Original Article

Title: Positive intrapleural pressure with carbon dioxide may limit intraoperative pulmonary arterial bleeding: verification by animal model

Running title: Positive pressure for pulmonary arterial bleeding

Authors: Momoko Asami, MD[1*], Eiichi Kanai, BVSc, PhD[2*], Yoshikane Yamauchi, MD, PhD[1], Yuichi Saito, MD, PhD[1], Noriyuki Matsutani, PhD[3], Masafumi Kawamura, MD, PhD[1], Yukinori Sakao, MD, PhD[1]

*Momoko Asami and Eiichi Kanai contributed equally to this work.

Institutions:

[1]Department of Surgery, Teikyo University School of Medicine, Tokyo, Japan

[2]Laboratory of Small Animal Surgery, Azabu University School of Veterinary Medicine, Tokyo, Japan

[3]Department of Surgery, Teikyo University Mizonokuchi Hospital, Tokyo, Japan

Corresponding Author:

Yukinori Sakao, MD, PhD, Department of Surgery, Teikyo University School of Medicine, 2-11-1 Kaga, Itabashi, Tokyo 173-8605, Japan. Tel: +81 3 3964 1211, Fax: +81 3 3964 6888, Email: ysakao070@gmail.com

Key words: thoracoscopic surgery, intraoperative bleeding, carbon dioxide insufflation, intrapleural pressure

Meeting presentation: None

Word count: 4329 words

Abstract

Purpose: Intraoperative complications, especially unexpected bleeding, are of great concern in the safety of thoracoscopic surgery. We investigated the hemostatic efficacy and safety of positive intrapleural pressure with carbon dioxide insufflation by assessing the amount of blood loss in a pulmonary arterial hemorrhage model.

Methods: An ex vivo experimental model of saline flow into a swine vessel was created in a container simulating a chest cavity. From the results, in vivo experiments (swine model) were conducted to compare the bleeding volume from the pulmonary artery while applying positive intrapleural pressure.

Results: In the ex vivo experiment, regardless of the incision type, the outflow volumes did not significantly differ at flow pressures of 20, 30, and 40 mmHg. At each flow pressure, the outflow volumes at 10, 15, and 20 mmHg of positive pressure in the container were significantly smaller than those of the control. Similarly, the in vivo experiments showed that bleeding decreased as intrapleural pressure increased.

Conclusion: It may be possible to temporarily suppress bleeding from the pulmonary artery by increasing the intrapleural pressure to 10 to 20 mmHg using carbon dioxide insufflation. This method may be an adjunctive hemostatic maneuver for intraoperative bleeding.

Introduction

In recent years, the use of positive intrapleural pressure (PIP) with carbon dioxide (CO_2) has increased in video-assisted and robot-assisted thoracoscopic surgery for the purpose of obtaining a good view and large working field in the thoracic cavity. We speculated that using PIP during bleeding events as described above can temporarily control the bleeding speed and ultimately reduce the occurrence of fatal complications. We previously published in vivo data from swine that underwent CO_2 insufflation(1) and showed that CO_2 insufflation can be safely introduced intraoperatively. Additionally, PIP suppressed bleeding from the injured site of the pulmonary vein with no fatal changes, such as air embolisms, accompanying the PIP in the systemic condition. We also confirmed in another clinical study that PIP of up to 15 mmHg can be safely applied intraoperatively in humans(2). However, the efficacy of PIP in treating pulmonary artery (PA) injury must be evaluated because PA laceration is a common cause of hemorrhage and is difficult to treat, as described in previous reports(3,4). Because the PA is also a low-pressure circulatory system, PIP is expected to have a certain bleeding control effect. Therefore, we conducted this study to investigate the suppressive effect of PIP on bleeding from the PA.

In the field of general thoracic surgery, minimally invasive techniques such as video-assisted and robot-assisted thoracoscopic surgery are gaining popularity because of the increased detection rate of early-stage lung tumors. These techniques allow surgeons to remove the lesions with decreased postoperative pain, a shorter air leak duration, shorter hospital stays, and lower overall complication rates with oncological results equivalent to those of traditional open thoracotomy(5–7). However, the safety of thoracoscopic surgery has caused great concern because of reports of devastating intraoperative complications, especially bleeding(3). A retrospective analysis of 8,563 thoracoscopic lung surgeries listed in the United States national database in 2016 revealed that 9.2% of patients required blood transfusions for management of intraoperative bleeding(8). Specifically, bleeding from the PA is also problematic because of secondary injury associated with surgical hemostasis. Several reports have described how to manage such severe intraoperative bleeding(4,9–11), and use of these techniques is very important. However, if the bleeding from the laceration site remains excessive, the probability of stopping it decreases, even with these techniques. Therefore, devices that will reduce the bleeding flow rate, even temporarily, are very useful.

Because proper use of laboratory animals is required to ensure animal welfare, we first collected detailed data from an ex vivo model using swine PAs immediately after slaughter. Subsequently, to validate these results, we applied the same procedure to in vivo models using a small number of experimental animals.

