

Superficial anatomic landmarks can be used to triangulate the location of canine peripheral lymphocentrums: superficial cervical, axillary, and superficial inguinal

Natalie J. Worden, DVM¹; Judith Bertran, DVM, MS, DACVS^{1*}; Meghan M. Watt, DVM¹; Penny S. Reynolds, PhD²; Carlos H. de Mello Souza, DVM, DACVS, DACVIM¹; Elizabeth A. Maxwell, DVM, MS, DACVS¹; Christopher A. Adin, DVM, DACVS¹; Kathleen Ham, DVM, MS, DACVS¹; Penny J. Regier, DVM, MS, DACVS¹

¹Department of Clinical Sciences, College of Veterinary Medicine, University of Florida, Gainesville, FL

²Department of Anesthesiology, College of Medicine, University of Florida, Gainesville, FL

*Corresponding author: Dr. Bertran (bertran.judith@ufl.edu)

Received November 21, 2022.

Accepted January 1, 2023.

doi.org/10.2460/javma.22.11.0518

OBJECTIVE

To utilize the geometry of superficial anatomic landmarks to guide incisional location and orientation for peripheral lymphadenectomy, document deep anatomic landmarks for lymphocentrum identification, and develop novel surgical approaches to the superficial cervical, axillary, and superficial inguinal lymphocentrums in dogs.

ANIMALS

12 canine cadavers.

PROCEDURES

2 cadavers were used for a pilot investigation to determine optimal body positioning, select superficial anatomic landmarks for lymphocentrum identification, and evaluate novel surgical approaches to the 3 lymphocentrums. These lymphocentrums were then dissected in 10 additional cadavers using these novel surgical approaches. Measurements of the distances from lymphocentrum to landmark and between landmarks were obtained for each lymphocentrum. Deep anatomic landmarks were recorded for each dissection. The mean and SD were calculated for each measurement and used to develop geometric guidelines for estimating the location of each lymphocentrum for these surgical approaches.

RESULTS

Each peripheral lymphocentrum was found in the same location relative to the respective, predetermined, superficial, anatomic boundaries in all cadavers. Briefly, the superficial landmarks to each lymphocentrum were as follows: (1) superficial cervical: wing of atlas, acromion process of scapula, greater tubercle of humerus; (2) axillary: caudal border of transverse head of superficial pectoral muscle, caudal triceps muscle, ventral midline; and (3) superficial inguinal: origin of pectineus muscle, ipsilateral inguinal mammary gland, ventral midline. The proposed superficial and deep surgical landmarks were identified within every cadaver. The previously undescribed surgical approaches were effective for lymphocentrum identification.

CLINICAL RELEVANCE

Anatomic landmarks provided in this study may help reduce surgical time and tissue trauma during peripheral lymphadenectomy in dogs. This study was also the first to describe a surgical approach to the superficial inguinal lymphocentrum and ventral approaches to the superficial cervical and axillary lymphocentrums and provided previously unpublished anatomic landmarks for a lateral approach to the superficial cervical lymphocentrum.

Lymphadenectomy is a commonly performed procedure in veterinary medicine and a recommended method of screening for metastatic disease in animals with certain neoplasms.^{1,2} The presence of lymph node (LN) metastasis is an important factor when developing treatment plans and estimating prognosis for veterinary oncology patients.^{1,3-6} Removal of metastatic LNs has also been demonstrated to confer a survival benefit in dogs.^{3,5,6}

In dogs, clinically normal, nondiseased peripheral lymphocentrums can be divided into 2 categories: nonpalpable and palpable. Nonpalpable peripheral lymphocentrums include parotid, medial retropharyngeal, deep cervical, axillary, and inguinal. Palpable lymphocentrums include mandibular, superficial cervical, and popliteal. The limited number of published reports describing surgical approaches to canine peripheral lymphocentrums mostly describes palpable lymphocentrums,

which are typically easier to find and dissect.^{1,7-10} Well-described approaches to nonpalpable lymphocenters are limited to the medial retropharyngeal and parotid lymphocenters, which are commonly approached in combination with the palpable mandibular lymphocenter.^{1,7-11} Unilateral approaches to the superficial cervical and axillary lymphocenters have each been described in a single report.^{1,12} The paucity of described surgical techniques and landmarks to allow for systematic dissection of some peripheral lymphocenters may result in prolonged surgical times and excessive soft tissue dissection, particularly when lymphocenters are nonpalpable or the ability to palpate is lost during the process of surgical dissection, body positioning, or draping. A recent study² demonstrated that, on average, 40% ± 27% of the total surgical time for primary mass removal and lymphadenectomy in dogs was dedicated to identification and removal of the sentinel LN. Additionally, surgeons perceived peripheral LN identification to be, on average, moderately difficult (55/100 on a visual analog scale) regardless of their depth and location, with axillary LNs rated as the most difficult to identify (75/100 and 85/100).² Given these challenges, veterinarians may be more likely to forego lymphadenectomies despite the potentially high diagnostic and treatment value.

Incisional location is likely the first key step in lymphocenter identification during lymphadenectomy, especially when the LNs are not palpable. Improved accuracy of incisional placement may facilitate LN identification and reduce both surgical times and tissue trauma during LN dissection. Patient positioning is a second factor that may impact the difficulty of the procedure by altering the location of lymphocenters relative to the surrounding anatomy. The description of consistently identified deep anatomic landmarks may aid surgeons in orienting themselves when performing lymphadenectomies, which could reduce unnecessary damage to vital structures during tissue dissection. To the authors' knowledge, no studies exist that determine repeatable guidelines for positioning and approaches to the superficial cervical, axillary, and superficial inguinal lymphocenters.

Despite the importance of lymphadenectomy in the management of veterinary oncological cases, guidelines for the surgical extirpation of many of the peripheral LNs are either infrequently reported, incompletely described, or absent within the veterinary literature. The objectives of this study were to (1) utilize the geometry of superficial, anatomic landmarks on canine cadavers to guide incisional placement for lymphadenectomy of the superficial cervical, axillary, and superficial inguinal lymphocenters; (2) propose novel surgical approaches to the superficial inguinal and axillary lymphocenters; and (3) identify deep anatomic landmarks that can be consistently used for identification of each lymphocenter. Our hypothesis was that each identified lymphocenter would be in approximately the same location relative to its respective, preselected, superficial, and anatomic landmarks and that the identified deep anatomic landmarks would be consistently identified across a group of canine cadavers.

