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Clinical Importance of Lymphatic Territories with Special Reference to Mammary Glands and Uterus in Canine: A Review

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ABSTRACT

The lymphatic circulatory system consists of two major components *viz.,* the lymphatic vessels and nodes. This system plays important role in immune surveillance, lipid absorption and maintenance of tissue fluid balance. The lymphatic vasculature also serves as the primary route for metastatic spread of tumour cells to regional lymph nodes. Swelling of lymph node usually indicates the existence of a disease process in its tributary territory. Medial retropharyngeal lymph node serves as the collecting centre of the head region. The buccal lymph node is only found in a minority of dogs mainly larger purebreds. The axillary lymph centre is involved in vascularisation of the thoracic limb and first three mammary glands considered during mammary gland surgery. The popliteal lymph node is the important lymph centre found in hindlimb provides valuable data about health status of canine. The heterogeneity and plasticity of the lymphatic drainage of mammary glands is common in the bitch. Sentinel lymph nodes of uterus might be used to reach an early diagnosis of uterine diseases. There is great individual variability within the lymphatic system due to presence or absence of some lymph nodes. In this review we summarize the clinical importance of lymphatic territory and anatomical position of some important lymph nodes in dog.

Keywords: Dogs, lymph node, lymphatic territory, vascular system

In mammalian body, there are two major circulatory systems; the blood vascular system and lymphatic system. The lymphatic system is as essential as blood vascular system for individual health and wellbeing. The lymphatic system is a linear network of lymphatic vessels and secondary lymphoid organs. The lymphatic capillaries form plexuses in most of the body tissue from which larger lymphatic vessels take origin. These larger vessels closely resemble veins in structure and are easily identified as closely spaced valves, give them a distinctive beaded appearance when they are well filled (Dyce *et al.,* 2010). Lymphatic vessels can be found in all of the vascularized organs and tissues except retina, brain, bone and tissues like without blood supply *viz.*, cartilage, cornea and epidermis (Stan *et al.*, 2010; Choi *et al.*, 2012). All lymph passes through at least one lymph node in its passage from the tissues to the blood stream except very few (Konig and Liebich, 2014). The primary function of lymphatic vessels is to transport excess interstitial fluid and macromolecules from tissues back to the blood circulation via the thoracic duct (Charman and Stella, 1992). Few life-threatening diseases

result from malfunction of the lymphatic vasculature and failure of lymph transport promotes lymphedema - a disfiguring, disabling and occasionally life threatening disorder (Johnson and Oliver, 2009). The lymphatic vasculature plays essential roles in tissue fluid balance, immune defense, metabolism and cancer metastasis through the lymph nodes located on the route of the lymphatic vessels (Pepper and Skobe, 2003; Wang and Oliver, 2010).

The lymph node is the structural and functional unit of the lymphatic system. Lymph nodes produce lymphocytes, filter lymph, phagocytose foreign material and form antibodies (Banks, 1993). It provides a barrier to the spread of infectious agent and also facilitates the metastasis of cancer cells. Good understanding of local anatomy and lymphatic drainage patterns, peripheral lymph node extirpation (lymphadenectomy) can be easy method for providing valuable information for treatment, recommendations and prognosis in these patients (Wright and Oblak, 2016). Lymphatic drainage therapy offer myriad benefits of patients facing surgery and treatment of lymphedema (Bruno and Alaya, 1997). Moreover, it is essential to understand lymphatic pathways of certain regions of the body. Therefore, aim of present review is to explore the position, accessibility, drainage territory and the destination of the efferent flow of all major nodes in canines especially in dogs.

GENERAL ANATOMY OF LYMPH NODES

Lymph nodes are usually ovoid or bean-shaped and vary in size from a millimetre to several centimetres. Some lymph nodes are superficial and can be identified on palpation through the skin. Microscopically, lymph node is divided into a cortex and a medulla. The cortex contains the germinal centers in which lymphocytes continuously produced. The medulla consists of branching trabeculae of reticular fibres and cells surrounded by medullary sinuses and lymph capillaries. The free cells include lymphocytes, plasma cells and macrophages. Each node is bounded by a capsule from which septa and trabeculae extend into the organ to form an internal framework. Afferent vessels enter a lymph node at capsule and empty into the subcapsular sinus. The region of lymph node contains efferent lymphatics and blood vessels are referred to as hilus. Lymph flows through lymph node is unidirectional from capsule to hilus (Banks, 1993).

