Paws claws and udder things

Gribbles VETERINARY

December 2022

Hidden in fat

KATHRYN JENKINS

A recent interesting case highlighted the issue of a lesion which can mimic the clinical appearance of a benign lipoma.

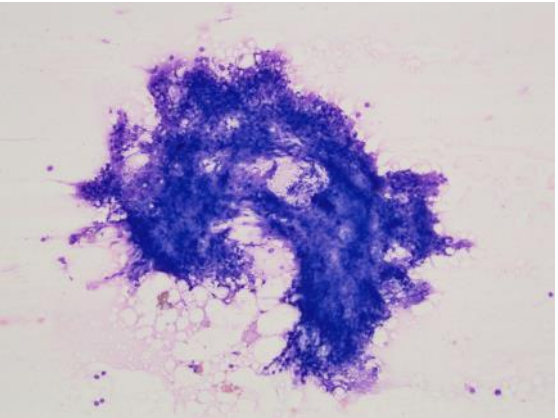
Clinical history:

A 10-year-old dog presented with a 7 cm diameter mass located on the upper forelimb, which had been slowly growing over the last 4-6 months. The mass palpated like a lipoma, and greasy material was noted when making the smears for cytology. Four smears were submitted for pathologist review.

Cytological findings:

Cell preservation was excellent and the cytology comprised several dense clumps of mesenchymal tissue embedded within a large amount of fat. The spindle cells appearing variably vacuolated with mild to moderate nuclear pleomorphism (Figure 1). Given the unusually large amount of stromal tissue, surgical biopsy for histopathology was recommended. This was performed and confirmed a well-differentiated low grade liposarcoma.

Figure 1. Low power. Dense clump of mesenchymal tissue within a fatty background. The finding of spindle cell morphology on cytology often requires surgical biopsy for histopathology for further characterisation.



Discussion:

Liposarcoma is the rare malignant counterpart to a benign lipoma. Liposarcomas can vary in appearance histologically (from well-differentiated, anaplastic, or myxoid variants). And as a result they can also vary in clinical appearance, from mimicking lipomas as in this case, through to presenting as a firm infiltrative mass. Although these tumours are considered rare in veterinary medicine, Shetland sheepdogs may be predisposed. Similar to other soft tissue sarcomas, liposarcoma are locally infiltrative, with distant metastasis less commonly reported.

In contrast, lipomas are a common benign tumour of well-differentiated adipocytes in dogs. These are less commonly found in other species, although Siamese cats may be over-represented. Lipomas are often well circumscribed, unencapsulated, freely moveable soft masses, found most often on the trunk and proximal limbs. They are slow growing expansile masses, often cured by complete excision. Grossly the aspiration of fat appears clear and glistening, with a slide that does not dry completely. On cytology the adipocytes of lipomas are indistinguishable from normal fat, and appear as large round balloon-like cells, with a compressed nucleus to the side of the cell. The cohesive adipocytes often have a characteristic "chicken wire" type appearance on low power (Figure 2).

In addition, a small percentage of lipomas occur as infiltrative forms. These tumours invade adjacent connective tissue and skeletal muscle, appearing marbled in appearance grossly. They are also