Materials and Methods

Specimens

The project was exempt from IACUC approval in our institution and occurred from August 12, 2021, through February 1, 2022. Twelve adult canine cadavers were obtained from a local animal shelter after euthanasia for reasons unrelated to this study. Cadavers were free of diseases that could induce lymphadenomegaly. Sample size and breed selection were dictated by the limited availability of canine cadavers at the time of the study. Two fresh canine cadavers (1 sexually intact male and 1 sexually intact female) were used after 24-hour storage at 3 °C to pilot choice of landmarks and proposed surgical approaches. For the definitive study, dissections and measurements were performed on the remaining 10 adult mixed-breed canine cadavers. The 10 cadavers were stored at -16 °C until needed for data collection. Prior to dissections, frozen cadavers were thawed at room temperature for 48 to 72 hours. Hair was clipped bilaterally on the regions of interest prior to dissections.

Selection of superficial and deep landmarks

Consultation with a veterinary anatomist (M.S. Sapper, BS, University of Florida Department of Physiological Sciences, verbal communication, August 12, 2021) using formalin-embalmed canine cadavers at the University of Florida College of Veterinary Medicine Anatomy Laboratory was initially conducted to determine identifiable deep landmarks surrounding each lymphocenter: superficial cervical, axillary, and superficial inguinal. This was followed by bilateral dissections of the superficial cervical, axillary, and superficial inguinal lymphocenters on 2 canine cadavers (1 sexually intact male and 1 sexually intact female) to determine a set of major, palpable superficial landmarks that could be used to triangulate each lymphocenter in both cadavers and practice the proposed novel surgical approaches. Superficial landmarks surrounding each lymphocenter were selected on the basis of the following criteria: the landmark (1) is an anatomic structure that is superficial and externally palpable, (2) is expected to be reliably identified in every dog regardless of surgical positioning or body condition/conformation, (3) is commonly known to clinicians, and (4) when used along with the other selected landmarks, has the lymphocenter located within their bounds. Deep anatomic structures and their locations relative to the lymphocenters were also recorded during these pilot dissections and compared to the landmarks identified in the embalmed cadavers.

Cadaver positioning, dissection, and measurement of lymphocenters

Dissections were performed bilaterally on 10 canine cadavers by 3 investigators (NJW, MMW, and an additional investigator). Measurements were obtained on all dissected peripheral lymphocenters by a single investigator (NJW) using a flexible tape measure and analog calipers. The distance was recorded between each of the selected landmarks and from a

metal pin placed in the center of each LN (or within the center of a lymphocentrum containing multiple LNs) to each landmark. For the axillary and superficial cervical dissections in which the LN(s) were located deeper within the tissues, measurements between the LN(s) and superficial landmarks were made on the epidermal plane. Depth measurements were not recorded for lymphocenters due to varying degrees of superficial-to-deep displacement during dissection of overlying soft tissues and differences in cadaver body condition score that affected lymphocentrum depth but not the distance between bony landmarks.

Superficial cervical lymphocentrum—For superficial cervical lymphocenters, ventral and lateral approaches were performed. The ventral approach was randomly assigned to the right or left lymphocentrum (QuickCalcs Random Number Calculators; GraphPad Software), and the lateral approach was then assigned to the opposite side. For the ventral approach, cadavers were positioned in dorsal recumbency with the forelimb tied caudally in full shoulder flexion and elbow extension. For the lateral approach, the cadaver was positioned in lateral recumbency with the dissection side facing up and the forelimb resting in approximately natural standing joint angles, parallel to the floor.

Axillary lymphocentrum—For axillary lymphocenters, cadavers were positioned in dorsal recumbency with the forelimbs in full abduction, positioned with approximately 90° of flexion at the shoulder and elbow.

Superficial inguinal lymphocentrum—Superficial inguinal lymphocentrum dissections were performed with cadavers positioned in dorsal recumbency. The hind limbs were abducted into a frog-leg position with the femur approximately perpendicular to midline and parallel with the floor.

Lymphocentrum triangulation

In all cadavers, each lymphocentrum was surgically identified. For each cadaver and lymphocentrum, a minimum of 3 superficial landmarks were identified to create a virtual triangle or polygon inside of which the lymphocentrum should be located. The sample means were calculated from the previously described measurements and used to create triangle or polygon diagrams in which each vertex represented a superficial landmark (**Figure 1**). Measurements were then taken from each landmark to its respective lymphocentrum. The sample means were calculated from these measurements. Thereafter, circles centered around each vertex were drawn with a radius equal to the mean distance between the respective lymphocentrum and landmark. Shading around each circle represented the SD for each measurement. These geometric diagrams were drawn to scale using the GeoGebra online geometry tool.¹³

Surgical approaches

Superficial cervical lymphocentrum: ventral approach—A 5-cm craniodorsal to caudoventral skin

incision was made spanning the middle third of the triangle created using the selected superficial anatomic landmarks (caudal aspect of the wing of the atlas, distal aspect of the scapular acromion process, and craniomedial aspect of the humeral greater tubercle), oriented approximately parallel to the anatomic plane connecting the caudal aspect of the wing of the atlas to the craniomedial aspect of the greater tubercle of the humerus (**Figure 2**). The subcutaneous tissue was incised, and dissection was performed until the division in muscle fibers between the omotransversarius muscle (lateral) and brachiocephalicus muscle (medial) could be identified. The fascia between the 2 muscles was incised, and the brachiocephalicus muscle was retracted medially, revealing perinodal adipose tissue deep to both muscles. The adipose tissue was grasped and elevated using thumb forceps, and the LN was identified within. The lymphocentrum was superficial and cranial to branches of the superficial cervical artery and vein and a muscular branch of the suprascapular nerve.

Superficial cervical lymphocentrum: lateral approach—A 5-cm craniodorsal to caudoventral skin incision was made spanning the middle third of the superficial landmark triangle (vertices: caudal aspect of the wing of the atlas, distal aspect of the scapular acromion process, and craniomedial aspect of the humeral greater tubercle), oriented parallel to the anatomic plane connecting the caudal aspect of the wing of the atlas to the craniomedial aspect of the greater tubercle of the humerus (**Figure 2**). Identification of the division between the omotransversarius muscle (dorsolateral) and brachiocephalicus muscle (ventromedial) was performed as described above. Subjectively, less depth of dissection was required to identify the muscle division compared to the ventral approach. The fascia between the 2 muscles was incised, and the omotransversarius muscle was retracted dorsally, revealing perinodal adipose tissue deep to the muscles. The adipose tissue was similarly elevated, and the LN was identified within. Again, the lymphocentrum was found superficial and cranial to branches of the superficial cervical artery and vein and a muscular branch of the suprascapular nerve. Typically, a solitary LN structure was identified; however, dissection of the LN capsule often revealed multiple contiguous LNs within the lymphocentrum.

