Interactive CardioVascular and Thoracic Surgery 30 (2020) 535–537
doi:10.1093/icvts/ivz314 Advance Access publication 21 January 2020

BRIEF COMMUNICATION

Cite this article as: Tontodonati M, Ridley D, Remie R, Clements P. Improved method for the catheterization of the right ventricle in a rat model of pulmonary artery hypertension. Interact CardioVasc Thorac Surg 2020;30:535–7.

Improved method for the catheterization of the right ventricle in a rat model of pulmonary artery hypertension

Marcello Tontodonati[a,*], Debbie Ridley[b], René Remie[c] and Peter Clements [a]

[a] Comparative and Translational Sciences
[b] In Vivo Science Delivery, R&D GlaxoSmithKline, Stevenage, UK
[c] René Remie Surgical Skills Centre (RRSSC), Almere, Netherlands

* Corresponding author. GlaxoSmithKline, Park Road, Ware SG12 0DP, UK. Tel: +44-1992-502970; e-mail: marcello.2.tontodonati@gsk.com (M. Tontodonati).

Received 4 November 2019; received in revised form 28 November 2019; accepted 2 December 2019

Abstract

Ventricle catheterization in the rat is widely practiced in cardiopulmonary research. The catheters deployed are either fluid filled or solid tip pressure or pressure–volume catheters. The access to the right ventricle is through the right jugular vein, most commonly without direct visualization such as fluoroscopy. Advancement of the catheter tip is aided by visualizing the pressure signals of the monitoring/recording systems used. This approach may present challenges due to various reasons, including the stiffness of new catheters, their dimensions or anatomical changes associated with the animal disease model. In this article, we present a novel approach, which has been optimized, successfully validated surgically and adopted in current projects. It has been shown to improve both the overall quality of the signals recorded and the time to access the right ventricle, thus reducing the overall time of surgery. The method presented in this article is safe, easy to reproduce and does not require additional skills compared to a more 'standard' approach.

Keywords: Rat catheterization • Animal models • *In vivo* measurements • Right ventricle pressure • Right ventricle catheterization

INTRODUCTION

Catheterization of the heart has been widely used in research in both mouse and rat. The techniques deployed for left and right ventricular access can be performed either with an open chest approach or a closed chest approach, depending on the main purpose of the procedure to be conducted. Usually, the catheterization of ventricles is conducted as a terminal procedure under deep anaesthesia; however, multiple catheterizations in the same animal have been described [1]. To simultaneously record pressure and volume data with a single catheter, a specialized transducer needs to be used with appropriate dimensions and diameter suitable for the species of interest. At present, there is a limited range of available transducers for this technique in the rodent species. We opted to use the Millar Mikro-Tip® pressure volume loop catheters. The main advantages of this product are the very small diameter for rodent use (1 and 2 Fr) and 2 options for the electrode spacing (6 and 9 mm) which are suitable for most ventricle dimensions in these species. With such small diameters there is no provision of J or U tips on catheters, only straight tips. This is an advantage when the target point is the left ventricle accessed through the right carotid, but in our experience, this is a 'limitation' which can impact on catheter advancement when the primary location is the right ventricle, via the right anterior vena cava. The risk of damaging either the right atrium, the right ventricle and/or the anterior vena cava, is increased when a straight tip catheter is used, which occurred during one of the surgical sessions before the adoption of the methodology described in this article. We describe an improved approach to right ventricle catheterization. On this basis, we developed a solution, a guide cannula, to facilitate the insertion of the catheter in the right ventricle, enabling a reliable recording of haemodynamic data and significantly reducing surgery time. The method developed has been further enhanced at GSK, by refining the design of the guide tube and the surgical approach to access the right ventricular cavity, allowing a better performance both in the overall procedure and the quality of data recorded.

MATERIALS AND METHODS

All animal studies were ethically reviewed and carried out in accordance with Animals (Scientific Procedures) Act 1986 and the GSK Policy on the Care, Welfare and Treatment of Animals.

Two different studies were conducted: in the first study 27 male CD (SD) rats of an average body weight of 396 g and in the second study 16 male CD (SD) rats (Charles River, Margate, UK) of approximately average body weight of 445 g (at surgery) were used. In the first study the standard procedure was adopted, whilst in the second study the guide tube procedure described in this article was used. All the animals were induced with SU-5416 (a vascular endothelial grow factor receptor protein tyrosine kinase inhibitor, 20 mg/kg; Sigma-Aldrich, Dorset, UK) immediately exposed to 3 weeks of hypoxia in a hypobaric chamber followed