Materials and Methods

Ex vivo experiment

Figure 1a illustrates the ex vivo experiment. The right PA from the lung of a swine immediately after slaughter was harvested and cut to a length of approximately 5 cm. A polypropylene container resembling a chest cavity was prepared, with a hole on one side and attachment of a single-use retractor (Alnote®-LAPSINGLE; Alfresa Pharma Corporation, Osaka, Japan). The two ends of the harvested PA were connected to the infusion route in a leak-proof manner, then led out of the container through a port. One side was connected to an infusion bag containing saline solution. On the other side, the infusion route was raised to the same height as the infusion bag. Saline solution was dripped from a height equivalent to the corresponding pressure. Measurements were taken when the water level in the opposite infusion route increased to the same level as that in the infusion bag, indicating that the designated pressure had been reached in the artery. First, different injury types, including 10-mm-long incisions and 5-Fr sheath placement, were prepared in the vessel. The amount of liquid outflow from the injury site was measured for 30 seconds under various conditions, including the inflow pressure and the pressure inside the container, using an AirSeal® Intelligent Flow System (CONMED, Utica, NY, USA). No difference was found between the 10-mm incision and the 5-Fr sheath model as a PA injury model; therefore, the 5-Fr sheath model was used in the experiment with positive pressure in the container because of the reproducibility, measurement accuracy, and ease of the experiment.

In vivo experiments

All animals have received humane care in compliance with the “Principles of Laboratory Animal Care” formulated by the National Society for Medical Research and the “Guide for the Care and Use of Laboratory Animals” prepared by the Institute of Laboratory Animal Research (ILAR), published by the National Academies Press(12). The Azabu University Animal Experimentation Committee approved the in vivo experiments (approval number 200206-1), which were performed in accordance with institutional guidelines and with the National Institutes of Health guidelines regarding the principles of animal care. Two specific-pathogen-free, 50-day-old female swine weighing 32 to 35 kg each were fed a standard diet and allowed water ad libitum. Both swine underwent thoracoscopic surgery with target intrapleural pressures with CO_2 insufflation. Anesthesia was induced via intravenous injection of ketamine (10–15 mg/kg), xylazine (2 mg/kg), and propofol (2.5–3.5 mg/kg), and the animals were intubated with a 5.5-mm flexible silicone endotracheal tube (Univent; Fuji Systems Corporation, Tokyo, Japan) connected to a mechanical ventilator. The animals inhaled 2% isoflurane/100% oxygen for the entire experiment. We cannulated the left femoral artery and vein with a 20-gauge needle intravascular catheter using a cut-down technique and monitored the central venous pressure (CVP). Biological parameters, including blood pressure from the left femoral artery and left main PA, were monitored and recorded during the operation. The swine were placed in the left decubitus position (Fig. 1b). The single-use retractor (Alnote®-LAPSINGLE) was then placed through the incision in the fourth intercostal space at the anterior axillary line. A 10-mm-diameter 30-degree rigid scope was introduced through this incision. One of the four ports was connected to an AirSeal® for CO_2 insufflation, and artificial pneumothorax was maintained at a designated pressure. All surgical procedures were performed thoracoscopically. A 5-Fr intravascular catheter was inserted into the right pulmonary trunk (Fig. 1c). The intrapleural pressures varied between 0, 5, 10, 15, and 20 mmHg, and the amount of bleeding was measured for 10 seconds after applying pressure. The blood pressure of the left femoral artery, CVP, and heart rate were monitored throughout the experiment. The PA pressure before bleeding at each intrathoracic pressure (IP) was measured. However, it was technically impossible to evaluate the PA pressure at the end of bleeding in order to check the amount of bleeding.

Statistical analysis

GraphPad Prism, version 9.3.1 (GraphPad Prism Software Inc., San Diego, CA, USA) was used for the statistical analyses and to construct the figures. The results were assessed using the Kruskal–Wallis test and simple linear regression analysis to compare multiple groups and the unpaired two-tailed Student's t-test with Welch's correction (Welch's t-test) to compare two selected groups. A p value of <0.05 was considered statistically significant.

Results

Ex vivo experiments with two different vascular injury models (5-Fr sheath insertion, 10-mm incision)

First, the ex vivo model was used to compare the outflow at different flow pressures for various wound types. Two groups were created: one with a 10-mm-long wound in the blood vessel (10-mm group), and one with 5-Fr sheath placement (5-Fr group). Figure 2a shows the results of the outflow volume comparison for six samples per group. For both wound types, the outflow volume increased as the inflow pressure increased. Kruskal–Wallis nonparametric analysis showed significant differences between the two groups at outflow pressures of 10 and 50 mmHg but no significant differences between the two groups at outflow pressures of 20, 30, or 40 mmHg (Table 1). The results showed that 5-Fr sheath placement, which was easily applied during the experiment in the container, could be used as the representative wound type to measure the outflow volume at these outflow pressures.