Axillary lymphocentrum: ventral approach—Centered over the point at which the caudal border of the superficial pectoral muscle creates a cutaneous depression at the axilla, a 5- to 6-cm mediolateral incision was made through the skin and subcutaneous tissue immediately caudal and parallel to the caudal border of the transverse head of the superficial pectoral muscle (**Figure 3**). Blunt dissection was continued immediately caudal to the superficial pectoral muscle until the muscle fibers of the deep pectoral muscle could be identified. Blunt dissection through the deep pectoral muscle, parallel to the muscle fibers, was performed at the division between the sections of the deep pectoral muscle overlying the body wall and overlying the proximal humerus. A pair of self-retaining Weitlaner or Gelpi retractors were placed in

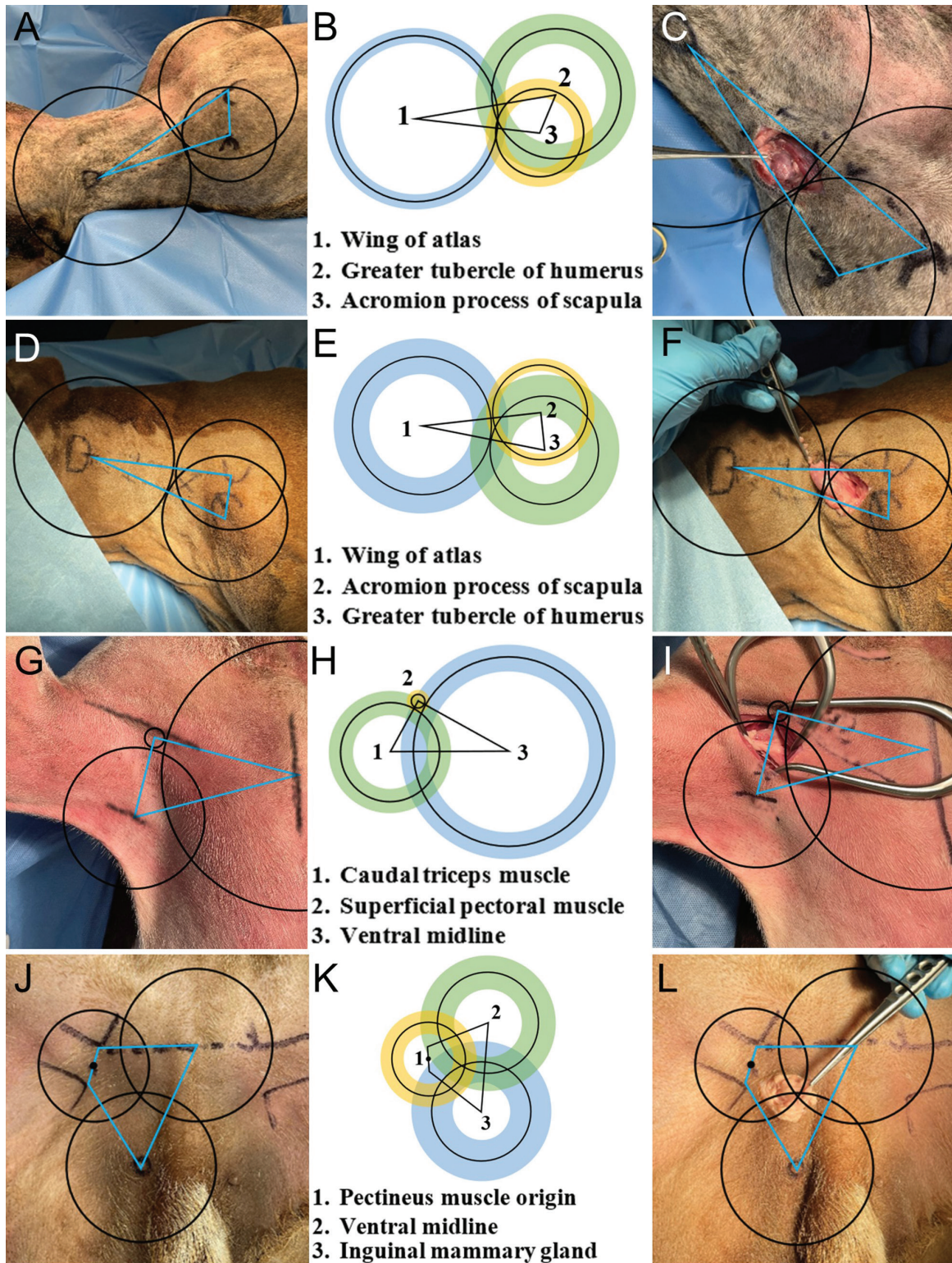


Figure 1—Depictions of the lymphocentrum triangulation process used in a canine cadaveric study (n = 12) between August 12, 2021, and February 1, 2022, to guide incisional location and orientation for peripheral lymphadenectomy of the superficial cervical lymphocentrum by a ventral (A, B, and C) or lateral (D, E, and F) approach, the axillary lymphocentrum (G, H, and I), and superficial inguinal lymphocentrum (J, K, and L). For each circle, the center represents an identified superficial landmark (ie, wing of the atlas), the radius represents the mean distance between the landmark and the respective lymphocentrum, and the shading represents the SD of the respective mean distance. For each triangulation, the region of intersection of the 3 circles within the bounds of the triangle approximates the expected location of the respective lymphocentrum.