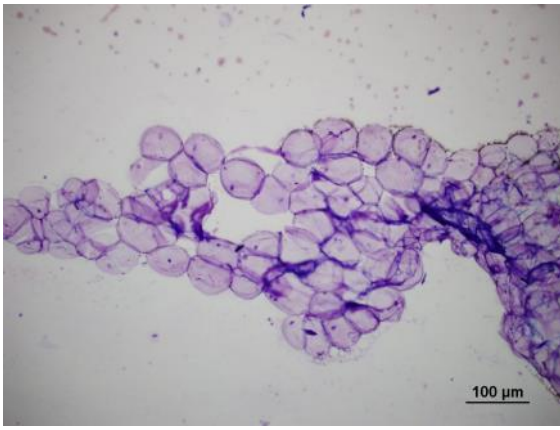


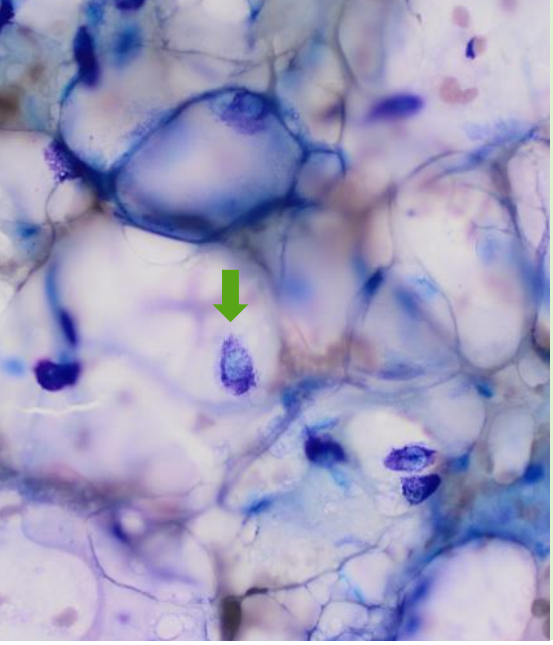
Figure 2. Low power. Well-differentiated adipocytes (fat cells). Incidental aspiration of normal subcutaneous fat appears identical to the cells present in lipoma, with a characteristic "chicken wire" or balloon-like appearance. Note the large size of these cells.

considered benign in behaviour, however they can be more difficult to completely excise.

It is important to recognize that other tumours can mimic the feel and clinical appearance of benign lipomas, and fat can be aspirated surrounding a lesion of a different nature (due to the focal bias of cytology).

Although liposarcoma are considered rare, other types of soft tissue sarcoma (especially perivascular wall tumours), and subcutaneous mast cell tumours, can also mimic a benign lipoma on palpation, and produce fatty material on cytology slides (Figure 3).

In these cases the power of cytology to provide a useful interpretation is improved by sampling multiple areas within the same mass.



We accept up to six smears per site for cytology. The finding of fatty tissue on a single slide could represent aspiration of normal subcutaneous fat, a benign lipoma, or simply fat surrounding a lesion of a different nature, including malignant neoplasia.

Figure 3. High power. Note the numerous large balloon-like adipocytes dominating this image. This mass felt like a lipoma, however several mast cells (arrow) are present on cytology which raises concern for an underlying mast cell tumour.

The moral of the story; even if the mass palpates like a lipoma and has material which appears greasy on the slide, always review the cytology and sample multiple areas within the mass.

Thank you to David Sheehan, CareVets Kilbirnie for this interesting case.

Festive season opening hours

We are open throughout the holiday season but are closed on all the public holidays.

To ensure you receive consumable orders prior to Christmas, please endeavour to have all orders to us by close of business on Wednesday 14 December. We are unable to guarantee pre-Christmas delivery of orders received after this time.

Delays with consumable orders may occur over the Christmas and New Year weeks due to the restricted courier service and reduced staffing levels. Please ensure any urgent orders sent during this period are clearly marked as such.

Some referral laboratories will also be closed during the Christmas holiday period, which may affect testing turn-around-times. Please contact the laboratory (0800 GRIBBLES) if you have any concerns.

CHRISTMAS HOURS

Saturday 24 December - OPEN
Christmas day - CLOSED
Monday 26 December - CLOSED
Tuesday 27 December - CLOSED

We will be open normal working hours 28-31 December.

NEW YEAR HOURS

Saturday 31 December - OPEN
New Year's day - CLOSED
Monday 2 January - CLOSED
Tuesday 3 January - CLOSED

We will be open normal working hours from Wednesday 4 January.

Lab tip - keep cool when it's hot!

We're heading into warmer weather, so please remember to keep samples cool after collection.

Traditionally as summer heats up, we notice a deterioration in the quality of some samples received at our laboratories. Some even arrive in an unsuitable condition for testing.

We recommend you include an ice-pack of some description with your samples if you are sending them to the laboratory via courier. We are happy to return the ice-packs to you with your shipping containers.