Effect of pressure changes in the cabinet on vascular outflow in a series of 5-Fr sheath models

We then varied the pressure in the cabinet to 5, 10, 15, and 20 mmHg (PIP model) in the same experimental system with 5-Fr sheath placement and compared the outflow volumes. Table 2 and Figure 2b show the results of the experiments performed by changing the flow pressures to 20, 30, and 40 mmHg. For all flow pressures, the outflow decreased as the container pressure increased, and the F-test showed the validity of the model's approximation. Welch's t-test showed that the outflow volumes at 10, 15, and 20 mmHg of container pressure were significantly smaller than that of the control ($p = 0.027$, $p = 0.002$, and $p = 0.005$, respectively).

In vivo experiment with 5-Fr sheath model

To verify the results of the ex vivo experiment, we conducted in vivo experiments on two swine. Figure 3a shows the results of the bleeding volume measured by varying the intrapleural pressures between +5 mmHg and +20 mmHg. Although the results could not be fully validated because we used only two swine, simple linear regression analysis confirmed that the bleeding volume decreased in accordance with the increased intrapleural pressure (slope = −0.22, $F = 55.13$, $p < 0.0001$) (Fig. 3a). Additionally, recorded video confirmed that the momentum of bleeding was suppressed as the intrapleural pressure increased (supplementary video 1). No significant changes in arterial blood pressure, CVP, or heart rate were observed between the pre- and post-experiments. As the intrapleural pressure increased, the blood pressure decreased, and the CVP tended to increase in both the pre- and post-experiments (Fig. 3b and c, respectively). The mean PA pressure (average of two trials) before bleeding was 26 mmHg at an IP of 0 mmHg, 28 mmHg at an IP of 5 mmHg, 29 mmHg at an IP of 10 mmHg, 30 mmHg at an IP of 15 mmHg, and 24 mmHg at an IP of 20 mmHg.

Discussion

This study demonstrated that PIP with CO_2 insufflation can effectively control PA injury-induced bleeding during thoracoscopic surgery. We found that increasing the intrapleural pressure to >15 mmHg should reduce the amount of bleeding by less than half compared with that of surgery without CO_2 insufflation. Moreover, the in vivo experimental results suggest that the mechanism of suppressing the bleeding volume involves both applying CO_2 pressure to the bleeding point and decreasing the cardiac output. To our knowledge, this is the first study to demonstrate that PIP may help control intraoperative bleeding from the PA.

To achieve reliable hemostasis, it is necessary to identify the bleeding point. However, the bleeding point is difficult to identify in the presence of excessive bleeding. In addition, the procedure used to identify the bleeding point may increase the amount of bleeding. Therefore, additional surgical procedures (e.g., securing the central side of the blood vessel) are performed to avoid increased bleeding when checking for bleeding points. However, performing these procedures under compression hemostasis often carries additional risks. Conversely, PIP can reduce the bleeding volume without additional surgical intervention and can help identify bleeding points. In other words, it may be regarded as a safer hemostasis procedure in the event of unexpected intraoperative PA bleeding.

The growing demand for minimally invasive surgery has led to widespread use of thoracoscopic surgery without definitive evidence. The expert opinions of many medical professionals on how to handle bleeding during surgery have been summarized(13). This expert consensus clarified the following points. First, in the event of bleeding, surgeons must remain calm and use compression as the first step. Next, surgeons should convert the procedure to thoracotomy when the laceration site is large, bleeding is poorly controlled, no endoscopic view is available, or the laceration site has spread during repair. This expert consensus did not mention PIP with CO_2 insufflation. Our results suggest that additional application of PIP will temporarily suppress bleeding, making it easier to control bleeding by compression and thoracoscopically repair the laceration. Furthermore, we believe that even if surgeons convert the procedure to thoracotomy, reduced bleeding during the thoracotomy should reduce the surgical invasiveness and thus contribute to the safety of the thoracoscopic surgery.

Here, the question regarding whether it is safe to introduce high intrapleural pressure during surgery may be raised. Two safety concerns may arise: the effect of PIP on cardiopulmonary function and the occurrence of an air embolism associated with CO_2 insufflation. The former has been difficult to evaluate in experimental animals. Scholars have concluded that PIP should not be applied because destabilization of cardiopulmonary function has been demonstrated in dogs(14), swine(15), and horses(16). Conversely, studies have shown that gradually increasing the intrapleural pressure up to 14 mmHg has little effect on cardiopulmonary function in humans(17,18). We used CO_2 insufflation in a previous study and found it to be stable up to 15 mmHg(2). Thereafter, in our clinical practice, when a procedure requires applying PIP during general thoracic surgery, we set it at 10 to 15 mmHg using an AirSeal® Intelligent Flow System, and patients' vital signs remain stable for several hours. This difference between humans and non-humans may be associated with the availability of isolated lung ventilation with a double-lumen tube because positive thoracic pressure has a minimal direct effect on ventilation.