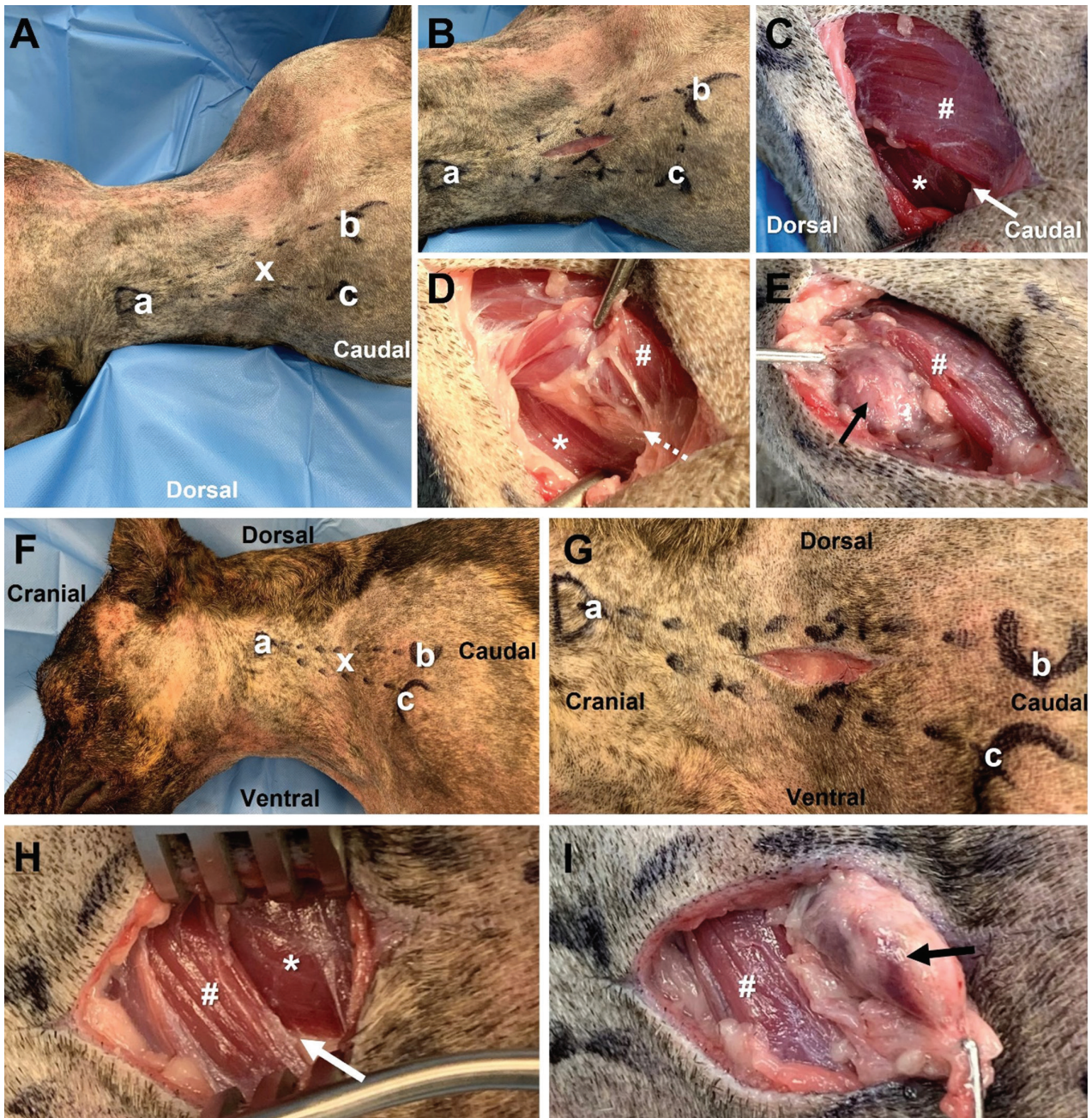


Figure 2—Serial images for triangulation of the superficial cervical lymphocentrum from a ventral approach (A through E; dorsal recumbency) and lateral approach (F through I; right lateral recumbency) performed during the study described in Figure 1, showing location and orientation of the skin incisions centered over the expected location (x) of the superficial cervical lymphocentrum, division (white arrow) between the omotransversarius muscle (asterisk) and brachiocephalicus muscle (pound sign), underlying perinodal adipose tissue (dashed white arrow), and a superficial cervical lymph node (black arrow). Dashed black lines drawn on the skin of the cadavers outline the triangle used to predict lymphocentrum location. A and B—Landmarks are the caudal aspect of the wing of the atlas (a), craniomedial aspect of the greater tubercle of the humerus (b), and distal aspect of the acromion process of the scapula (c). F and G—Landmarks are the caudal aspect of the wing of the atlas (a), distal aspect of the acromion process of the scapula (b), and craniomedial aspect of the greater tubercle of the humerus (c).

the deep pectoral incision to improve exposure of deeper structures. Blunt dissection was continued deep to the deep pectoral muscle, medial to the latissimus dorsi muscle, and medially toward the rectus thoracis muscle (at the thoracic wall) until the perinodal adipose tissue was identified, grasped, and elevated. The axillary LN was

identified within this perinodal adipose tissue. The lateral thoracic nerve, artery, and vein were consistently identified in all cadavers after dissection through the deep pectoral muscle, and dissection was continued medially and dorsally to this neurovascular bundle to find the lymphocentrum.