The main exceptions to the "keep it cool rule" are blood/cytology smears, histology samples and faeces for larval culture. Please leave these at room temperature prior to transport.

See this 'How To' guide on our website for more information.

Update on the prevalence of anthelmintic resistance

SARAH RIDDY

With the routine use of Gribbles Veterinary's automated faecal egg count reduction test (FECRT) report system, we are pleased to announce the resumption of our annual update on anthelmintic resistance. This report details the analysis of data from fully differentiated FECRTs submitted to the Gribbles network for the season November 2021 to May 2022.

Data was obtained from submissions that clearly identified the test anthelmintic and sample groups with ≥10 animals per treatment group. Data at genus level required a pre-treatment of 50 epg per genus to be included. All other test requirements and methodology were as described by McKenna (2018). The final data set contained 3,272 data points collected from 166 FECRT submitted over this period. 1,811 data points were excluded due to insignificant numbers of a genus in a FECRT (<50 epg in the pre-FECRT). A number of factors contribute to this including the season the FECRT was performed, prevalence of different genus on different farms and regions across New Zealand, and egg output of the genus present.

Twenty five data points were excluded due to the test anthelmintic not being identified. 215 data points were excluded due pre-FECRT case numbers not being identified on the post -FECRT submission form. Submissions that did not clearly identify they were FECRT were not included in the dataset. Submissions where part of the FECRT was performed in-clinic and only the larval culture being performed at Gribbles, were also excluded from the data set due to lack of compliance with the laboratory's accreditation and quality procedures.

These results are from 64 cases from the North Island and 102 cases from the South Island, and are presented in Table 1. The percentage of resistance for single active anthelmintic remains relatively similar to those reported in the 2018 update (McKenna 2018). Of note is the increase of double and triple combination anthelmintic treatment groups submitted in this update in comparison to that in 2018. For Teladorsagia the prevalence for triple active resistance in 2016-17 to 2021-22 increased from 6% to 18%, for Benzimidazole/Levamisole (BZ/LEV) dual increased from 18% to 36% and for Abamectin/Levamisole (ABA/LEV) dual increased from 3% to 30%. For Trichostrongylus, the increase in prevalence from 2016-17 to 2021-22 for triple actives* increased from 1% to 33%, BZ/LEV dual from 13% to 42% and ABA/LEV dual from 3% to 29%.

Table 1: The prevalence of anthelmintic resistance identified in sheep nematodes by fully differentiated faecal egg count reduction tests (FECRTs) undertaken on case submissions to Gribbles Veterinary during 2021-2022 (n=3272)

PARASITE	BZ	LEV	IVE	ABA	MOX	BZ/LEV	LEV/ ABA	DERQ/ ABA	ABA/ OXF	ABA/ MONE	TRIPLE
Cooperia	10/60 (17%)	1/76 (1%)	0/5 (0%)	10/49 (20%)	5/32 (16%)	0/8 (0%)	10/167 (6%)	0/45 (0%)	0/13 (0%)	0/17 (0%)	1/115 (1%)
Haemonchus	3/22 (14%)	0/35 (0%)	0/5 (0%)	0/21 (0%)	0/16 (0%)	0/5 (0%)	0/79 (0%)	0/23 (0%)	0/7 (0%)	0/10 (0%)	1/58 (2%)
Nematodirus	32/50 (64%)	13/64 (20%)	0/3 (0%)	3/46 (7%)	4/21 (19%)	1/9 (11%)	8/114 (7%)	0/40 (0%)	0/10 (0%)	0/7 (0%)	4/93 (4%)
Oesophagostomum / Chabertia	6/51 (12%)	0/59 (0%)	0/3 (0%)	0/39 (0%)	0/15 (0%)	0/6 (0%)	3/104 (3%)	0/36 (0%)	0/11 (0%)	0/16 (0%)	0/78 (0%)
Teladorsagia	42/80 (53%)	48/100 (48%)	5/6 (83%)	31/70 (44%)	14/35 (40%)	4/11 (36%)	53/177 (30%)	4/63 (6%)	6/22 (27%)	2/27 (7%)	26/144 (18%)
Trichostrongylus	43/84 (51%)	36/104 (35%)	0/6 (0%)	17/75 (23%)	15/39 (38)	5/12 (42%)	57/195 (29%)	3/68 (4%)	5/23 (22%)	2/28 (7%)	53/160 (33%)