Groups of neighbouring lymph nodes occuring constantly in specific regions of the body and receive afferent vessels from approximately the same region in most species are known as lymphocentres. There are important species-specific differences in the lymphocenters. Lymphocentres of carnivores and ruminants contain fewer but individually large lymph nodes, while those of pigs and horses contain a large number of relatively small lymph nodes (Konig and Liebich, 2014).

LYMPHATIC DRAINAGE OF THE SUPERFICIAL LYMPH NODES AND ITS CLINICAL IMPORTANCE

Due to the importance of the dog as a companion animal and as a transplantation model, its immune system has been the subject of many studies (Fujita *et al.*, 1972; Fossum and Ford, 1985). The difference exists in different species of the lymphatic pathways associated with its lymph nodes and suggests that the information available may not apply equally to all animals (Belz and Heath, 1995). Therefore, the applied importance of lymphatic drainage, its organization and palpable lymph nodes in dog has been presented here for better understanding of canine model (Table 1).

The Lymph Nodes of the Head and Face Region

(a) Parotid lymph centre: The parotid centre consists of one or more nodes placed on the masseter close to the temporo-mandibular joint

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Table 1: Anatomical position of some important lymph nodes in dog

Name of Lymph Node	Anatomical Position
Parotid lymph node	⊙ One or more parotid lymph nodes are present. ⊙ Under the rostro-dorsal border of parotid gland. ⊙ Cranial to the base of ear. ⊙ Caudal border of *masseter m.*
Mandibular lymph node	⊙ 2 to 3 mandibular lymph nodes are present. ⊙ Placed around the facial vein and near the angle of the mandible.
Medial retropharyngeal lymph node	⊙ Medial to the mandibular gland and *sternomastoideus m.* ⊙ Dorso-laterally on the pharynx. ⊙ Ventro-medial to the wing of atlas.
Lateral retropharyngeal lymph node (If present)	⊙ Caudal border of the parotid and mandibular glands.
Superficial cervical (Prescapular) lymph node	⊙ Lies in-front of the shoulder, under cover of the lateral superficial muscles of the neck.
Buccal lymph node (If present)	⊙ It always placed dorsal to the *zygomatic m.* and rostral to the *masseter m*. in the region where the superior labial vein drains into the facial vein.
Axillary lymph node	⊙ Within the axilla on the medial muscles of the shoulder.
Popliteal lymph node	⊙ In popliteal space caudal to stifle joint. ⊙ Between the *biceps femoris* and *semitendinosus* muscles at the proximal insertion of the gastrocnemius muscles. ⊙ Placed on the distal caudal femoral artery.

and commonly covered by the parotid gland. This lymph node is not always palpable. These nodes receive lymph from dorsal structures of the head including skin, the dorsal bones of the skull, the contents of the orbit and the masticatory muscles (in part). Belz and Heath (1995) reported 1 or 2 efferent vessels emerge from parotid lymph node and these forms afferents to the medial retropharyngeal lymph node.

(b) Mandibular lymph centre: The mandibular lymph nodes are the easiest to palpate and subsequently the easiest to surgically remove (Wright and Oblak, 2016). It comprised of groups of 2 or 3 and occasionally up to 5 oval lymph nodes. It lies in two groups immediately dorsal and ventral to the facial vein. The dorsal mandibular lymph node is typically flattened, 3-sided, and approximately 10 mm long in the dog. The ventral mandibular lymph node is typically long and ovoid, size 10 mm x 20 mm and flattened transversely (Belz and Heath, 1995). It drains lymph from lateral surface of the head and from the facial node when present.

