of pulmonary artery banding is performed with an adjustable arterial clamp with needles of different diameters. $a =$ diameter of needles, $b =$ diameter of pulmonary trunk. The degree of pulmonary artery banding =
| Control | PAB   |
|---------|-------|
| 0%      | 14.75%| 45.00% | 65.41% | 75.35% | 100% |

**Figure 2**

The change of right ventricle area after gradient pulmonary artery banding.
Figure 3

(A) RV/LV cross sectional area (B) RV/(RV+LV) cross sectional area. Data presented as mean ± SEM. One-way ANOVA with post hoc Bonferroni analysis. *p < 0.05, **p < 0.01, and ***p < 0.001, PAB vs Control.
Figure 4

(A) The max pressure of right ventricle after gradient pulmonary artery banding. (B) The min pressure of right ventricle after gradient pulmonary artery banding.
Figure 5

(A) The carotid arterial systolic blood pressure after gradient pulmonary artery banding. (B) The carotid arterial diastolic blood pressure after gradient pulmonary artery banding. (C) The mean heart rate after gradient pulmonary artery banding.
Figure 6

(A) The positive rate of rise in RV pressure (+dp/dt) after gradient pulmonary artery banding. (B) The negative rate of rise in RV pressure (-dp/dt) after gradient pulmonary artery banding.

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