(Benzimidazole BZ, Levamisole LEV, Ivermectin IVE, Abamectin ABA, Moxidectin MOX, Derquantel DERQ, Monepantel MONE, Oxfendazole OXF, TRIPLE includes several brands with 3 actives in combination)

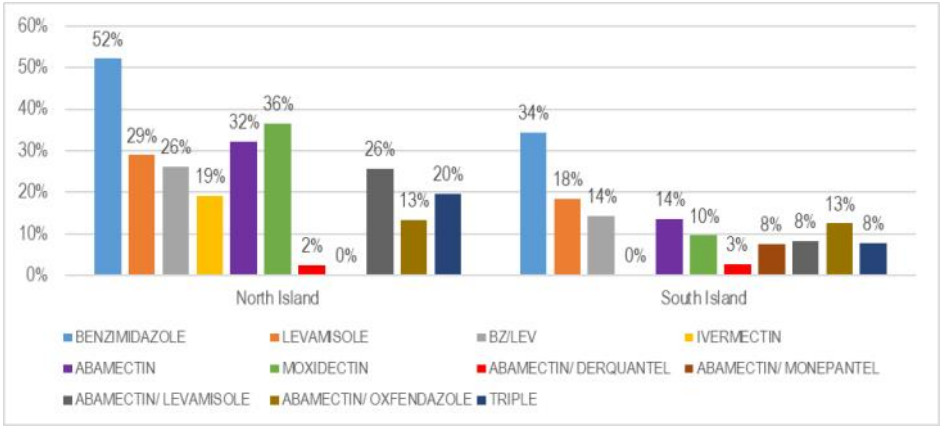


Figure 1: Prevalence of resistance to anthelmintic recorded in sheep FECRTs submitted to Gribbles Veterinary laboratories during 2021-2022 stratified to North Island and South Island (n = 3272).

It is noted by the author that a number of these single actives and one of the dual combinations reported are not available commercially at present. The data collected by Gribbles is done passively through submissions to the laboratory and relies on the information supplied by the submitting veterinary. Incorrect recording of the test anthelmintic, use of older stock, or off-licence use of products may explain these abnormalities.

Stratification of cases by geographical location for both anthelmintic and genus for the percentage of resistance can be found in Figure 1 (previous page) and Figure 2. The corresponding data regarding total data points analysed (including both resistant and susceptible nematodes) can be found in Tables 2 and 3.

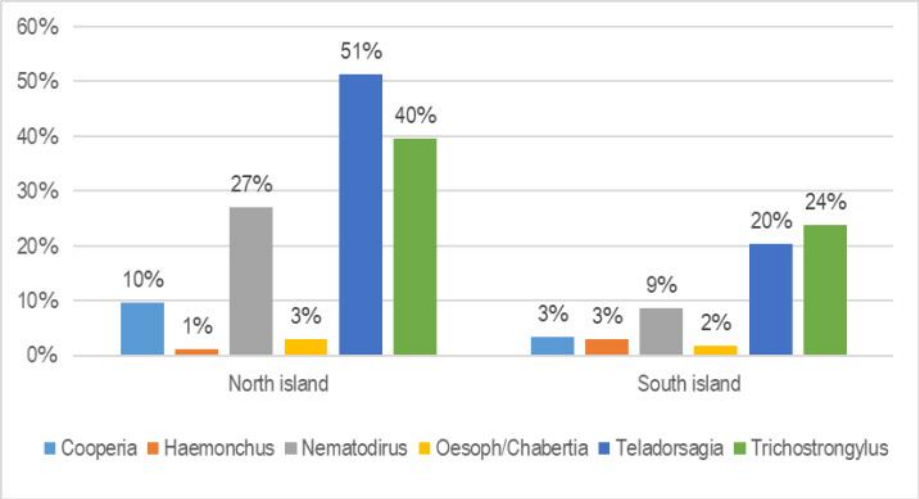


Figure 2: Prevalence of resistance to anthelmintic to genus level recorded in sheep FECRTs submitted to Gribbles Veterinary laboratories during 2021-2022 stratified to North Island and South Island (n=3272).

