Short Communication

External jugular vein catheterization technique for continuous blood sampling in gilts

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Abstract

External jugular vein blood collection is common in gilts and sows, but continuous sampling for hormonal and pharmacokinetic research requires catheterization. The standard protocol has remained largely unchanged for nearly 40 years. This study aimed to develop a modified external jugular vein catheterization technique in gilts. Ten gilts (72.0 ± 3.4 kg) underwent surgery under general anesthesia after 12-hour fasting. Sedation was induced with intramuscular Xylazine (4.4 mg/kg) and Tiletamine–Zolazepam (2.2 mg/kg), followed by intravenous Thiopental sodium (10 mg/kg). Once anesthetized, gilts were placed in dorsal recumbency, and the external jugular fossa was prepared under sterile conditions. A 10 cm incision was made above the external jugular vein, and connective tissue was carefully cleared. A cranial ligature blocked blood flow, and a 0.5 cm longitudinal incision was made caudally on the external jugular vein wall. A #5 feeding tube (50 cm) was modified as a catheter with three external rings for fixation. One ring was placed at the exit site, another inside the vein, and the third at the fixation point. The internal ring was secured with 3-0 vicryl sutures to ensure blood flow. The catheter was flushed with heparinized saline (10 IU/ml) to prevent clotting and routed subcutaneously before incision closure with 3-0 monofilament nylon sutures. After a 48-hour recovery, the catheters remained functional for at least 4 days, ensuring sample quality and procedural success. This modified technique enhances continuous blood sampling feasibility in gilts.

Keywords: animal welfare, catheterization, external jugular vein, surgery, pig

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Received February 10, 2025

Accepted March 2, 2025

Introduction

Chronic external jugular vein catheterization has been widely utilized in pig research for over 40 years, facilitating frequent or continuous blood sampling without the need for repeated venipuncture (Karlbom *et al*., 1982; Rodriguez and Kunavongkrit, 1983; Pluschke *et al*., 2017). Since 1983, this method has been specifically designed and routinely employed in long-term studies, particularly for blood sampling in unrestrained pigs (Rodriguez and Kunavongkrit, 1983). It is an essential technique for pharmacokinetic studies, enabling the monitoring of drug absorption, distribution, metabolism, and excretion through serial blood sampling (Kong *et al*., 2022). Similarly, it is invaluable for hormonal studies that require frequent sampling to observe physiological changes in hormone levels (Marchant-Forde *et al*., 2012). By minimizing the stress and discomfort associated with repeated needle insertions, this technique not only improves animal welfare but also enhances the reliability of the data collected.

Chronic external jugular vein catheterization is adaptable for pigs of different ages and sizes. For pigs weighing over 40 kg, a surgical approach involving sedation and catheter insertion through an incision in the external jugular fossa has proven effective (Pluschke *et al*., 2017). Over the past decade, this method has remained in use and demonstrated that catheters can stay in place for at least 72 hours in pigs weighing 40–60 kg, allowing repeated blood collection with minimal stress and preventing sample hemolysis (Pluschke *et al*., 2017). In Thailand, this method has not been reported, nor has it been documented for gilts weighing over 60 kg. Therefore, it is crucial to develop a precise external jugular vein catheterization technique in gilts to support pharmacokinetic and endocrinological research effectively. This study aimed to develop a refined protocol for external jugular vein catheterization using a modified catheter in gilts weighing over 60 kg. The objective was to demonstrate the feasibility of collecting multiple large-volume blood samples (10 ml) over a 4-day period

Materials and Methods

***Animal and general management*:** This study adhered to the Ethical Principles and Guidelines for the Use of Animals for Scientific Purposes established by the National Research Council of Thailand. Approval was granted by the Institutional Animal Care and Use Committee (IACUC) in accordance with Chulalongkorn University regulations and policies on experimental animal care and use (Approval number 2331036). A total of 10 healthy pre-pubertal crossbred gilts (Landrace × Yorkshire × Duroc) with an average body weight of 72.0 ± 3.4 kg were included. The gilts were sourced from a commercial swine herd in western Thailand and transported to the Livestock Animal Hospital, Faculty of Veterinary Science, Chulalongkorn University, in Nakhon Pathom, Thailand. The animals were housed in an open housing system in 2 × 2 m^2 pens, with two gilts per pen. They received a standard commercial diet, and drinking water was provided *ad libitum* via water nipples. One week before surgical external jugular vein catheterization, a veterinarian assessed their health through physical examination and serological testing. Before surgery, the gilts were fasted for 12 hours, with water remaining accessible. Throughout the experiment, both feed and water were provided.