We found no clear reports of air embolisms caused by the use of thoracic CO_2 insufflation. However, various studies on air embolisms due to CO_2 insufflation were conducted in the field of laparoscopic surgery in the 1990s and 2000s, and we believe that these studies will help in evaluating the risks during thoracoscopic surgery. A meta-analysis showed that air embolisms occurred in only 7 of 489,335 laparoscopic procedures (0.001%)(19). However, using transesophageal echocardiography to monitor for CO_2 embolism during laparoscopic surgery enabled the more frequent detection of bubbles(20). Dion et al.(21) reported that a single 15-mL dose to the vena cava in dogs resulted in no intravascular bubbles, a single 100-mL dose resulted in only increased PA pressure, and a single 300-mL dose resulted in bubbles in the left ventricle and death. Mayer et al.(22) performed an in vivo experiment in which CO_2 was infused into the inferior vena cava at various rates for 2 hours in swine, resulting in air embolism and death in three of five animals after more than 50 minutes of infusion at 1.2 mL/kg/min. Graff et al.(23) concluded from experiments on dogs that the 50%

lethal dose for transvenous CO_2 infusion in 70-kg humans is 1750 mL. These results suggest that a rapid and massive influx of CO_2 is required to develop air embolisms with clinical symptoms because CO_2 is highly soluble in water. Because one death due to air embolization caused by artificial pneumothorax using non-CO_2 air has been documented(24), a certain amount of gas entry should be expected during general thoracic surgery. When CO_2 is applied for insufflation, symptomatic air embolism is unlikely to occur because of the high solubility of CO_2 in water.

This study had some limitations. First, because of the small number of swine in both the in vivo and ex vivo experiments, especially in the in vivo experiments, the statistical data may be unreliable. However, the ultimate goal of this study was to establish a method for using CO_2 insufflation to safely stop intraoperative bleeding in humans. Therefore, we believe that a porcine experimental model should be established as soon as possible and that a small number of samples would provide a sufficient bridge to clinical practice. Moreover, constructing in vivo models would require sacrificing many animals, which should be avoided as much as possible to promote animal welfare. Second, anatomical differences exist between swine lungs and human lungs. However, obtaining the necessary length of human vessel to conduct the experiment is difficult; therefore, we chose swine as the experimental models.

Conclusion

Ex vivo and swine in vivo experiments showed that increasing the intrapleural pressure (10–20 mmHg) via CO_2 insufflation may temporarily suppress bleeding from PA injury.

This method may be useful as an adjunctive hemostatic maneuver for intraoperative bleeding if it is limited to bleeding from low-pressure systems such as the venous and pulmonary circulatory systems. Further investigation is needed to clarify the safety and efficacy of this method in clinical practice.

Acknowledgments

We thank the students of Azabu University for their technical assistance with the swine anesthesia in the in vivo experiments. We thank Dr. Shuji Ando for advising us on the statistical analysis and Dr. Fumi Yokote for greatly contributing to the figure production. We also thank Traci Raley, MS, ELS and Angela Morben, DVM, ELS, from Edanz (https://jp.edanz.com/ac), for editing a draft of this manuscript.

Author contributions statement

Conceptualization: MA, MK, YukSak

Data curation: MA, ST, NM, EK, YukSak

Formal analysis: YoYa, YukSak

Funding acquisition: EK, MK, YukSak

Investigation: MA, TY, YukSak

Methodology: MA, EK, YukSak

Project administration: MK, YukSak

Resources: YaYa, NM, EK, MK, YukSak

Software: YoYa

Supervision: YuiSai, MK

Validation: YuiSai, MK

Visualization: MA, YoYa, YukSak

Writing – original draft: MA, YoYa, YukSak

Writing – review & editing: NM, EK, YuiSai, MK

Funding

A portion of the scholarship donations provided to Dr. Kanai and Dr. Kawamura was used for this study. None of the sponsors were involved in the study design; data collection, analysis, or interpretation; or writing or publishing of this manuscript.

Disclosure statement

All authors have no conflicts of interest.

