Lymph node aspirates

Sandra Forsyth of SVS Laboratories outlines how to aspirate lymph nodes with a light touch.

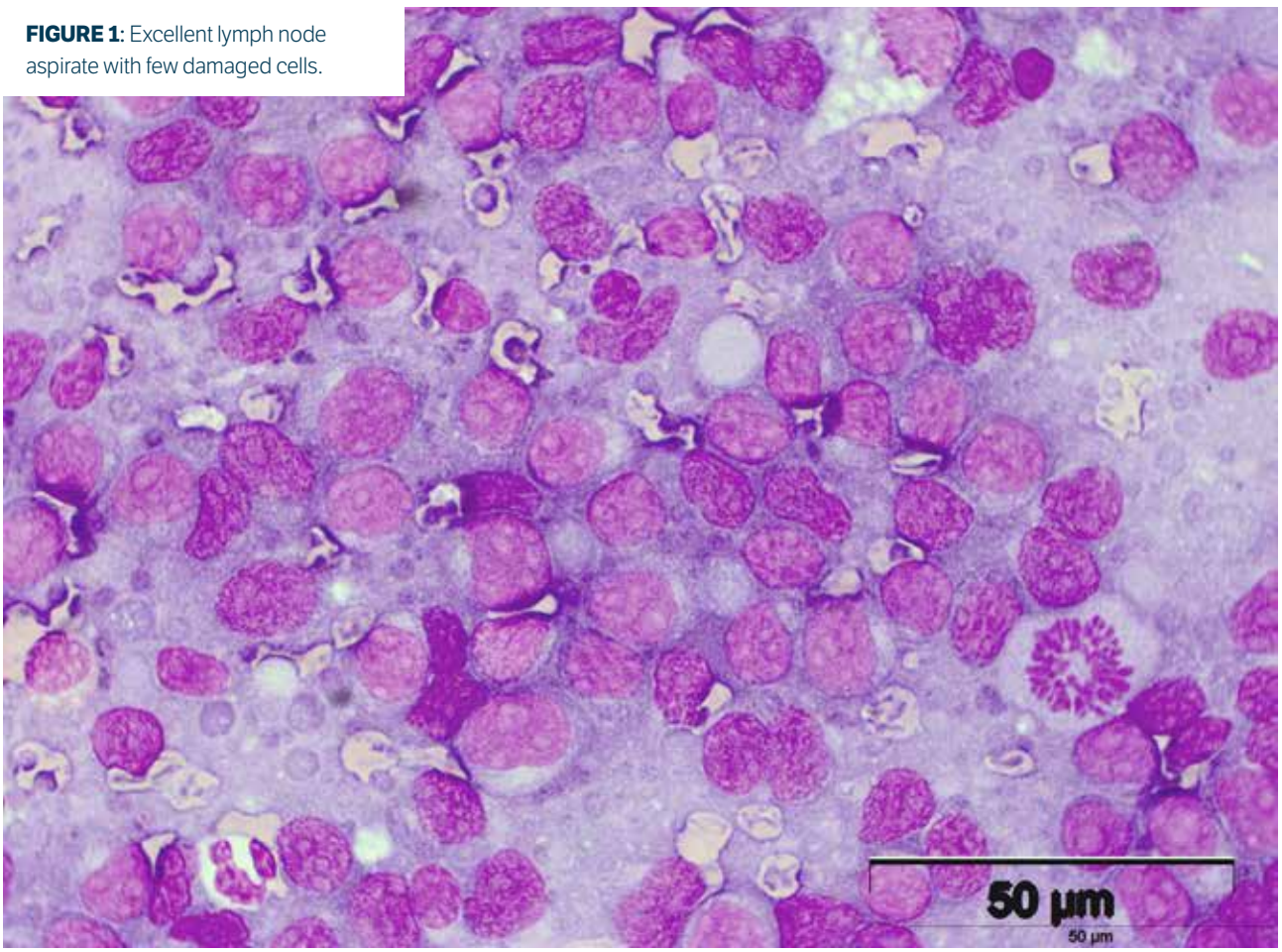
LYMPH NODE ASPIRATES are commonly submitted to the lab for cytological examination, usually because the nodes are enlarged, or to check for metastatic disease.