Premedication: The gilts were sedated via intramuscular injection of a combination of Xylazine (4.4 mg/kg, 100 mg/ml, Xylavet 100®, Thai Meiji Pharmaceutical, Bangkok, Thailand) and Tiletamine–Zolazepam (2.2 mg/kg, 100 mg/ml, Zoletil®, Virbac Laboratories, Carros, France) in the neck region. Approximately 10–15 minutes after injection, general anesthesia was induced with an intravenous injection of Thiopental sodium (10 mg/kg body weight, 100 mg/ml, Scott-Edil Pharmacia Ltd., Chandigarh, India) through the ear vein. Thereafter, the animals were placed in dorsal recumbency for the procedure, and fluid therapy was initiated using 0.9% saline (1,000 ml, ANB Laboratories Co. Ltd., Bangkok, Thailand) administered via the ear vein using a butterfly intravenous catheter.

Surgical procedure: The external jugular fossa and surrounding area were carefully cleaned using sterile techniques, followed by a precise dissection of the cervical region at the external jugular fossa. A 10 cm incision was made along the external jugular fossa, directly above the external jugular vein (Fig. 1a). The connective tissue was gently separated to locate the external jugular vein, and once identified, the surrounding connective tissue was meticulously cleared. A ligature was tied at the cranial end of the vein to block blood flow. A longitudinal incision of approximately 0.5 cm was then made in the wall of the external jugular vein caudally the ligature (Fig. 1d). A customized catheter was prepared using a #5 feeding tube, 50 cm in length (Bever Medical Industry, Samut Prakan, Thailand), certified by ISO 13485. The feeding tube was modified into a catheter by trimming the tip to form three rings, each approximately 0.3 cm wide (Fig. 1b). Place the rings on the exterior part of the feeding tube (Fig. 1b), positioning the ring closest to the tip approximately 15 cm from the distal end (Fig. 1b). One ring was positioned at the site where the catheter exited the vein, while another was placed inside the vein at the fixation point (Fig. 1c). The internal ring was firmly secured to the catheter to ensure unobstructed blood flow. To anchor the catheter within the vein, three surgical knots were tied around the fixation ring using 3-0 vicryl sutures. Blood samples were taken to confirm that there were no leaks or obstructions in the vein. The catheter was then flushed with heparinized sodium (100 IU/ml, L.B.S. Laboratory Ltd, Samut Prakan, Thailand), diluted in saline solution at a ratio of 10 IU per 1 ml of saline solution to prevent clotting. The catheter was routed through the subcutaneous layer near the incision site, which was subsequently closed with 3-0 monofilament nylon sutures to minimize dead space. Close the wound using a horizontal mattress suture combined with simple interrupted sutures (Fig. 2a). An extension set was positioned at the upper part of the incision, and the area was securely sutured close with 3-0 monofilament nylon.

The catheter was secured to the skin using simple interrupted or Chinese finger trap stitch before the

wound was covered with Betadine and sterile gauze (Fig. 2a). The catheter and extension were then taped to the back of the neck for stability (Fig. 2b). During recovery, post-operative medications, including analgesics (2 mg/kg tolfenamic acid, 40 mg/ml, Tolfedine® CS, Vetoquinol S.A., Lure, France) and antibiotics (15 mg/kg amoxicillin trihydrate, 150 mg/ml, Longamox®, Vetoquinol S.A., Lure, France), were administered. The antibiotic was administered every 48 hours for a total of three doses following the surgery. The catheter was flushed twice daily, in the evening and at night, with heparinized saline. The pigs were given 48 hours to recover before the blood sampling schedule began at 8:00 AM, with collections at 0, 5, 10, 15, 30, and 45 minutes, as well as 1.5, 2, 3, 4, 5, 6, 7, and 8 hours. Additionally, the catheter was flushed with heparinized saline between each 10 ml blood collection. The catheters remained in place for 4 days of sampling before the pigs were humanely euthanized with an overdose of anesthetic.