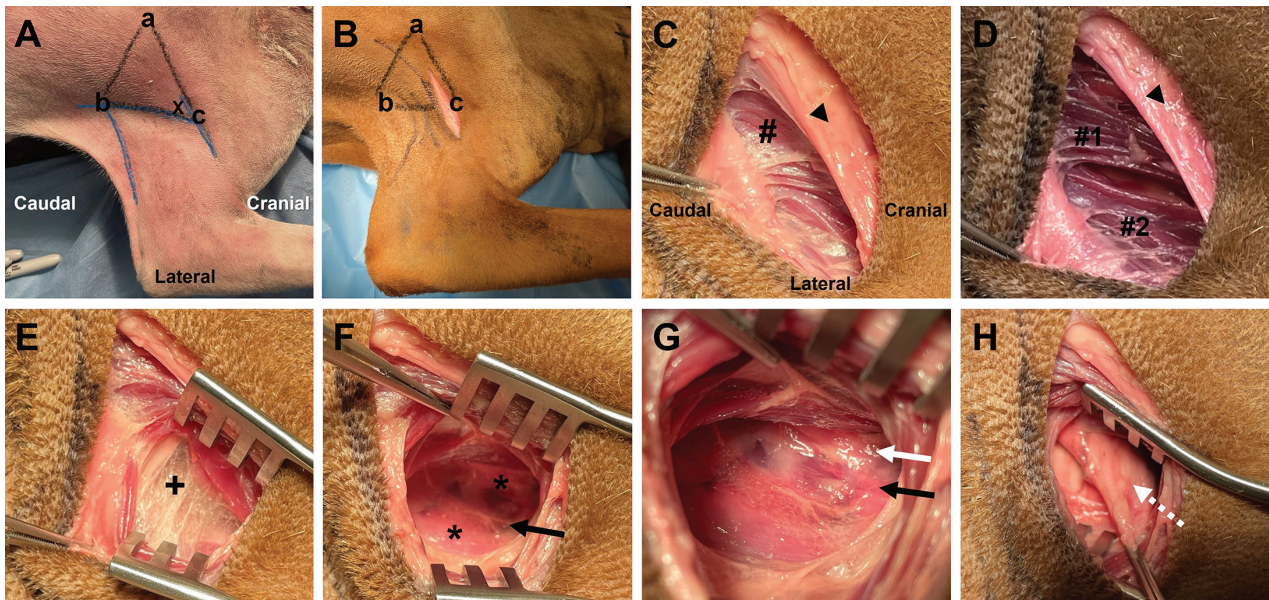


Figure 3—Serial images for triangulation of the axillary lymphocentrum from a ventral approach performed during the study described in Figure 1, showing the location and orientation of the skin incision centered over the expected location (x) of the axillary lymphocentrum and the deep (pound sign) and superficial (arrowhead) pectoral muscles. The dog is in ventral recumbency with its head toward the right. The deep pectoral muscle is bluntly dissected between its ventromedial leaf (#1) overlying the ventrolateral thorax and its dorsolateral leaf (#2) superficial to the proximal humerus. Subsequent images show the deep fascia (plus sign), adipose tissue (asterisk) bisected by the lateral thoracic artery, vein, and nerve (black arrow), region for continuation of dissection (white arrow) located medial to the lateral thoracic neurovascular bundle, and an axillary lymph node (white arrow). Blue lines drawn on the skin of the cadaver represent the palpable caudal borders of the triceps muscle caudally and the superficial pectoral muscle cranially, with a horizontal line representing the palpable lateral margin of the thorax. Black lines drawn on the skin of the cadaver outline the triangle used to predict lymphocentrum location. a = Ventral midline at a level bisecting the muscular landmarks (b and c). b = Caudal aspect of triceps muscle where it meets the ventrolateral thorax. c = Cutaneous depression within the axilla created by the caudal aspect of the superficial pectoral muscle where it meets the ventrolateral thorax.

Superficial inguinal lymphocentrum: ventral approach—A 2- to 3-cm skin incision was started medial to the center of the origin of the pectineus muscle and extended craniomedially so that it was centered between the superficial landmarks (origin of the pectineus muscle, ventral midline at the level of the caudal border of the pectineus origin, and the ipsilateral inguinal mammary gland), angled approximately 45° from ventral midline (**Figure 4**). A combination of blunt and sharp dissection was used to dissect through the superficial layers of underlying adipose tissue until the LN(s) were identified. The excess adipose tissue associated with the inguinal mammary gland in the female dog subjectively increased the difficulty of the dissection; however, dissection of mammary tissue was not required to locate the LN(s). The external pudendal artery and vein were found medial to the lymphocentrum, and the caudal superficial epigastric artery and vein were seen branching from the external pudendal vessels at the level of or immediately caudal to the lymphocentrum.

Statistical analysis

Summary data for distances between specified landmarks (cm) are reported as mean and SD limits, calculated as (mean – SD, mean + SD). For superficial cervical lymphocenters, landmark distances were measured for both ventral and lateral surgical approaches,

and SDs were calculated separately. For axillary and superficial inguinal lymphocenters, measurements were obtained on both left and right sides for each dog using only the ventral approach; therefore, SDs for paired differences were calculated for these distance metrics. Differences between ventral-lateral and left-right measurements paired by dog were described by mean differences and 95% confidence limits. Lymph node counts were categorized by frequency of occurrence in each count category (number of LNs [1, 2, 3] per lymphocentrum site per dog). Descriptive statistics were calculated in SAS *proc means* and pooled SD for paired data in *proc glm* (SAS version 9.4; SAS Inc).

Results

Specimens and lymphocentrum measurements

Data were collected from 10 adult canine cadavers with a mean weight of 23.4 kg (SD, 6 kg; range, 15 to 33 kg). There were 4 females (F) and 6 males (M). No lymphadenomegaly was palpated on initial examination of cadavers.

Lymphocentrum dissections

Superficial cervical lymphocentrum—Twenty superficial cervical lymphocenters were dissected in