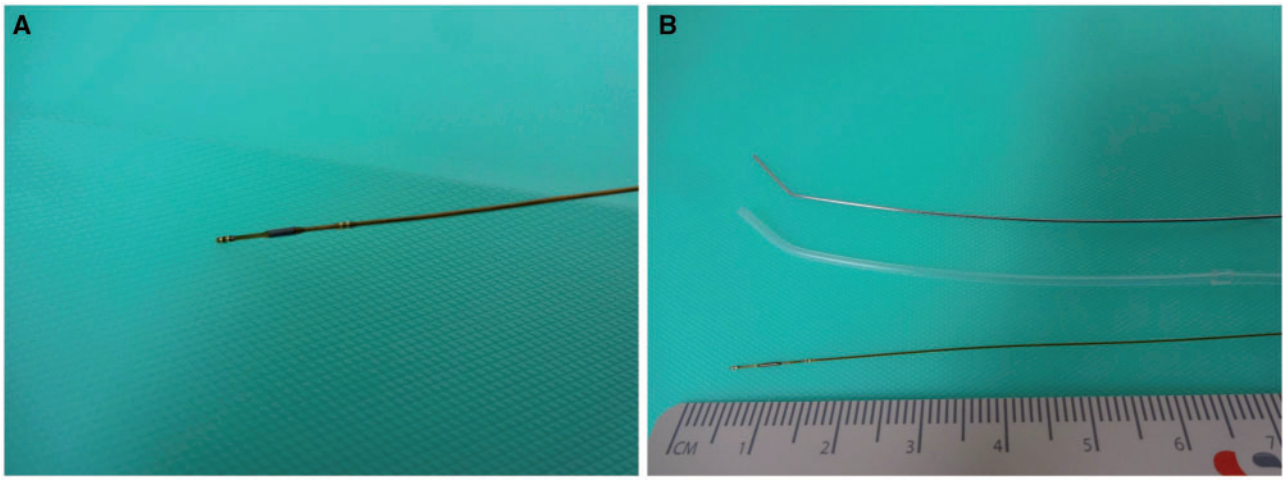

Figure 1: (**A**) 2-Fr Millar Mikro-Tip® pressure volume loop catheter with straight tip used for the procedures described in the article. (**B**) The stainless steel wire used to shape the polyethylene tube (in the middle) and the straight tip 2-Fr catheter for the pressure/volume assessment.

by 3 weeks of normoxia. This procedure is well described for the development of an irreversible pulmonary arterial hypertension and right ventricle heart failure [2]. Development and progression of the disease was monitored weekly by magnetic resonance imaging scans ($n = 8$) up to the terminal haemodynamic measurements. In both studies, rats were group-housed ($n = 4$/cage, standard house cages for rats) with environmental enrichment, according to GSK internal guidelines on animal welfare and ethics.

Mikro-Tips catheters by Millar (model SPR-838, 2 Fr with electrode spacing of 9 mm, ADInstruments, Oxford, UK) for pressure-volume loop were used under general anaesthesia at the terminal surgery, for haemodynamic assessment (Fig. 1A). One catheter was used per animal for both the right and left ventricle pressure and volume measurements and all the data were recorded on a LabChart system through a Power Lab system (ADInstruments). Animals were anaesthetized with a mixture of isoflurane (at ∼1.8%) and oxygen through a rat face mask specifically designed and 3-dimensional printed internally at GSK. At the end of the haemodynamic measurements (right and left ventricle pressure and volume) each rat was killed with an overdose of intravenous pentobarbitone administered in the caudal vena cava followed by confirmation of death by severing the major vessels (caudal vena cava and abdominal aorta).

The shape of the guide wire was obtained by using a metallic wire of adequate dimensions to fit the tubing, as a mould (Fig. 1B). The polyethylene tubing (PT51A, 0.76 mm internal diameter and 1.22 mm external diameter) with the metallic wire inserted were placed in hot water for at least 30 s to allow the polyethylene to take the required shape and cooled down immediately after in fresh cold water to retain the shape permanently. The length of the tubing was ∼59 mm and the proximal tip was bent at ∼30°. At ∼40 mm from the tip of the guide a mark was scorched to estimate the maximum depth of insertion of the catheter in the right external jugular vein. This distance was chosen based on anatomical historic data (for rats of this size) and represents the approximate distance of the right atrium from the insertion point of the catheter in the jugular vein (just caudal of the bifurcation of the right external jugular vein into the linguofacial vein and the maxillary vein). The correct position of the catheter proximal to the tricuspid valve was identified during the procedure by the evidence of pulsating blood flow in the guide tube.

The access to the right external jugular vein has been widely described elsewhere [3]. Once the right jugular vein has been exposed, the guide catheter is inserted and pushed towards the right atrium. At the first marker (∼40 mm), a slight turn counterclockwise will place the tip of the guide right in front of the tricuspid valve. The correct placement will be confirmed by observing the pulsating wave of the blood or the lock solution inside the guide tube. At this point the transducer is inserted into the guide tube and pushed to the correct location within the right ventricle and visual assessment of both the pressure and volume signals will be confirming the correct position of the tip. If the signal is not 'clean' (too much noise) the guide may be gently moved to optimize the quality of the trace to be recorded. At the end of the haemodynamic assessment of the right ventricle, gentle withdrawal of both the guide and the transducer will end the procedure.