References

1. Okamura R, Takahashi Y, Dejima H, Nakayama T, Uehara H, Matsutani N, et al. Efficacy and hemodynamic response of pleural carbon dioxide insufflation during thoracoscopic surgery in a swine vessel injury model. Surg Today. 2016 Dec;46(12):1464–70.
2. Yokote F, Yamauchi Y, Uehara H, Shirai S, Yasuda A, Saito Y, et al. Intrathoracic use of a small ultrasonic probe for localizing small lung tumors in thoracoscopic surgery: Empirical results and comparison with preoperative CT images. Gen Thorac Cardiovasc Surg. 2021 Mar 20;69(3):516–24.
3. Flores RM, Ihekweazu U, Dycoco J, Rizk NP, Rusch VW, Bains MS, et al. Video-assisted thoracoscopic surgery (VATS) lobectomy: Catastrophic intraoperative complications. J Thorac Cardiovasc Surg. 2011 Dec;142(6):1412–7.
4. Miyazaki T, Yamasaki N, Tsuchiya T, Matsumoto K, Hatachi G, Kitamura Y, et al. Management of unexpected intraoperative bleeding during thoracoscopic pulmonary resection: a single institutional experience. Surg Today. 2016 Aug 28;46(8):901–7.
5. Oh DS, Reddy RM, Gorrepati ML, Mehendale S, Reed MF. Robotic-Assisted, Video-Assisted Thoracoscopic and Open Lobectomy: Propensity-Matched Analysis of Recent Premier Data. Ann Thorac Surg. 2017 Nov;104(5):1733–40.
6. Kent M, Wang T, Whyte R, Curran T, Flores R, Gangadharan S. Open, video-assisted thoracic surgery, and robotic lobectomy: Review of a national database. Ann Thorac Surg. 2014 Jan;97(1):236–44.
7. McKenna RJ, Houck W, Fuller CB. Video-assisted thoracic surgery lobectomy:

experience with 1,100 cases. Ann Thorac Surg. 2006 Feb;81(2):421–5; discussion 425-6.

8. Ghosh SK, Roy S, Daskiran M, Yoo A, Li G, Fegelman EJ. The clinical and economic burden of significant bleeding during lung resection surgery: A retrospective matched cohort analysis of real-world data. J Med Econ. 2016 Nov 1;19(11):1081–6.

9. Yamashita S, Tokuishi K, Moroga T, Abe S, Yamamoto K, Miyahara S, et al. Totally thoracoscopic surgery and troubleshooting for bleeding in non-small cell lung cancer. Ann Thorac Surg. 2013 Mar;95(3):994–9.

10. Mei J, Pu Q, Liao H, Ma L, Zhu Y, Liu L. A novel method for troubleshooting vascular injury during anatomic thoracoscopic pulmonary resection without conversion to thoracotomy. Surg Endosc. 2013 Feb 18;27(2):530–7.

11. Kohno T. Management of complications in thoracoscopic surgery. J Thorac Dis. 2018 Jun;10(S14):S1620–3.

12. Guide for the Care and Use of Laboratory Animals. Guide for the Care and Use of Laboratory Animals. Washington, D.C.: National Academies Press; 2011.

13. Liu L, Mei J, He J, Demmy TL, Gao S, Li S, et al. International expert consensus on the management of bleeding during VATS lung surgery. Ann Transl Med. 2019 Dec;7(23):712–712.

14. Polis I, Gasthuys F, Gielen I, Van Ryssen B, Van Bree H, Laevens H, et al. The Effects of Intrathoracic Pressure During Continuous Two-Lung Ventilation for Thoracoscopy on the Cardiorespiratory Parameters in Sevoflurane Anaesthetized Dogs. J Vet Med Ser A. 2002 Apr;49(3):113–20.

15. Jones DR, Graeber GM, Tanguilig GG, Hobbs G, Murray GF. Effects of

insufflation on hemodynamics during thoracoscopy. Ann Thorac Surg. 1993;55(6):1379–82.

16. Bohaychuk-Preuss KS, Carrozzo MV, Duke-Novakovski T. Cardiopulmonary effects of pleural insufflation with CO2 during two-lung ventilation in dorsally recumbent anesthetized horses. Vet Anaesth Analg. 2017 May;44(3):483–91.

17. Ohtsuka T, Nakajima J, Kotsuka Y, Takamoto S. Hemodynamic responses to intrapleural insufflation with hemipulmonary collapse. Surg Endosc. 2001 Nov;15(11):1327–30.

18. Wolfer RS, Krasna MJ, Hasnain JU, McLaughlin JS. Hemodynamic effects of carbon dioxide insufflation during thoracoscopy. Ann Thorac Surg. 1994;58(2):404–8.

19. Bonjer HJ, Hazebroek EJ, Kazemier G, Giuffrida MC, Meijer WS, Lange JF. Open versus closed establishment of pneumoperitoneum in laparoscopic surgery. Br J Surg. 1997 May;84(5):599–602.

20. Derouin M, Couture P, Boudreault D, Girard D, Gravel D. Detection of Gas Embolism by Transesophageal Echocardiography During Laparoscopic Cholecystectomy. Anesth Analg. 1996 Jan;82(1):119–24.

21. Dion YM, Lévesque C, Doillon CJ. Experimental carbon dioxide pulmonary embolization after vena cava laceration under pneumoperitoneum. Surg Endosc. 1995 Oct;9(10):1065–9.

22. Mayer KL, Ho HS, Mathiesen KA, Wolfe BM. Cardiopulmonary responses to experimental venous carbon dioxide embolism. Surg Endosc. 1998 Aug;12(8):1025–30.

23. Graff TD, Arbegast NR, Phillips OC, Harris LC, Frazier TM. Gas embolism: A

comparative study of air and carbon dioxide as embolic agents in the systemic venous system. Am J Obstet Gynecol. 1959 Aug;78(2):259–65.