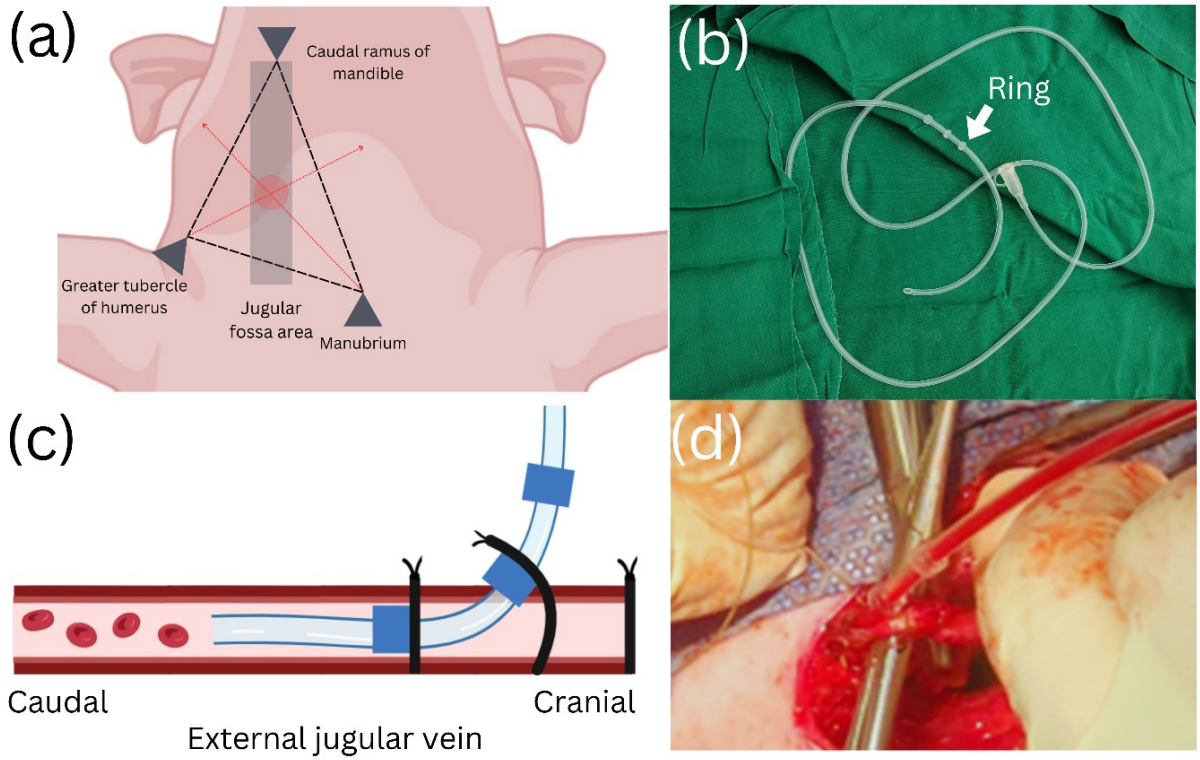

Figure 1 (a) Demonstrating the location for exploring jugular vein (b) This image shows the external jugular catheter modified from feeding tube (c) Illustration of the positioning of the catheter and locking ring placed in the external jugular vein (d) The picture demonstrates the positioning of the modified catheter fixed to the external jugular vein with sutures.

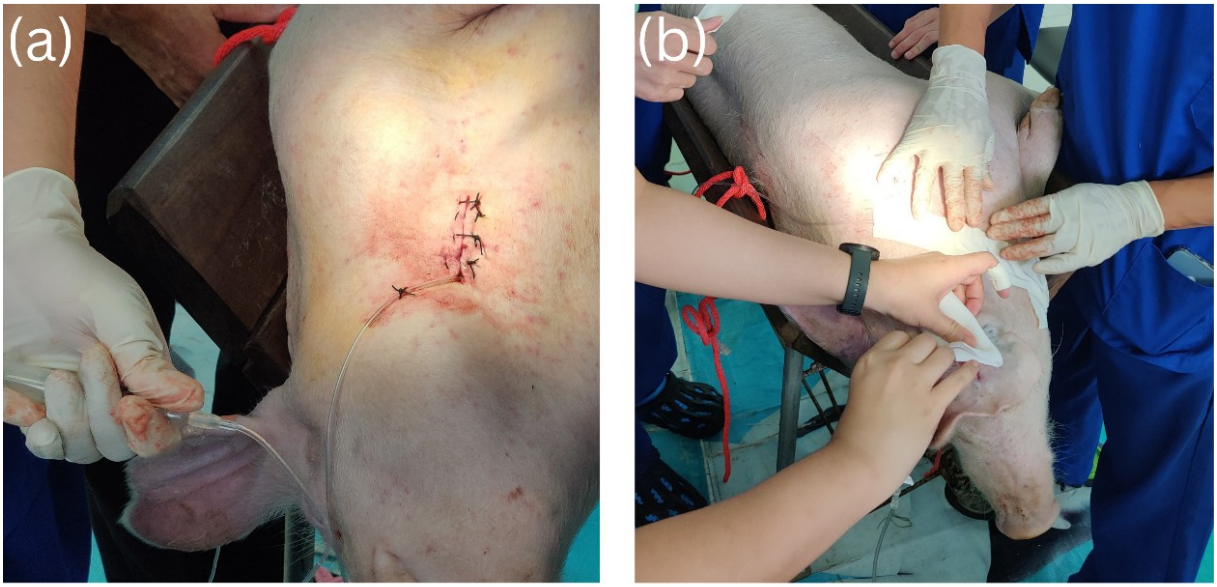

Figure 2 After completing the operation, the catheter was secured to the skin with a stitch (a), after which the wound was covered with Betadine and sterile gauze, then taped to the back of the neck for stability (b).