RESULTS

This procedure has been successfully applied in all the animals included in the study and was an integral part of evaluating this pulmonary hypertension model. The quality of the signals recorded was not compromised by the presence of the guide and in all animals, we were able to record stable pressure and volume data and derive functional end points of the right ventricle (Fig. 2). Most importantly, by adopting this approach we were able to routinely use the straight tip transducers at our disposal with no damage either to the expensive catheters or to the vascular structures of the animals used in the study. An important refinement obtained adopting this approach was a significant reduction in total time of surgery. In the first of the studies described in the article using the straight tip catheter only, the median time to complete the procedure (from the start of the procedure to termination: to catheterize both right and left ventricles and obtain readings) was ∼47 min (interquartile range 30 min) from start of the procedure to termination (with a maximum time recorded of 1 h and 32 min and a minimum time of 29 min achieved only in 1 subject) according to the anaesthesia records. Using the guide in the second study, the median

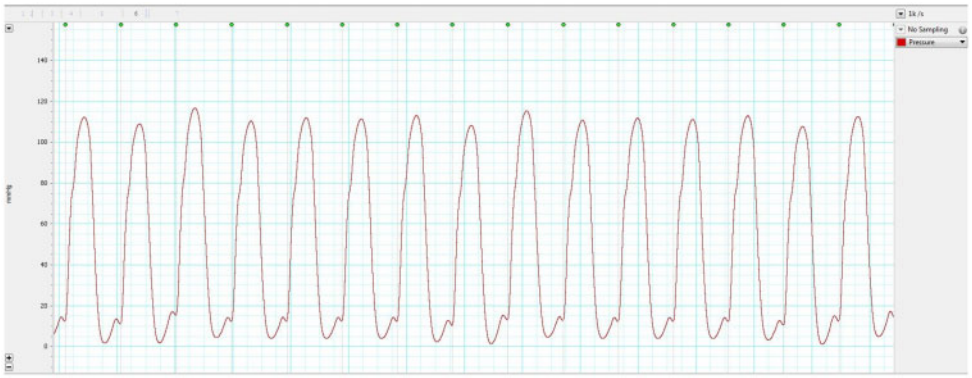

Figure 2: Sample of right ventricular pressure signal obtained using the technique described in the article.

completion time was ∼30 min (with interquartile range of 17 min) (with a maximum of 1 h and 42 min—only in a single animal—to a minimum of 15 min). Procedure times were documented in the anaesthesia records for each animal, archived at GSK. Unfortunately, we have no records of the time required to catheterize either the right ventricle or the left ventricle in isolation and are unable to determine the precise time required for each phase of the procedure. Time variability in each study is mainly due to the anatomical vascular variations frequently seen in the rat, in either the external right jugular and the right common carotid artery accesses.

DISCUSSION

Catheterization of the right ventricle in the experimental rat is not an innovative technique and has been adopted to assess the ventricular function particularly to support and study chronic pulmonary diseases as hypertension and as in our case pulmonary arterial hypertension. The challenge with this procedure is mainly associated with the current technology of the transducers when pressure/volume loop recordings are required for the haemodynamic assessment. All the current commercially available transducers for catheters up to 2 Fr of dimension have a straight tip and this may pose a risk of involuntary damage to cardiovascular structures, including perforation of the right auricle and or the right ventricle. Although sporadic and occasional, this has occurred in our laboratory where 3 rats undergoing the procedure showed pericardial effusion due to right atrium puncture at the gross macroscopic examination at the end of the procedure. In our hands, the adoption of a guide tube as described in this brief communication has proven to not only minimize the risk of incidents, but also facilitate the access to the right ventricle with minimal effort and significantly improve the time for surgery. An important consideration is related to the refinement of the surgical technique which allows a significant reduction in time spent on each animal, with important welfare and ethical implications. In particular, by minimizing the time of the surgical procedure, there is lesser impact on the cardiovascular function due to the anaesthesia, and therefore more reliable readouts. The procedure adopted in our case was performed under non-recovery anaesthesia. However it is our belief that this approach may also be used in longitudinal studies which may require a serial functional assessment of the right ventricular function [4].

Conflict of interest: none declared.

REFERENCES

[1] Migneco F, Huang Y-C, Coyan G, Birla RK. New and simplified method for multiple left ventricle catheterizations in small animals. Interact CardioVasc Thorac Surg 2008;7:925–7.

[2] Kohtaro A, Toba M, Alzoubi A, Ito M, Fagan KA, Cool CD *et al.* Formation of Plexiform lesions in experimental severe pulmonary arterial hypertension. Circulation 2010;121:2747–54.

[3] Zimmer HG, Zierhut W, Seesko RC, Varekamp AE. Right heart catheterization in rats with pulmonary hypertension and right ventricular hypertrophy. Basic Res Cardiol 1988;83:48–57.

[4] Feng J, Fitz Y, Li Y, Fernandez M, Cortes Puch I, Wang D *et al.* Catheterization of the carotid artery and jugular vein to perform hemodynamic measures, infusions and blood sampling in a conscious rat model. J Vis Exp 2015:51881.