ASPIRATING ENLARGED NODES

When aspirating lymph nodes it is very easy to obtain poor preparations, with many cells damaged beyond recognition because lymphocytes are fragile, particularly neoplastic lymphoid cells.

To manage the delicacy of these cells successfully, we need to treat them tenderly to ensure that useful cytological preparations are obtained (Figure 1). Attention to handling begins by using gentle aspiration or, even better, using a non-aspiration technique. Unlike epithelial and mesenchymal cells, lymphocytes do not adhere to other cells and are not embedded in matrix, so moving the needle sharply forward at various angles will often provide plenty of cells. The additional benefit of a non-aspiration technique is that blood contamination is reduced. Using the non-aspiration technique is worth a shot, but if no cells are collected then aspiration with a light touch is the next step.

FIGURE 1: Excellent lymph node aspirate with few damaged cells.



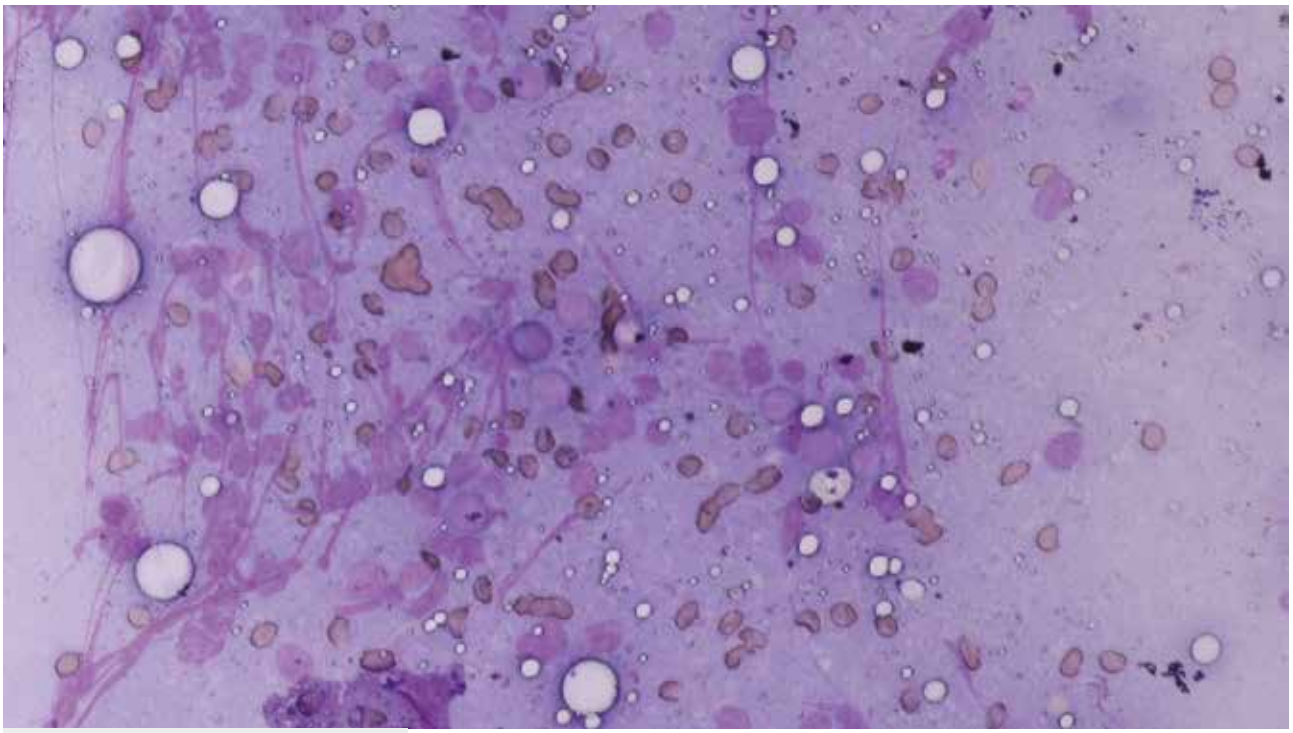


FIGURE 2: Damaged lymphocytes showing strands of nuclear material as a consequence of over-vigorous aspiration.