(c) Retropharyngeal lymph centre: The retropharyngeal lymph centre may consist of both medial and lateral retropharyngeal lymph nodes (Schummer & Nickel, 1979), but lateral retropharyngeal lymph nodes are not always present in dogs. Belz and Heath (1995) dissected 24 dogs and reported lateral retropharyngeal lymph nodes only in 2 dogs. The medial retropharyngeal lymph node lies dorso-laterally on the pharynx, caudal to the digastric muscle and ventro-medial to the wing of the atlas. Medial retropharyngeal lymph nodes in dogs and cats receive all lymph draining from tissues throughout the head, including the tonsils along

with lymphatic vessels to the facial, parotid, lateral retropharyngeal and mandibular lymph nodes (Evans, 1993). Its efferents form the tracheal lymph trunk. Based on lymphatic drainage patterns, the medial retropharyngeal lymph node may provide information regarding regional metastasis of oral neoplasm (Belz and Heath, 1995). These lymph nodes cannot be palpated when normal in size and are one of the most challenging types of lymph nodes to identify surgically (Wright and Oblak, 2016).

(d) Buccal lymph centre: The buccal lymph node is only found in a minority of dogs and can be unilateral, bilateral or absent. Shelton and Forsythe (1979) studied 250 dogs of various breed and found buccal lymph nodes unilaterally in 11 dogs and bilaterally in 11 dogs (observed prevalence 8.8 %). Rumph *et al.* (1980) examined 171 Grey hounds dog and reported presence of buccal lymph nodes only in 15 dogs (8.7%). Casteleyn *et al.* (2008) found 6 bilateral and 7 unilateral buccal lymph nodes in larger purebred dogs out of 150 dogs of various breed that they examined. Authors observed that buccal lymph node always placed rostral to the masseter muscle and dorsal to the zygomatic muscle in the region where the superior labial vein drains into the facial vein. Moreover, no buccal lymph node was found in small or medium sized breeds and in the all 41 mongrels examined in this study. Similar to dogs, in other species *viz.* guinea pigs, primates and camels buccal lymph nodes are also found inconsistently (Barone *et al.*, 1950; Spira, 1962; Grau, 1974). In contrast to this, it is always present in humans and rabbits (Grau, 1974 and Barone, 1996). Scarce information is available about the drainage area of the canine buccal lymph node. It drains lymph from facial structures including the nose, upper lip and buccal region and efferents of buccal lymph node, which discharge lymph into the mandibular lymph nodes (Shelton and Forsythe, 1979). The canine buccal lymph node is important clinically as it can enlarge due to tumour metastasis or inflammation of the buccal region (Casteleyn *et al.*, 2008). Thus, clinicians should be aware of its existence while dealing with nodular buccal swelling in clinical cases.

The Lymph Nodes of the Neck

The superficial cervical (prescapular) lymph node centre lies infront of the shoulder under cover of the superficial muscles of the neck. It comprised of one or more nodes that drain a very wide but predominantly superficial territory. The majority of these lymph nodes are oval in shape, somewhat flattened and approximately 30 mm long by less than 10 mm thick (Wright and Oblak, 2016). The deep cervical centre comprises a chain of nodes, occasionally found in the vicinity of the thyroid gland and the cervical portion of the trachea (Dyce *et al.,* 2010). These nodes are packeted in cranial, middle and caudal groups but often irregular in disposition. They mainly drain deeper and more ventral structures of cervical region. They send their efferents cranio caudally to others in the chain and then the thoracic duct, tracheal trunk or cranial mediastinal lymph node.

In the neck, commonly only the superficial cervical lymph nodes are excised as the deep cervical lymph nodes are deeply located, variable in number and position, small in size, and require a much more extensive dissection for removal (Wright and Oblak, 2016).

The Lymph Nodes of the Forelimb

The axillary lymph centre is one of the important lymph centre being involved in vascularisation of the thoracic limb and first three mammary glands, which is unanimously accepted by many authors. This lymph centre reflects health status of the mammary gland (guard lymph nodule) and considered when surgical removal of mammary gland is performed. This lymph centre consists of two big lymphoid groups; their own axillary lymph nodules and accessory axillary lymph nodules (Stan *et al.,* 2007). The

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axillary nodes are located within the forelimb on the medial muscles of the shoulder and accessory nodes may be found in relation to the first rib or more caudally on the chest wall. Stan *et al.* (2007) reported accessory axillary nodules in 4 out of 12 dogs examined with mentioned that axillary lymph nodules were bigger in size than in cases where the accessory nodules are present.