Table 2: Total number of nematode data points analysed from sheep FECRTs submitted to Gribbles Veterinary laboratories for the North Island and South Island of New Zealand during 2021-2022 stratified by test anthelmintic.

Anthelmenthic	North Island	South Island
BENZIMIDAZOLE	94	253
LEVAMISOLE	166	272
BZ/ LEVAMISOLE	23	28
IVERMECTIN	26	2
ABAMECTIN	109	191
MOXIDECTIN	85	73
ABAMECTIN/ DERQUANTEL	123	152
ABAMECTIN/ MONEPANTEL	52	53
ABAMECTIN/ LEVAMISOLE	354	482
ABAMECTIN/ OXFENDAZOLE	30	56
TRIPLE	292	356

Table 3: Total number genus data points analysed from sheep FECRTs submitted to Gribbles Veterinary laboratories for the North Island and South Island during 2021-2022 stratified by location.

Genus	North Island	South Island
Cooperia	267	320
Haemonchus	247	34
Nematodirus	137	320
Oesophagostomum/Chabertia	130	288
Teladorsagia	277	458
Trichostrongylus	296	498

Analysis from the FECRT data obtained across the Gribbles Veterinary network for the season 2021-2022 shows a rapid rise in the prevalence of anthelmintic resistance in dual- and triple- active drench categories when compared with the previous report in 2016-2018 (McKenna, 2018). The landscape of anthelmintic resistance is continuously changing, due in part to the variation of active combinations available and in use. Gribbles Veterinary is therefore committed to providing this analysis report on an annual basis to track the prevalence trends on a nationwide level.

Accurate trend reporting is dependent on accurate data. The more FECRT data that Gribbles produces, the more detailed and true this trend reporting can be. In order for results to be considered for this report, the pre- and post-drench FEC and larval cultures must be tested within the Gribbles network of laboratories. This is to ensure the quality of the testing is guaranteed, as all Gribbles locations are IANZ accredited.

Acknowledgements to everyone who contributed to the creation of this report.

Reference:

McKenna PB. Update on the prevalence of anthelmintic resistance. *VetScript* 31: 46–47, 2018.

A collaborative case

CATHY HARVEY & REBECCA ALLAN

Clinical history:

Buddy, a 4-year-old neutered male Border Collie presented with lethargy and an in-house PCV of 12%. There was no free blood in the abdomen and the spleen was normal. He had progressive pancytopenia without regeneration, a single dose of dexamethasone and two blood transfusions, before bone marrow samples and blood smears were submitted to the laboratory.

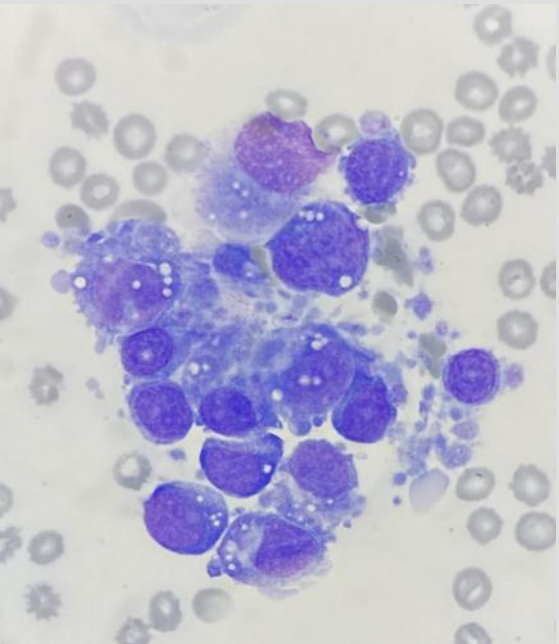
Haematology findings:

In the peripheral blood smears, platelets were clumped but appeared decreased, with occasional large and atypical shaped platelets. There were rare cells containing fine cytoplasmic eosinophilic granules, which may be megakaryocyte/blast in origin. Erythrocytes appeared normal morphologically.