Results and Discussion

Ten pigs underwent catheterization, and none were excluded due to complications related to blood collection. The catheters remained functional for at least 4 days during the experiment, and blood sampling was successfully performed in all cases. However, occasional difficulties in blood collection arose due to improper animal positioning, despite correct catheter placement. This issue was resolved by adjusting the animal's position appropriately.

The successful blood collection for a minimum of 4 days after catheterization is consistent with a previous study by Pluschke *et al*. (2017), which reported that surgical external jugular catheterization in grower pigs maintained catheter patency for at least 96 hours, enabling blood sampling without occlusion. This technique allows for repeated blood sampling with minimal stress and no sample hemolysis. Additionally, the youngest reported age for successful catheterization is 12-day-old piglets, in which external jugular vein catheterization under anesthesia enabled frequent blood sampling, though it was associated with a slight reduction in weight gain (De Leonardis *et al*., 2023). These findings suggest that external jugular vein catheterization can be maintained for at least 4 days post-surgery without complications, ensuring continuous blood collection.

The present study demonstrated that feeding tubes can serve as an alternative to the Silastic catheter used in a previous study (Rodriguez and Kunavongkrit, 1983) or other intravenous catheter sets (Pluschke *et al*., 2017). The feeding tubes used in this study are made of medical-grade polyvinyl chloride (PVC), a material commonly used in neonatal and pediatric medicine for enteral feeding or gastric decompression (Freeman *et al*., 2012). The #5 size refers to its small diameter, making it suitable for delicate and small patients, such as newborns and premature infants, while its 50 cm length is appropriate for gastric placement, minimizing discomfort. This study also demonstrated that the length of the feeding tubes can be modified for use in intravenous catheterization in pigs. However, previous research has suggested that PVC may induce blood coagulation, as contact between whole blood and PVC tubing can increase thrombin generation (Frank *et al*., 2013). Despite this, coagulation is a time-dependent process. In the present study, heparin was regularly used as an anticoagulant during blood collection, ensuring no coagulation issues as long as the tube was flushed with heparinized saline and maintained in that condition until the next collection. Additionally, the preference for these modified venous catheters over conventional intravenous catheter sets is due to their lower cost and greater availability, making them a more practical and accessible option.

Alternatively, an external jugular vein catheterization technique has been performed in pigs via the ear vein using the Seldinger technique (Ringer *et al*., 2023). The key advantage of this method is that it is non-surgical and minimally invasive, making it suitable for frequent blood collection (Ringer *et al*., 2023). The procedure involves using a thin-walled needle to access the external jugular vein, followed by the insertion of a guidewire through the needle. The needle is then removed, allowing a dilator or sheath to widen the access and facilitate catheter placement (Ringer *et al*., 2023). However, applying the Seldinger technique in pigs can be challenging due to anatomical differences and the requirement for specialized expertise and proper facilities to ensure procedural success (Damm *et al*., 2000). In Japan, external jugular vein catheterization via the auricular vein has been used and maintained for up to three days after catheter insertion (Niiyama *et al*., 1985). However, this technique has occasionally faced difficulties in blood collection due to the stiffness of the catheter (Niiyama *et al*., 1985). Based on the present findings, intravenous catheterization remains a reliable technique for frequent blood sampling in pigs, with no observed issues related to blood obstruction.

A limitation of the present study was that surgical catheterization in pigs was performed without endotracheal intubation. However, the absence of intubation did not compromise the success of the procedure or the stability of the animals. Despite this, endotracheal intubation remains the recommended standard for surgical procedures, as it is essential for airway management, aspiration prevention, and respiratory support, ensuring both animal safety and procedural success (Malavasi, 2024). In pigs, the appropriate endotracheal tube size varies based on body weight: 3–4 mm internal diameter (ID) for piglets, 6–7 mm ID for pigs weighing 10–25 kg, and 16–18 mm ID for large sows (Henrikson *et al*., 1995). However, the surgical duration for intravenous catheterization in pigs is relatively short, typically lasting 30–45 minutes, and no cases of respiratory apnea were observed in the present study. Nevertheless, for safety reasons, the use of endotracheal intubation is recommended during general anesthesia in pigs to minimize potential risks.

Acknowledgements

This research was funded by the National Research Council of Thailand (NRCT) (N42A660892). We sincerely appreciate the invaluable expertise and support provided by Dr. Theerawat Tharasanit, Cong Bang Ngo, Nest Dale Bartolome, and Sittat Chumsri from the Department of Obstetrics, Gynaecology, and Reproduction, Chulalongkorn University, during the surgical procedure.

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