24. Wan Y-Y, Zhai C-C, Lin X-S, Yao Z-H, Liu Q-H, Zhu L, et al. Safety and complications of medical thoracoscopy in the management of pleural diseases. BMC Pulm Med. 2019 Jul 10;19(1):125.

Figure legends

Fig. 1 Operative procedures. (a) Ex vivo experimental setting. The right pulmonary artery (PA) from the lung of a swine was harvested. In a polypropylene container, the two ends of the harvested PA were connected to the infusion route in a leak-proof manner. One side was connected to an infusion bag containing saline solution, and on the other side, the infusion route was raised to the same height as that of the infusion bag. Saline solution was dripped from a height equivalent to the corresponding pressure. (b) In vivo experimental setting. The swine were placed in the left decubitus position. A single-use retractor (Alnote®-LAPSINGLE) was placed through the incision in the fourth intercostal space at the anterior axillary line. A 10-mm-diameter 30-degree rigid scope was introduced through this incision. One of the four ports was connected to an AirSeal® for CO_2 insufflation, and artificial pneumothorax was maintained at a designated pressure. (c) Intrathoracic view of the in vivo experiments. A 5-Fr intravascular catheter was inserted in the right pulmonary trunk. The intrapleural pressure was varied, and the amount of bleeding was measured.

Fig. 2 Ex vivo experimental results. (a) Comparison of the outflow volumes for the two groups: the 10-mm group (10-mm-long incision) and the 5-Fr group (5-Fr sheath). For both wound types, the outflow volume increased with the inflow pressure. There was no significant difference between the two groups at outflow pressures of 20, 30, or 40 mmHg. (b) Experimental results of 5-Fr model after varying the flow pressure to 20, 30, and 40 mmHg. For all fluid pressures, the outflow decreased as the container pressure increased.

Fig. 3 In vivo experimental results of 5-Fr sheath model. (a) Bleeding volumes measured by varying the intrapleural pressure between 5 mmHg and 20 mmHg. The blue line shows a simple linear regression model. The bleeding volume decreased as the intrapleural pressure increased. (b) Arterial blood pressures measured before and after the experiment while varying the intrapleural pressure between 5 mmHg and 20 mmHg. (c) Central venous pressures measured before and after the experiment while varying the intrapleural pressure between 5 mmHg and 20 mmHg.

Supplementary video 1. This is a demonstration video in which positive intrapleural pressure was applied with carbon dioxide to reduce pulmonary artery bleeding after removal of the 5-Fr sheath in a swine model. The amount of bleeding decreased as the positive pressure increased (the intrapleural pressure varied from 5 to 20 mmHg).