Bone marrow cytology:

The seven smears had low to moderate cellularity and consisted of variably sized aggregates of cells. A 100-cell differential consisted of 8% erythroid cells, 15% myeloid cells and 77% atypical cells, plus low numbers of plasma cells, macrophages and lymphocytes. The atypical cells had a pleomorphic appearance with round to oval nuclei, variably prominent nucleoli and low to moderate amounts of basophilic cytoplasm, variably containing small vacuoles and fine eosinophilic granules, frequently with peripheral cytoplasmic blebs that have the appearance of platelets. These cells were surrounded by platelets, some of which were large and atypical in morphology. Binucleate cells, rare small multinucleated cells, mitotic figures and erythrophagocytic cells were noted in low numbers. Both myeloid and erythroid cells were too few to assess.

Figure 1. Cytology – atypical cells with moderate anisokaryosis, and peripheral cytoplasmic blebs which have the appearance of platelets. These cells are surrounded by platelets, some of which are large and atypical in morphology.



These findings suggested proliferation of pleomorphic cells with features suspicious of megakaryocytic origin.

Bone marrow histopathology:

The samples consisted of blood with scattered islands of adipocytes and haematopoietic tissue. The haematopoietic tissue was cellular but lacked trabeculae. The majority of cells consisted of immature round cells with moderate anisokaryosis, multiple small nucleoli and a small amount of eosinophilic material. Some of the cells had multiple nuclei. There were mitotic figures readily evident. There were rare mature granulocytic cells and no normal mature megakaryocytes. The erythroid line seemed complete, but erythropoiesis appeared reduced.

Histopathology indicated myeloid leukaemia (probably megakaryocyte origin) with hypoplasia or ineffective haematopoiesis of all lines.

Diagnosis:

The large numbers of atypical cells with features consistent of megakaryocytic origin are strongly suggestive of megakaryoblastic leukaemia (acute myeloid leukaemia, AML with megakaryoblastic differentiation); however, other myeloid cell lines couldn't be excluded.

Discussion:

This case was the subject of extensive discussion by anatomic and clinical pathologists throughout our network of laboratories. The cytology and histopathology preparations (slides examined microscopically and also on digital platforms) as well as digital images were examined collaboratively across the Gribbles' pathologist "hive-mind" in multiple geographic sites and multiple disciplines.

Myeloid neoplasms in animals are relatively uncommon, but probably also under

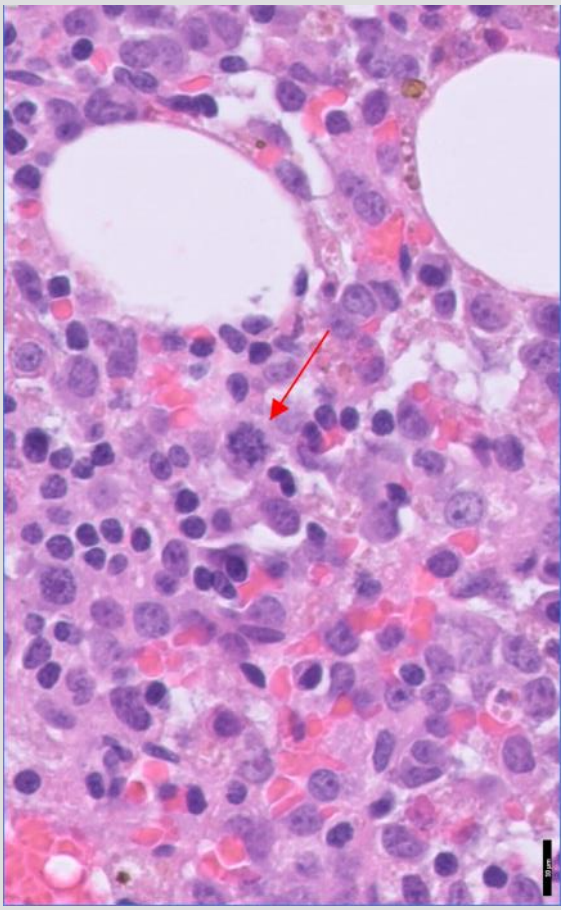


Figure 2. Histology – the majority of cells consist of round to oval cells with moderate anisokaryosis, multiple small nucleoli and small amount of eosinophilic material. Some of the cells have multiple nuclei. There are mitotic figures readily evident (arrow).