Cells can also readily lyse if they are vigorously squirted onto a glass slide, and it is much better to ooze them out of the needle hub by placing the tip of the needle on the slide before depressing the syringe plunger. When too many damaged cells are present, a cytological diagnosis cannot be made (Figure 2).

As said, lymphocytes are readily aspirated, which means that large numbers of cells can be collected and thick smears are a potential consequence. If cells are packed like sardines it can be impossible to make out cytological features, and a diagnosis may not be forthcoming. It is better to make a large, thin smear than a small, thick one. When spreading cells by using the slide-over-slide technique, simply allow the top slide to rest on the bottom slide; don't force them together, as this can also damage cells.

Even using the best technique there is always at least some cell damage, and for

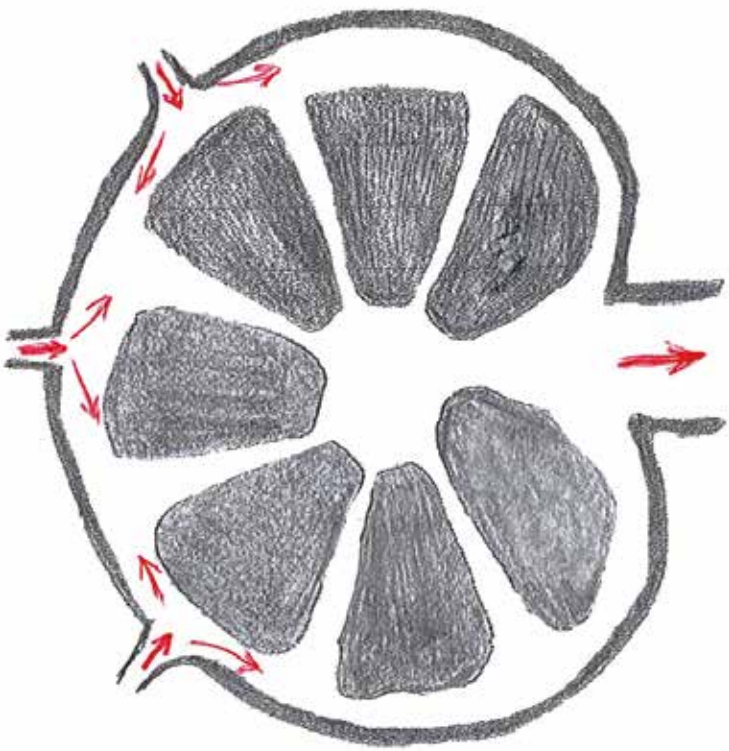


FIGURE 3: Flow of lymph from afferent vessels into the subscapular space, and eventually out through the single efferent vessel.