Figure 1

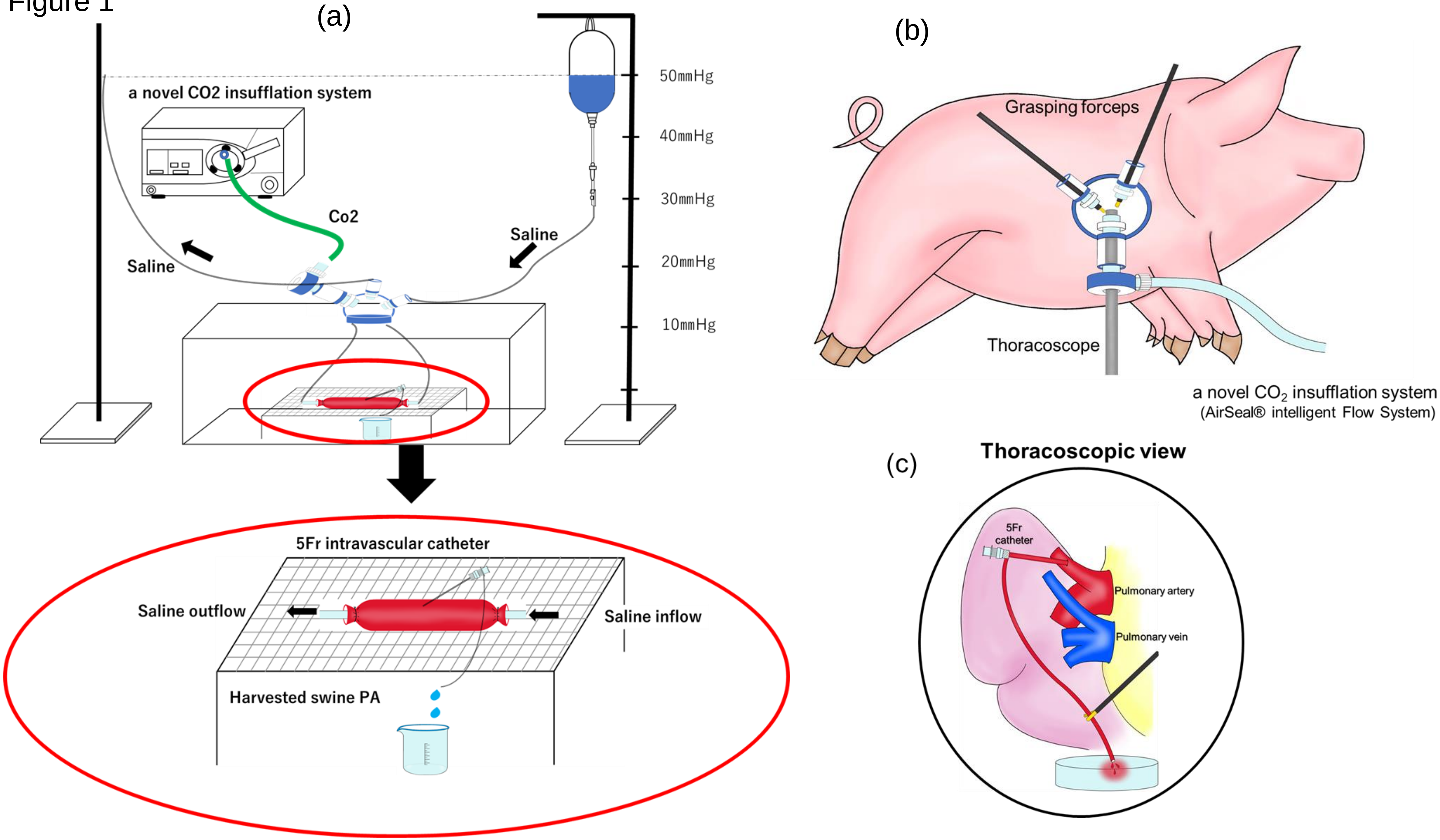

(a)
a novel CO2 insufflation system
50mmHg
40mmHg
30mmHg
20mmHg
10mmHg
Co2
Saline
Saline
5Fr intravascular catheter
Saline outflow
Saline inflow
Harvested swine PA
(b)
Grasping forceps
Thoracoscope
a novel CO2 insufflation system
(AirSeal® intelligent Flow System)
(c)
Thoracoscopic view
5Fr catheter
Pulmonary artery
Pulmonary vein