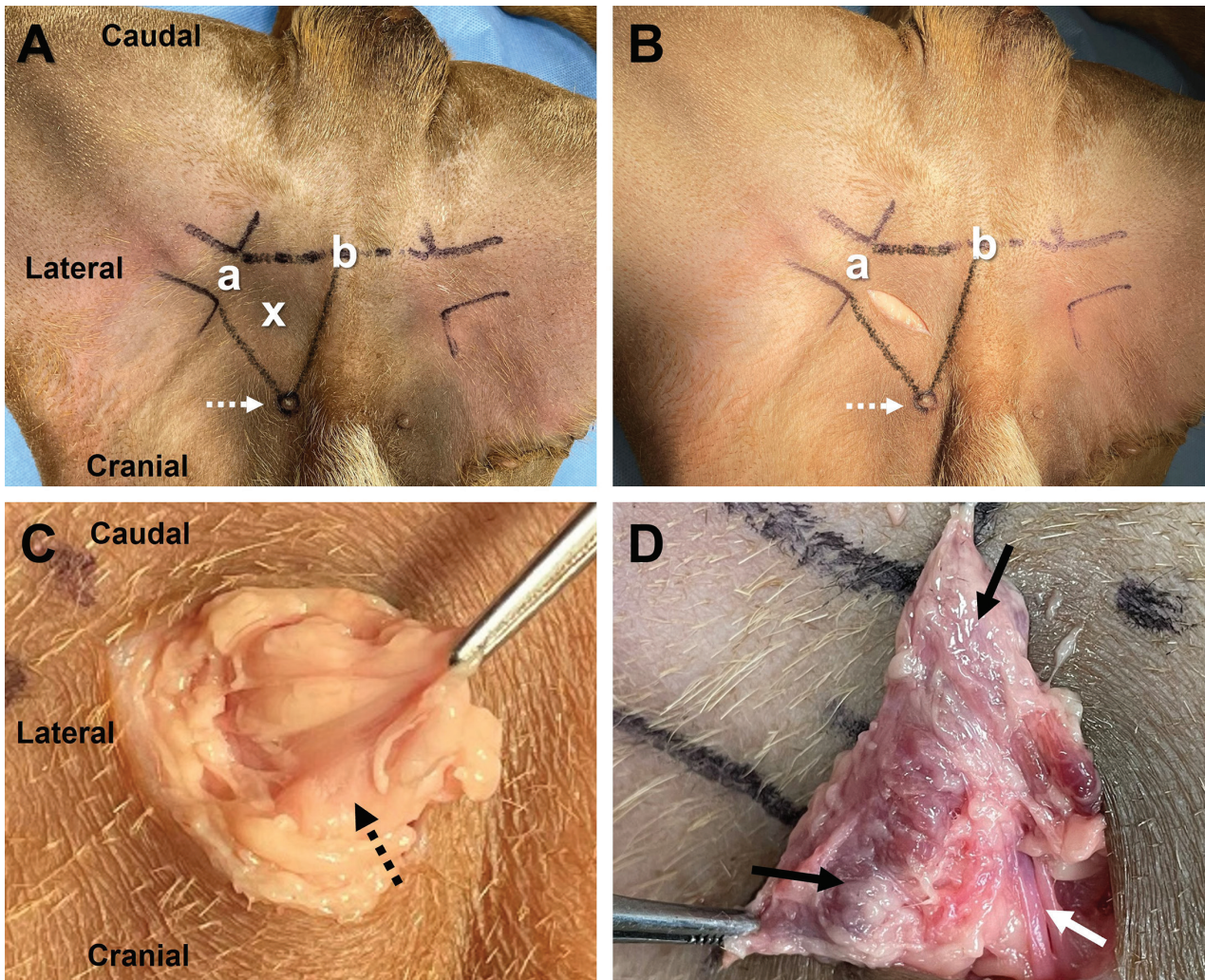


Figure 4—Serial images for triangulation of the superficial inguinal lymphocentrum from a ventral approach performed during the study described in Figure 1, showing the location and orientation of the skin incisions centered over the expected location (x) of the superficial inguinal lymphocentrum, perinodal adipose tissue (black dashed arrow), external pudendal artery and vein (white arrow), and 2 superficial inguinal lymph nodes (black arrows). Black lines drawn on the skin of the cadaver outline the trapezoid used to predict lymphocentrum location as well as to outline the borders of the pectineus muscle origin. The dog is in ventral recumbency with its head toward the bottom of the images. a = Origin of pectineus muscle. b = Ventral midline at the level of the caudal aspect of the pectineus muscle origin. White dashed arrow = Ipsilateral inguinal mammary gland.

10 canine cadavers. Ten lymphocenters (3 left [L], 7 right [R]) were dissected via a ventral approach and 10 lymphocenters (7 L, 3 R) were dissected via a lateral approach. For both ventral and lateral approaches, the lymphocentrum was found within the center of the triangle created by lines drawn to connect the 3 landmarks (Figure 1). Therefore, the skin incision was oriented craniocaudally, spanning the middle third of this anatomic triangle. Landmark distances were consistently longer (approx 1 to 3 cm, or 15% to 20% of total landmark distance) for the ventral approach compared to the lateral approach. One LN was found per lymphocentrum site for all 10 cadavers (**Table 1**).

Axillary lymphocentrum—Eighteen axillary lymphocentrum dissections (9 R, 9 L) in 9 canine cadavers (5 M, 4 F) were measured. The axillary lymphocenters from 1 cadaver were excluded due to the selected landmarks

being redefined to better account for cadavers of different body conditions. In all cadavers, the lymphocentrum was found immediately caudal to the caudal border of the transverse superficial pectoral muscle and midway along the length of this muscle, at the cutaneous depression formed at the axilla (Figure 1). Therefore, the skin incision was oriented mediolaterally, centered over the natural axillary depression formed by the caudal aspect of the transverse superficial pectoral and the axillary fold when the limb is in full abduction. Landmark distances showed no consistent differences between left and right sides. Measurements varied by approximately 1 cm on either side of the landmark. One LN was found per lymphocentrum site for all 9 cadavers (**Table 2**).

Superficial inguinal lymphocentrum—Seven superficial inguinal lymphocenters (9 L, 8 R) were dissected in 9 canine cadavers (5 M, 4 F). Three

Table 1—Mean distances between specified anatomic landmarks and 20 superficial cervical lymphocenters and differences in those distances for ventral (n = 10) versus lateral (10) approach to the superficial cervical lymphocenters in 10 canine cadavers during a study conducted between August 12, 2021, and February 1, 2022.

Measurement	Mean distance for ventral approach (cm) ^a	Mean distance for lateral approach (cm) ^a	Mean difference: lateral - ventral (cm) ^b
Acromion to wing of atlas	16.4 (15.2, 17.6)	13.2 (11.0, 15.4)	-2.0 (-4.7, 0.8)
Acromion to greater tubercle of humerus	5.5 (4.6, 6.4)	4.2 (3.5, 4.9)	-0.7 (-1.8, 0.4)
Greater tubercle of humerus to wing of atlas	18.7 (17.1, 20.3)	13.9 (10.8, 17.0)	-2.8 (-6.5, 0.9)
LN to acromion	5.8 (4.5, 7.1)	5.3 (4.6, 6.1)	-0.5 (-1.4, 0.5)
LN to wing of atlas	10.9 (9.9, 11.9)	7.7 (5.7, 9.7)	-1.7 (-4.3, 0.9)
LN to greater tubercle of humerus	8.6 (6.6, 10.6)	6.0 (3.7, 8.2)	-1.6 (-4.3, 1.2)

LN = Lymph node.

^aDistances reported as mean and SD limits (ie, mean - SD, mean + SD). ^bDifferences between distances for the lateral and ventral approach for each site are reported as mean and 95% CI limits.

Table 2—Mean distances between specified anatomic landmarks and the 18 axillary lymphocenters (9 right, 9 left) or 17 superficial inguinal lymphocenters (8 right, 9 left) and the differences in those distances for the left versus right side for 9 canine cadavers described in Table 1.

Lymphocenters	Measurement	Mean distance (cm) ^a	Mean difference: left - right, (cm) ^b
Axillary	Superficial pectoral m to triceps m	4.5 (3.7, 5.3)	0.0 (-0.9, 1.0)
	Superficial pectoral m to midline	8.1 (7.4, 8.7)	0.4 (-0.2, 1.0)
	Triceps m to midline	9.2 (8.3, 10.1)	0.5 (-0.5, 1.4)
	LN to superficial pectoral m	0.5 (0.1, 1.0)	0.3 (-0.1, 0.8)
	LN to triceps m	3.9 (2.7, 5.1)	-0.2 (-1.5, 1.2)
	LN to triceps-pectoral midline	7.4 (6.8, 8.1)	0.3 (-0.5, 1)
Superficial inguinal	Pectineus m to midline	4.0 (3.4, 4.5)	-0.2 (-0.8, 0.3)
	Pectineus m to inguinal mammary gland	3.8 (3.2, 4.4)	0.1 (-0.7, 0.9)
	Inguinal mammary gland to midline	5.2 (4.6, 5.7)	0.0 (-0.7, 0.7)
	LN to pectineus m	2.2 (1.9, 2.6)	-0.3 (-0.6, 0.1)
	LN to midline	3.1 (2.6, 3.7)	0.1 (-0.7, 0.9)
	LN to inguinal mammary gland	2.9 (2.6, 3.2)	-0.2 (-0.5, 0.1)

LN = Lymph node. m = Muscle.

^aDistances reported as mean and SD limits (ie, mean - SD, mean + SD). ^bDifferences between distances for the left versus right side for each site are reported as mean and 95% CI limits.

superficial inguinal lymphocenters could not be included due to previous dissections of these sites for a prior unrelated study. Based on the average measurements, the superficial inguinal lymphocenter was expected to be found near the center of the region bounded by the landmarks (Figure 1). Therefore, the skin incision was centered within the landmarks, starting medial to the midpoint of the origin of the pectineus muscle and extending craniomedially, at an angle of approximately 45° to midline. The superficial inguinal lymphocenters were subjectively more difficult to identify in female cadavers. Landmark distances showed no consistent differences between right and left sides and varied by less than 1 cm on either side of the designated landmark (Table 2). The number of LNs per superficial inguinal lymphocenter site was asymmetrical for 6 dogs, with 1, 2, or 3 LNs found on the left for 4, 4, or 1 dog, respectively, versus found on the right for 5, 1, or 2 dogs, respectively.