The Lymph Nodes of the Hindlimb

The popliteal lymph node centre is composed of the superficial popliteal lymph node of the hindlimb only. These lymph nodes are most frequently single but in rare cases can be double. They are consistent in position located in the subcutaneous fat deposit between the medial border of the biceps femoris and medial border of the semitendinosus muscle where the muscles diverge from each other. This oval-shaped lymph node is considered the largest lymph node of the pelvic limb and is approximately 20 mm long. The popliteus lymph node in dogs displays a rich vascular network; which accounts for the fact that it is seen as "a clinical sentinel organ"; of greatest importance in both clinical and semiological practice (Stan *et al.,* 2008).

LYMPHATIC DRAINAGE OF THE MAMMARY GLANDS

The heterogeneity and plasticity of the lymphatic drainage of mammary glands is reported by various authors in the bitches (Fig. 1 A & B). There are also variations in drainage among individuals. The lymphatic drainage of the mammary gland in bitches is considered for tracing the possible spread of tumour cells and implies for surgical removal of the mammary tumour along with the gland associated lymph nodes. In addition, drainage of the mammary gland may be altered in cases

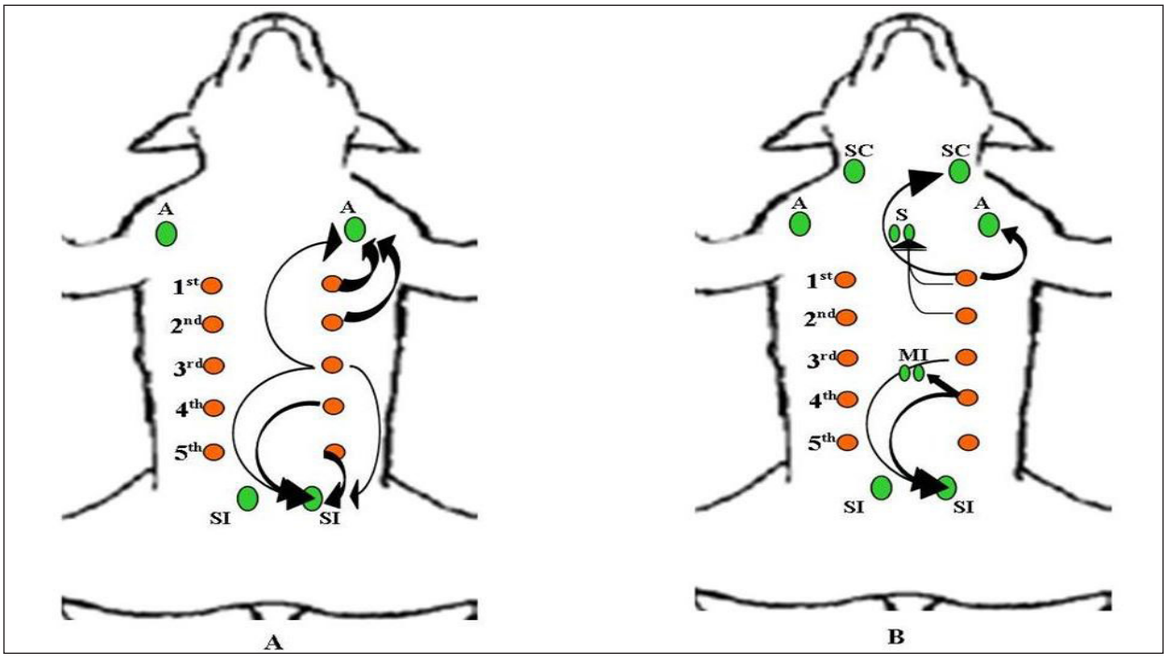

Fig. 1: (A) Schematic representation of the usual pattern of the lymph drainage from the 1st, 2nd, 3rd, 4th and 5th mammary glands in the bitch (A-Axillary nodes; SI- Superficial inguinal nodes). **(B)** Schematic representation of the rare pattern of the lymph drainage from the 1st, 2nd, 3rd and 4th mammary glands in the bitch (A-Axillary nodes; SC- Superficial cervical nodes; MI- Medial iliac nodes; S- Sternal nodes; SI- Superficial inguinal nodes)

of mammary tumours are sometime known to alter the lymphatic drainage pattern by forming new draining channels and recruiting a large number of lymph nodes (Pereira *et al.,* 2003; Patsikas *et al.,* 2006).