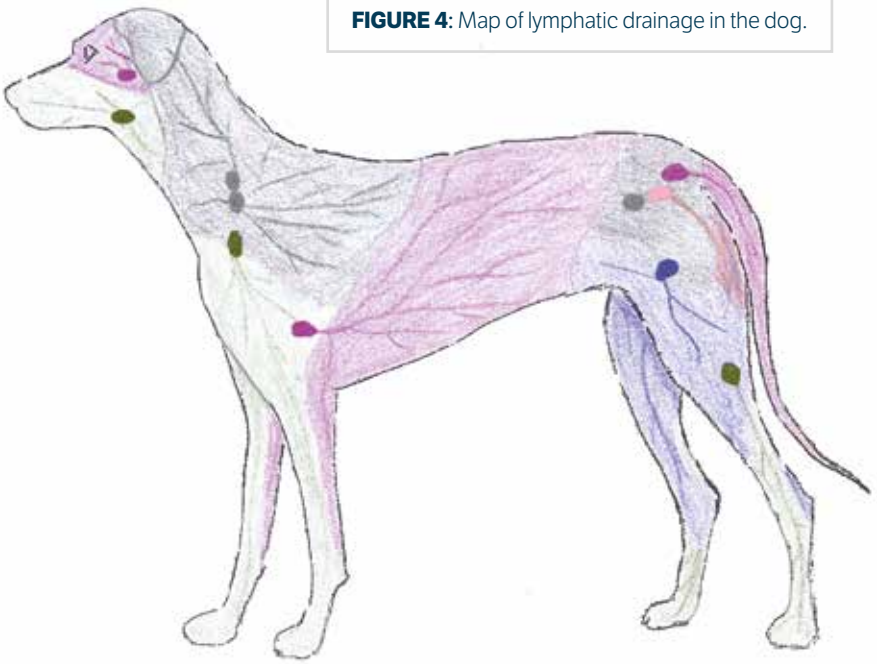


FIGURE 4: Map of lymphatic drainage in the dog.

IT IS SURPRISING THAT LYMPH NODE ASPIRATION FOR TUMOUR STAGING IS CARRIED OUT AS INFREQUENTLY AS IT IS, **BECAUSE IT IS AN EASY, RELATIVELY NON-INVASIVE, INEXPENSIVE METHOD FOR LOOKING FOR METASTATIC DISEASE.**

this reason immunocytochemistry is not as useful as immunohistochemistry for differentiating B and T cells, as cells need to be intact for an accurate diagnosis.

LYMPH NODES AND METASTATIC DISEASE

Lymph nodes consist of multiple lymphoid follicles surrounded by lymph-filled sinuses, all enveloped by an outer capsular membrane. Afferent lymphatic vessels deliver lymph to the node along its outer border. The lymph spreads through the subcapsular sinus, flows down the transverse sinuses and into the medullary sinuses, to leave via a single efferent vessel (Figure 3).

Metastatic cells enter the node through the afferent vessels and spread through the subcapsular space, initially being prevented from moving into the transverse sinuses. Consequently, metastatic cells 'lodge' in the subcapsular space and begin to proliferate there. From a clinical point of view, this means that when carrying out fine-needle aspiration (FNA) for a 'met check' the ideal spot for the needle is immediately under the capsule, rather than the centre of the node.

It is surprising that lymph node aspiration for tumour staging is carried out as infrequently as it is, because it is an easy, relatively non-invasive, inexpensive method for looking for metastatic disease. Perhaps there is a perception that samples are non-diagnostic and less useful than biopsy samples, radiographs or ultrasound. However, in a 2001 study by Langenbach et al., sensitivity (100%) and specificity (96%) were shown to be as good as or better than those shown by needle core biopsies, which had a sensitivity of 64% and specificity of 96%.

That said, the study was undertaken at a university hospital where the clinicians were experienced in tissue sampling, and perhaps sensitivity would be lower in samples taken by non-specialists.

The utility of lymph node aspiration was further reflected in a 2014 study by Warland et al., which determined that dogs with cutaneous mast cell tumours did not develop distant metastatic disease without showing regional lymph node metastasis first.

Sampling of the node was carried out via FNA, and it was found that if the local node was clear of mast cells, then further staging such as thoracic radiography, abdominal ultrasonography, buffy coat examination and bone marrow preparations provided no or little further benefit.

Naturally, this means that the correct regional node needs to be aspirated, several areas of the node need to be sampled as outlined in the section above, and the subcapsular area should be the primary site of sampling. Figure 4 provides a guide to the drainage areas of superficial lymph nodes in the dog.

CONCLUSION

With appropriate sampling and smear preparation, a cytologist will often be able to differentiate inflammatory, reactive, neoplastic and metastatic lymph nodes to provide an accurate diagnosis. VS

REFERENCES:

Langenbach A, McManus PM, Hendrick MJ, Shofer FS, Sorenmo KU. Sensitivity and specificity of methods of assessing the regional lymph nodes for evidence of metastasis in dogs and cats with solid tumours. *Journal of the American Veterinary Medical Association* 218, 1424–8, 2001

Warland J, Amores-Fuster I, Newbury W, Brearley M, Dobson J. The utility of staging in canine mast cell tumours. *Veterinary Comparative Oncology* 12(4), 287–98, 2014