Figure 2

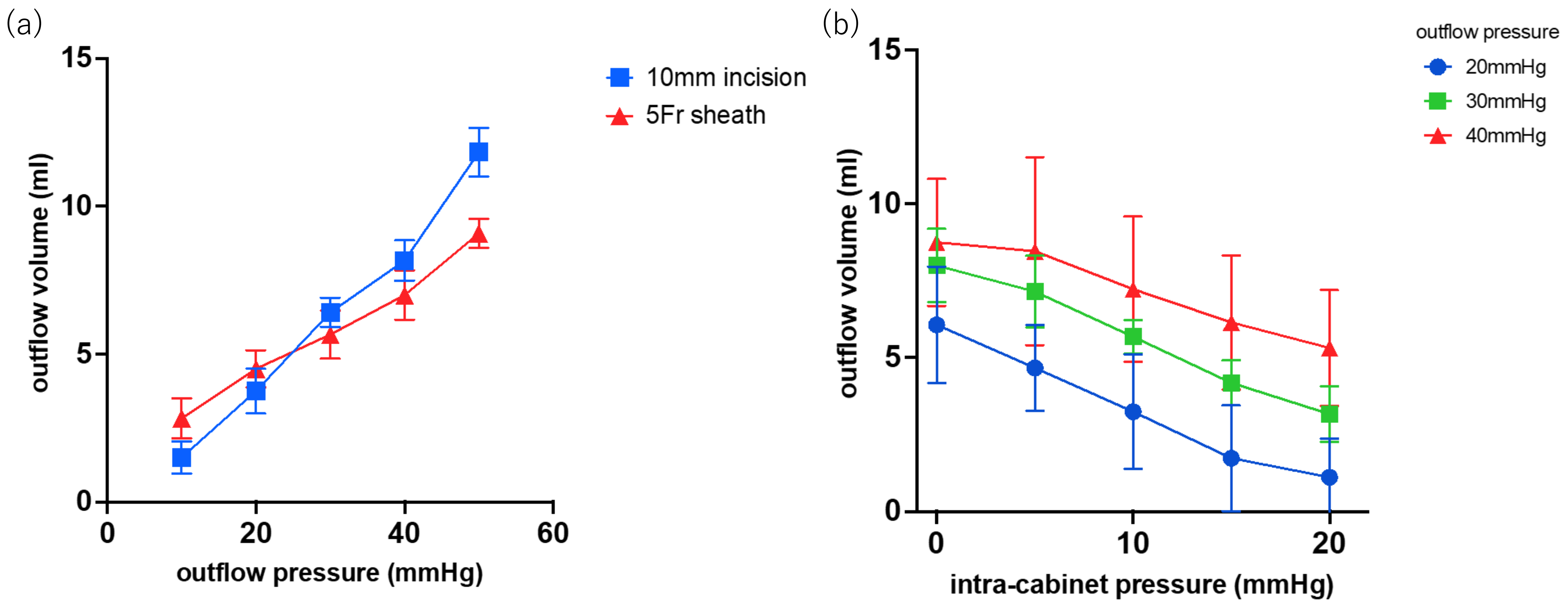
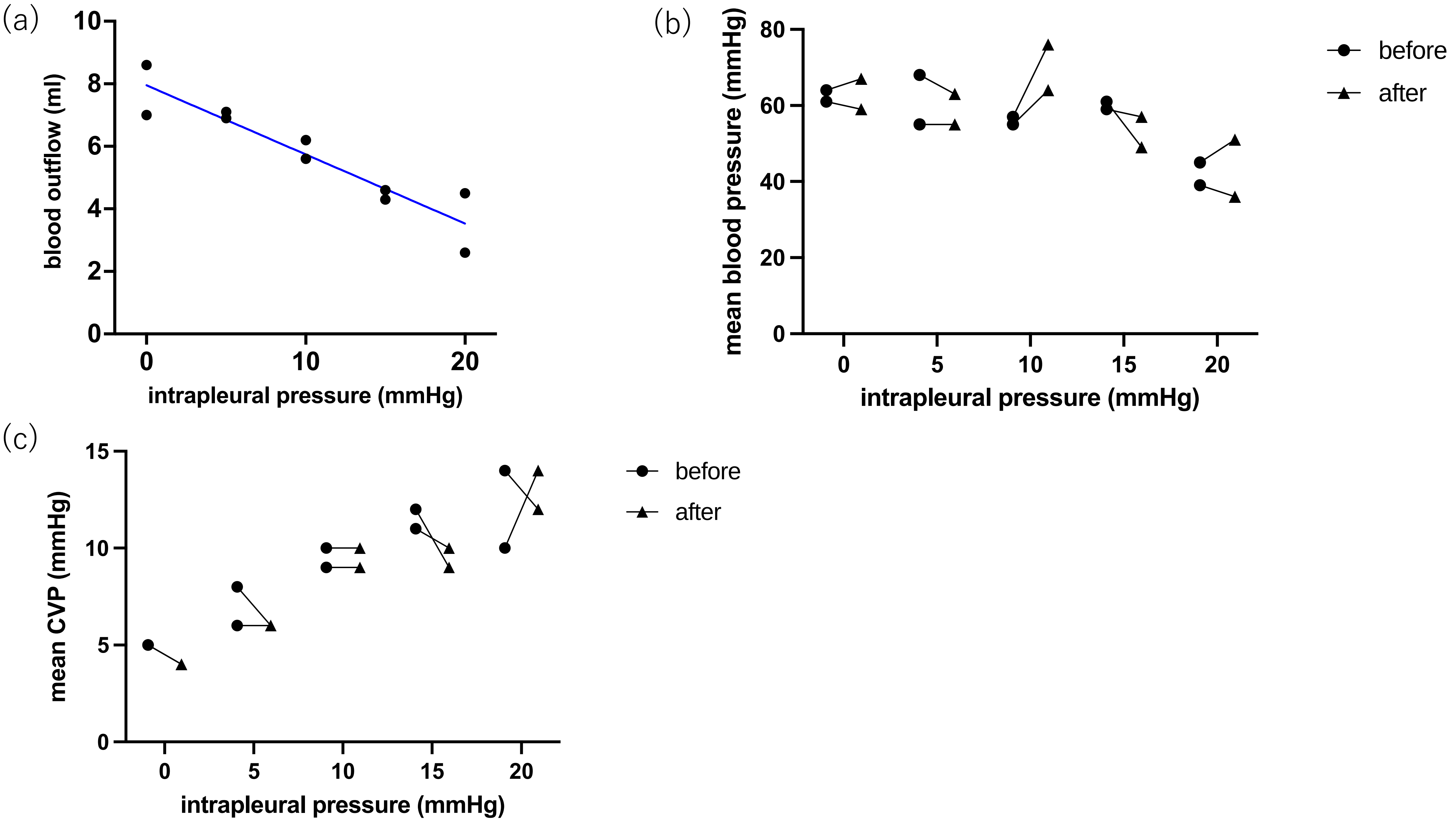

(a)
15
10
5
0
outflow volume (ml)
0
20
40
60
outflow pressure (mmHg)
10mm incision
5Fr sheath
(b)
15
10
5
0
outflow volume (ml)
0
10
20
intra-cabinet pressure (mmHg)
outflow pressure
20mmHg
30mmHg
40mmHg

Figure 3



(a)
blood outflow (ml)
10
8
6
4
2
0
0
10
20
intrapleural pressure (mmHg)
(b)
mean blood pressure (mmHg)
80
60
40
20
0
0
5
10
15
20
intrapleural pressure (mmHg)
before
after
(c)
mean CVP (mmHg)
15
10
5
0
0
5
10
15
20
intrapleural pressure (mmHg)
before
after