Discussion

The present study proposes a novel method for determining incisional location and orientation based on the geometry of superficial landmarks in a group of

canine cadavers. The findings of the present study support the hypothesis that each identified lymphocenter would be in approximately the same location relative to its selected superficial and deep anatomic landmarks across a group of canine cadavers. The results demonstrate that the 3 evaluated peripheral lymphocenters were consistently found in similar positions and within the bounds of the proposed superficial, anatomic landmarks in 10 canine cadavers in defined body positions. The superficial and deep anatomic landmarks used herein were consistently found in each cadaver, indicating that they may be reliable for intraoperative lymphocenter identification. Additionally, the previously undescribed approaches to the axillary and superficial inguinal lymphocenters were successfully used for lymphocenter identification in all dissected cadavers.

Although there is limited published information regarding surgical approaches to peripheral LNs in dogs, lymphadenectomy of the mandibular, medial retropharyngeal, parotid, and popliteal lymphocenters has been relatively well described in previous literature.^{1,7-11} Therefore, the present study is intended to fill the current literature gap by describing surgical approaches for lymphadenectomy of the superficial cervical, axillary, and superficial inguinal lymphocenters.

The present study evaluated both lateral and ventral approaches to the superficial cervical lymphocentrum. Despite the approaches being similar, the ventral approach was subjectively more difficult due to the redundant skin folds in the region of the lymphocentrum, necessitating deeper subcutaneous dissection for identification of our suggested muscular landmarks. Additionally, the ventral approach led to a reduced ability to palpate the LN as it was pulled dorsally into the tissues of the cervical region. The present study may provide a more distinguishable landmark for identification of this LN compared to previous descriptions: the division between the omotransversarius and brachiocephalicus muscles. Regardless of patient positioning, the superficial cervical lymphocentrum was repeatedly found within the perinodal adipose tissue deep to this muscular division, and identification of this landmark may reduce tissue dissection and surgical trauma to the omotransversarius muscle. Additionally, the authors emphasize the importance of neutral positioning of the thoracic limb to ease LN identification with the lateral approach, as alternative positioning complicated palpation and identification. Fully draping the limb into the surgical field may also improve intraoperative lymphocentrum identification by increasing the surgeon's ability to manipulate the limb.

A single caudoventral surgical approach to the axillary lymphocentrum has only recently been described in the veterinary literature, and while this approach allows for visualization of the axillary lymphocentrum, accessory axillary lymphocentrum, and axillary lymphatic trunk, it may require a larger incision and more extensive dissection for adequate exposure of the described landmarks.¹² In the present study, the axillary lymphocentrum dissection was subjectively the most challenging of the evaluated lymphocenters due to its deep position preventing palpation and limiting exposure. As a result, self-retaining retractors were necessary for adequate exposure during dissections. However, if performed as described, the surgical approach described in the present study may reduce the amount of dead space created during lymphadenectomy and requires dissection around fewer neurovascular structures than the previously described approach. The utility of the present approach in identifying the accessory axillary lymphocentrum was not evaluated.

A standard surgical approach to the superficial inguinal lymphocentrum has not been described in the veterinary literature. Despite the lack of a published, standard surgical approach, superficial inguinal lymphadenectomies are typically performed clinically in conjunction with mastectomies and removal of tumors in the caudal abdominal region, inguinal region, and hind limbs.¹⁴ A single previous report¹⁴ suggested the use of an ultrasound-guided hook-wire localization of this lymphocentrum due to the prolonged surgical time and extensive dissection associated with surgical identification. The standard surgical approach in the present study may also reduce intraoperative times and extent of tissue dissection, while eliminating the need for specialized tools. In the present study, identification of the superficial

inguinal lymphocentrum was subjectively more difficult in female cadavers, which was attributed to the increased adipose tissue associated with the developed mammary glands. This could be less of a factor in spayed females or in live dogs; however, the impact of reproductive status was not specifically examined in this study. The use of adjunctive intraoperative LN mapping techniques may prove particularly useful during superficial inguinal lymphadenectomy in female dogs.

This study was the first to test for a repeatable method of lymphocentrum identification using a geometric approach based on the superficial anatomy of canine cadavers to guide incisional location and orientation. The consistency of the superficial and deep anatomic landmarks and their locations relative to the 3 peripheral lymphocenters was evaluated across a group of canine cadavers, demonstrating repeatable identification. Furthermore, this study was the first to describe a surgical approach to the superficial inguinal lymphocentrum and a ventral surgical approach to the superficial cervical lymphocentrum, in addition to describing a novel approach to the axillary lymphocentrum. While differences in the complication rates and outcomes between the present study's proposed techniques and those previously described cannot be compared using the current cadaver models, these alternative approaches may influence preoperative patient positioning and therefore reduce the need for intraoperative repositioning of the patient between the primary tumor excision and lymphadenectomy. Further research on these techniques in live dogs would be necessary to compare outcomes for these techniques.

The primary limitations of this study included its cadaveric nature, low sample size, limited range of breeds and sizes of cadavers, measurement precision, and displacement of the LNs or landmarks during or after dissection. Additionally, visualization and palpation were the only methods used to validate LN identification in this study, and no postdissection imaging was performed to confirm that all LNs within a lymphosome had been identified; while this is consistent with how lymphadenectomies are typically performed in a clinical setting, potential unidentified LNs could have affected measurement locations in this study. All cadavers used in this study were apparently healthy, mesocephalic, and nonchondrodystrophic, and cadaver selection in this study was limited by the availability of fresh canine cadavers during the time of the study. Therefore, the accuracy of these lymphocentrum triangulations in dogs with other conformations or with pathologies affecting the LNs or surrounding landmarks is unknown. Further investigation is needed to determine the utility of the proposed surgical guidelines in dogs with other conformations. While measurement precision is likely user dependent, the method of using average measurements between anatomic landmarks in cadavers to guide surgeons in a clinical setting has been previously used in several studies.¹⁵⁻¹⁷ Finally, the cadaveric nature of this study was not perfectly representative of lymphadenectomies in live animals, so the visibility of certain deep landmarks may differ in a clinical setting.