Patsikas and Dessiris (1996a) demonstrated lymph drainage in mammary glands of normal lactating mongrel bitches. They observed that the first gland usually drained by axillary nodes and in few cases to the axillary and superficial cervical nodes simultaneously (Fig.1 A & B). The second gland drains to the axillary nodes. The fourth gland usually drains to the superficial inguinal nodes but it may rarely drain to the superficial inguinal and medial iliac nodes simultaneously. The fifth gland drains to the superficial inguinal nodes. In another study, Patsikas and Dessiris (1996b) investigated lymph drainage of the 3rd mammary gland and stated that lymph usually drains from the third gland to the axillary and superficial inguinal nodes simultaneously, but in some cases it drains only cranially to the axillary nodes (Fig. 1 A & B). It may rarely drain only caudally to the superficial inguinal nodes. In these studies, authors not observed lymphatic connection between the mammary glands, however, it was demonstrated that lymph can pass from one gland to another, through their common regional lymph nodes by retrograde flow. It was established that there is a connection between the superficial inguinal lymph nodes from either side. Furthermore, Lymphatics of the mammary glands that cross the midline were not confirmed. Somewhat similar findings were reported by Stan *et al.* (2010) in 9 bitches of the common breed. Authors observed that thoracic mammary glands (T1 and T2) and in some cases, cranial abdominal mammary gland (A1), are drained by the axillary or cranial sterna lymph nodes or even by the superficial cervical lymph nodes. The caudal (A2) and inguinal (I) abdominal mammary glands are drained by the superficial inguinal lymph nodes. There are cases where gland (A2) has a dual drainage, cranial into axillary lymph nodes and caudal toward the superficial inguinal lymph nodes, Similarly, Shida *et al.* (2012) also found individual variations of the lymphatic drainage pattern in intact female beagles showing once again the heterogeneity and plasticity of the lymphatic system in the bitch.

LYMPHATIC DRAINAGE OF THE UTERUS

The assessment of sentinel lymph nodes of uterus is considered for early diagnosis and prognosis of uterine disorders in bitch. The lymphatic system of this region is important for maintenance of homeostasis of the uterus and also facilitate the metastatic spread of tumour cells to regional lymph nodes in case of uterine cancer, however, the occurrence of uterine tumours are rare in bitch (Klein, 2007), as compared to high incidence of uterine cancers in humans. Some type of tumours like hemangiosarcomas, adenocarcinoma and leiomyosarcoma have been reported in bitches (Tivers *et al.,* 2009; Cave *et al.,* 2002; Serin *et al.,* 2010). Furthermore, cases of infection and inflammation of canine uterus are very common (Fontaine *et al.,* 2009). Several new lines of studies revealed that inflammation triggers lymphangiogenic signals (Kerjaschki *et al.*, 2006; Flister *et al.,* 2010). The lymphatic vasculature regulates inflammatory responses by transporting leukocytes and antigen-presenting cells from the site of inflammation to the lymph nodes and other secondary lymphoid organs (Wang and Oliver, 2010). The uterus is managed to prevent serious pathological processes, if good health status of patient, lymphatic drainage and resorption. Therefore, knowledge of the pattern of lymph drainage to regional lymph nodes might be used to reach an early diagnosis of uterine diseases. Only few studies have been conducted to gain the accurate knowledge of lymphatic drainage pattern of the uterus in canines. In one animal experiment, Wang *et al.* (2001) observed that the common iliac nodes and lumbar nodes became green within 5 min after chlorophyllin dye was injected into the canine uterus. In another

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study, Justino *et al.* (2014) demonstrated that the medial iliac lymph node chain is the sentinel chain of the uterine horns of bitches.

CONCLUSION

Increased knowledge of lymphatic vascular function, associated diseases, identification of lymphatic territories in different species and generation of valuable animal models could facilitate the design of therapeutic strategies against cancer, inflammatory conditions and some metabolic diseases.

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