diagnosed, because biopsies and bone marrow aspirates submitted to diagnostic laboratories are often of insufficient quality to establish a diagnosis. It is difficult to make a diagnosis from a single snapshot of even a good quality bone marrow biopsy. AML in some instances may be fatal before a bone marrow biopsy has been collected, and animals are not consistently submitted for post-mortem examination.

Myeloid neoplasia should be suspected if an animal has persistent anaemia, thrombocytopenia, or neutropenia without obvious cause such as blood loss, inflammation, or immune mediated destruction of blood cells. The clinician, clinical pathologist and anatomic pathologist have to carefully integrate a thorough history, the haematology results and bone marrow findings to derive a possible diagnosis of myeloid neoplasia.

In order to fully assess the bone marrow both cytological and histological evaluation is necessary. The strength of cytology

evaluation of aspirates is the qualitative assessment of cell features (granulation, vacuolation, haemoglobinization, dysplasia, stain intensity and chromatin patterns), while histology evaluation is essential to quantitatively evaluate overall cellularity, topography, mitotic rate, bony trabeculae and stroma. Identification of cell type is best accomplished by cytology. Histopathology is used to assess overall composition of the marrow and other haemolymphatic organs, but identification of cells is more difficult.

Steps to establish a diagnosis of myeloid neoplasia:

1. Rule out lymphoid neoplasia
2. Complete blood cell count
3. Review of the blood film
4. Aspirate bone marrow for cytology
5. Biopsy bone marrow for histopathology
6. Flow cytometry (currently not available in NZ at this time), or immunophenotyping (only lymphoid markers available in NZ)

The diagnosis is based on:

> Presence and persistence of cytopenia and or persistent increase in one cell type.
> Identification of increased blasts cells in the blood and/or bone marrow and/or myelopthisis and/or dysplasia.
> Subcategorization using immunoassays such as flow cytometry or immunohistochemistry can also be used if available.

AML (acute myeloid leukemia) is defined as cytopenia and > 20% blast cells in the blood or bone marrow. AML most often results from acquired genetic lesions in stem cells. All ages of animals can be affected, but larger surveys indicate that AML is most common in young to middle-aged animals. Large breed dogs may be over represented, in particular German Shepherd dogs. Clinical presentation consists of non-specific illness over days to weeks with ecchymosis due to lack of platelets, exercise intolerance due to anaemia, or fever due to neutropenia. AML is an aggressive neoplasm, generally causing grave illness, poor quality of life and leading to death. However, a multiagent intensive chemotherapy protocol has yielded approximately 2-year survival and quality of life and one dog with AML, and in another study some dogs have survived for greater than a year with single agent chemotherapy suggesting outcomes may not be uniformly fatal.

Buddy is still doing well and is continuing to go to his owner's bach in Franz Josef for weekends.

Many thanks to Rangiora Vet Centre and West Coast Vets Ltd Hokitika for submitting excellent history and samples from this challenging case.

Reference: Tumors of the Hemolymphatic System. Myeloid Neoplasms. In: Donald J. Meuten (eds). *Tumors in Domestic Animals* 5th Edtn. Pp 288-305. John Wiley & Sons, Inc., 2017.

In brief

> **Got Salmonella in dairy cattle?** Please participate in this study to help identify the risk factors for Salmonella outbreaks in dairy. Visit the study website here.

> **Our facial eczema Lab-Portal** is open for business any time you do pasture spore counts. Login or register here and help keep farmers and vets throughout New Zealand up-to-date with pasture conditions.

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