In conclusion, the superficial cervical, axillary, and superficial inguinal lymphocentrums are in similar positions relative to the superficial anatomic landmarks selected in our study and could be consistently identified using our proposed deep anatomic landmarks across a group of canine cadavers. The findings of the present study will provide veterinarians with a method of more accurately orienting skin incisions for extirpation of these 3 peripheral lymphocentrums using the geometry of superficial anatomic landmarks. Additionally, the deep surgical landmarks can be used to guide lymphocentrum identification when performing the proposed approaches to these lymphocentrums. Identification of the superficial cervical lymphocentrum was subjectively easier via the lateral approach, which should be considered when determining patient positioning for surgery. Compared to the previously reported approach to the axillary lymphocentrum, which allows for a more complete dissection of the axillary and accessory axillary lymphatic structures, the more targeted approach described in the present study may be better suited for excisional biopsies or instances in which metastatic disease has not already been confirmed.¹² Given the subjectively increased difficulty of identification of the superficial inguinal lymphocentrum in female canine cadavers, clinicians might consider adjunctive methods of LN mapping to aid in identification of this lymphocentrum, particularly in obese female dogs.

Use of the proposed method of determining incision orientation relative to superficial landmarks may contribute to reduced dissection times, reduced tissue trauma, and improved subjective ease of lymphocentrum identification during these peripheral lymphadenectomies, particularly for veterinarians who do not have access to intraoperative LN mapping techniques or have limited experience performing lymphadenectomies. Additionally, the proposed geometric approach to lymphadenectomy may improve accuracy of incisional orientation when approaching nonpalpable lymphocentrums. This novel approach to surgical planning for lymphadenectomies may assist veterinarians in envisioning the underlying, deeper anatomic structures based on the superficial geometry of the patient when starting their surgical approaches. Future directions include investigation of lymphocentrum positioning across a larger sample size or a wider range of conformations, evaluating for iatrogenic tissue trauma, and evaluation of other poorly described peripheral lymphocentrums.

Acknowledgments

No external funding was used in this study. The authors declare that there were no conflicts of interest. Drs. Bertran, Worden, Ham, Regier, and Adin contributed to study conception and design. Drs. Worden, Watt, and Bertran contributed to data collection. Drs. Worden and Watt contributed to data analysis and interpretation. Drs. de Mello Souza and Maxwell contributed to data interpretation. Dr. Reynolds contributed to statistical analysis. All authors contributed to drafting and revising the manuscript and approved the final manuscript.

The authors would like to thank Michael Sapper for assistance with identification of anatomic structures, Dr. W. Alex Fox-Alvarez for his assistance with study design, Shelby Straight for assistance with cadaveric dissections, and Chiquitha Crews for assistance with data collection.

References

1. Wright T, Oblak ML. Lymphadenectomy: overview of surgical anatomy & removal of peripheral lymph nodes. *Today's Vet Pract.* 2016;6(4):20-29.
2. Lapsley J, Hayes GM, Janvier V, et al. Influence of locoregional lymph node aspiration cytology vs sentinel lymph node mapping and biopsy on disease stage assignment in dogs with integumentary mast cell tumours. *Vet Surg.* 2021;50(1):133-141. doi:10.1111/vsu.13537
3. Ferrari R, Marconato L, Buracco P, et al. The impact of extirpation of non-palpable/normal-sized regional lymph nodes on staging of canine cutaneous mast cell tumours: a multicentric retrospective study. *Vet Comp Oncol.* 2018;16(4):505-510. doi:10.1111/vco.12408
4. Baginski H, Davis G, Bastian RP. The prognostic value of lymph node metastasis with grade 2 MCTs in dogs: 55 cases (2001-2010). *J Am Anim Hosp Assoc.* 2014;50(2):89-95. doi:10.5326/JAAHA-MS-5997
5. Marconato L, Polton G, Stefanello D, et al. Therapeutic impact of regional lymphadenectomy in canine stage II cutaneous mast cell tumours. *Vet Comp Oncol.* 2018;16(4):580-589. doi:10.1111/vco.12425
6. Mendez SE, Drobatz KJ, Duda LE, White P, Kubicek L, Sorenmo KU. Treating the locoregional lymph nodes with radiation and/or surgery significantly improves outcome in dogs with high-grade mast cell tumours. *Vet Comp Oncol.* 2020;18(2):239-246. doi:10.1111/vco.12541
7. Smith MM. Surgical approach for lymph node staging of oral and maxillofacial neoplasms in dogs. *J Vet Dent.* 2002;19(3):170-174. doi:10.1177/089875640201900306
8. Green K, Boston SE. Bilateral removal of the mandibular and medial retropharyngeal lymph nodes through a single ventral midline incision for staging of head and neck cancers in dogs: a description of surgical technique. *Vet Comp Oncol.* 2017;15(1):208-214. doi:10.1111/vco.12154
9. Skinner OT, Boston SE, Souza CHM. Patterns of lymph node metastasis identified following bilateral mandibular and medial retropharyngeal lymphadenectomy in 31 dogs with malignancies of the head. *Vet Comp Oncol.* 2017;15(3):881-889. doi:10.1111/vco.12229
10. Wainberg SH, Oblak ML, Giuffrida MA. Ventral cervical versus bilateral lateral approach for extirpation of mandibular and medial retropharyngeal lymph nodes in dogs. *Vet Surg.* 2018;47(5):629-633. doi:10.1111/vsu.12920
11. Herring ES, Smith MM, Robertson JL. Lymph node staging of oral and maxillofacial neoplasms in 31 dogs and cats. *J Vet Dent.* 2002;19(3):122-126. doi:10.1177/089875640201900301
12. Rehnbom ER, Skinner OT, Mickelson MA, Hutcheson KD. Axillary lymphadenectomy in dogs: a description of surgical technique. *Vet Comp Oncol.* 2022;20(3):664-668. doi:10.1111/vco.12820
13. *GeoGebra Geometry.* Version 6.0. Accessed February 11, 2022. <https://geogebra.org/geometry>
14. Pierini A, Marchetti V, Rossanese M, Finotello R, Cattai A, Pisani G. Ultrasound-guided hook-wire localization for surgical excision of non-palpable superficial inguinal lymph nodes in dogs: a pilot study. *Animals (Basel).* 2020;10(12):2314. doi:10.3390/ani10122314
15. Smith NM, Segars L, Kauffman T, Olinger AB. Using anatomical landmark to avoid phrenic nerve injury during balloon-based procedures in atrial fibrillation patients. *Surg Radiol Anat.* 2017;39(12):1369-1375. doi:10.1007/s00276-017-1895-y
16. Almutairi S. A cadaveric study on the efficacy of surface marking and bony landmarks used in sacral neuromodulation. *Cureus.* 2020;12(7):e9153. doi:10.7759/cureus.9153
17. Putzer D, Haselbacher M, Hörmann R, Thaler M, Nogler M. The distance of the gluteal nerve in relation to anatomical landmarks: an anatomic study. *Arch Orthop Trauma Surg.* 2018;138(3):419-425. doi:10.1007/s00402-017-2847-z