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Thrombocytosis in 715 Dogs (2011–2015)

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Background: Thrombocytosis is a hematologic abnormality in dogs that has been associated with various neoplastic, metabolic, and inflammatory conditions.

Objective: To classify thrombocytosis in dogs based on severity and evaluate whether there are associations between severity and underlying disease processes.

Animals: Seven hundred and fifteen dogs with thrombocytosis and 1,430 dogs with normal numbers of platelets.

Methods: Retrospective study. Medical records of dogs with increased (>500 × 10^3/μL; thrombocytosis group) and normal (300–500 × 10^3/μL; control group) platelet counts between 2011 and 2015 were reviewed. Dogs were characterized by severity of platelet increase and diagnosis. Diagnostic categories included neoplasia, endocrine disease, inflammatory disease, or miscellaneous.

Results: A total of 1,254 complete blood counts with thrombocytosis from 715 dogs were included in the study. Median platelet count in this population was 582 × 10^3/μL (500–1,810 × 10^3/μL). No correlation between severity of thrombocytosis and diagnosis was identified. Causes of secondary thrombocytosis included neoplasia (55.7%), endocrine disease (12.0%), and inflammatory disease (46.6%). Immune-mediated disease was common (22.2%), associated with frequent glucocorticoid administration, and had a significantly higher median platelet count (636 × 10^3/μL [500–1,262 × 10^3/μL] versus 565 × 10^3/μL [500–1,810 × 10^3/μL]) when compared to the other inflammatory processes ($P < 0.001$). The diagnoses in the thrombocytosis dogs differed significantly from the control population ($P < 0.001$).

Conclusions and Clinical Importance: Thrombocytosis is commonly associated with carcinoma and immune-mediated disease in dogs.

Key words: Inflammation; Neoplasia; Platelets; Pseudohyperkalemia.

Thrombocytosis is defined as an increase in number of platelets in peripheral blood and is categorized as being either a primary or secondary process. Primary thrombocytosis is a myeloproliferative disease resulting from increased platelet production and is also called essential thrombocythemia.[1,2] The mechanism for this disease in dogs has not been determined, and diagnosis is based on exclusion of presence of other disorders. In people, primary thrombocytosis is linked to a mutation in the JAK2 protein, which is part of the thrombopoietin signaling pathway, leading to proliferation of megakaryocytes.[3] Additionally, megakaryoblastic leukemia is a cause of primary thrombocytosis in people and is a rare finding in dogs.[4]

Secondary, or reactive, thrombocytosis is more common than primary thrombocytosis in both people and animals. Secondary thrombocytosis occurs due to an underlying process such as neoplasia, inflammation, trauma, or iron deficiency.[1,2,5,6] Neoplasia is the most common cause in several study populations of dogs.[7–9] Thrombocytosis is commonly reported in dogs with hyperadrenocorticism or which have concurrent administration of glucocorticoid, although the mechanism is not well understood.[2,9] Inflammatory disease causes thrombocytosis through a variety of mechanisms. IL-6, a cytokine involved in the acute phase response, increases the production of thrombopoietin (TPO).[10,11] Thrombopoietin is a glycoprotein hormone produced by the liver and is sometimes referred to as megakaryocyte growth and development factor (MGDF) due to its effect on the bone marrow, stimulating megakaryocyte proliferation and platelet production.[12]

Secondary thrombocytosis is seen as a rebound effect after a large number of platelets are lost acutely, as in splenectomy or immune-mediated thrombocytopenia.[13,14] Other associations with secondary thrombocytosis include administration of epinephrine or vincristine.[15] In addition to acute platelet loss, regenerative anemia after erythrocyte loss or destruction is a cause of secondary thrombocytosis.[16] The homology between amino acid sequences of erythropoietin (EPO) and thrombopoietin (TPO) has been proposed as a cause for cross-reactivity in people, but the mechanism is not completely understood.[16,17]

Thrombocytosis is often an incidental finding in animals, whereas in people, thrombocytosis is associated

Abbreviations:

EPO	erythropoietin
TCC	transitional Cell Carcinoma
TPO	thrombopoietin

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with increased risk for hemorrhage or thrombosis, and is used as a prognostic factor in certain disease states.[18,19] Increased platelet mass is proposed to cause shearing within the vasculature, leading to platelet hypersensitivity.[20] Such platelets are easily activated, and their aggregation can lead to microischemic events or depletion of von Willebrand factor multimers, increasing the risk for bleeding or thrombosis.[20] In people, thromboembolic risk is higher in patients with extreme thrombocytosis.[21] Extreme thrombocytosis is defined as a platelet count greater than $1,000 \times 10^3/\mu L$. In a previous study of dogs, extreme thrombocytosis was more commonly associated with a diagnosis of neoplasia, but there was no association with thromboembolic risk.[8] Case reports of thromboembolic complications due to thrombocytosis in dogs are limited, and there is a dearth of research investigating the use of thrombocytosis as a prognostic factor in veterinary medicine.[22–24]

Pseudohyperkalemia is a common finding associated with thrombocytosis. Potassium is greater in serum when compared to plasma due to its release from blood cellular components during clotting.[25] This difference has been noted in dogs, and the magnitude of this difference correlates with platelet number.[25] In cases of thrombocytosis, the serum concentration of potassium is increased when compared to the plasma concentration of potassium, and this difference could be interpreted as clinically impactful.[25–27] Pseudohyperkalemia can be identified through an increased potassium concentration in serum compared to plasma. Whereas the relationship between hyperkalemia and thrombocytosis is well documented in people, it has rarely been investigated in the veterinary literature.[25]

The objective of this study was to characterize the diseases associated with thrombocytosis in a large population of dogs and determine whether the severity of the thrombocytosis could predict the diagnosis.

Materials and Methods

A search of medical records of affected dogs at Purdue University Veterinary Teaching Hospital was completed for the period between January 1, 2011 and December 31, 2015. Dogs were identified based on at least 1 platelet count that was $>500 \times 10^3/\mu L$. Platelet counts were determined using the Abbott Cell Dyn 3700 hematologic analyzer[a] and confirmed by microscopic examination of a blood smear as part of a routine hematologic analysis. Additional information required for inclusion in the study included a complete medical record and a definitive diagnosis.

For dogs meeting the inclusion criteria, medical records were reviewed for signalment, diagnoses, and medications administered including any administration of glucocorticoids, chemotherapeutic agents (including vincristine), antithrombotics or anticoagulants, or oral potassium supplementation. Other complete blood count data including the CBC and leukogram, if available, were recorded. Serum and plasma potassium concentrations, if measured on the same day that the thrombocytosis was identified, were included as well. If a bone marrow aspirate, core biopsy, or both were collected at the time the thrombocytosis was identified, this information was included.

Platelet counts were categorized as mild ($500–600 \times 10^3/\mu L$), moderate ($601–800 \times 10^3/\mu L$), marked ($801–1,000 \times 10^3/\mu L$), or extreme ($>1,000 \times 10^3/\mu L$) thrombocytosis. This classification scheme was extrapolated from previous studies in both humans and animals, in which a cutoff of $\geq 500 \times 10^3/\mu L$ platelets defined thrombocytosis. Diagnoses were grouped into 1 of 5 categories: neoplasia, endocrine disease, inflammatory disease, miscellaneous, or multiple diagnoses. Once categorized, medical record review further identified the definitive diagnosis including the type of neoplasia, the single or multiple endocrine diseases present, and the primary inflammatory process. Inflammatory disease was subcategorized by process and included allergic disease, gastrointestinal disease, hepatobiliary disease, immune-mediated disease, infectious disease, musculoskeletal disease, neurologic disease, respiratory disease, renal/urinary disease, or other. If no underlying diagnosis could be identified, these dogs were considered to have idiopathic thrombocytosis. Data regarding diagnosis were collected from the medical record corresponding to the date that each platelet count was obtained. As some dogs had multiple complete blood counts performed during the time period, dog-specific data regarding diagnoses were determined using the initial visit at which a thrombocytosis was identified.

An additional search of medical records was performed to identify a control population of dogs with a platelet count between 300 and $500 \times 10^3/\mu L$, during the same 5-year span. These medical records were reviewed for signalment and diagnoses. All dogs, thrombocytosis and controls, were arranged by age. Control dogs included in the study were identified and age-matched by selecting 1 control dog on each side of a dog with thrombocytosis. Breed matching was not attempted due to the retrospective nature of the study and the number of mixed breed dogs for which matching would not be effective. Diagnoses were categorized in the same way as described above. If the dog had more than one complete blood count performed during the searched time period, only the initial complete blood count per dog was included.

Numerical variables were assessed for normality using the Shapiro-Wilk test. Due to the nonparametric distribution of the data, comparisons between 2 groups of numerical data were conducted using the Wilcoxon rank sum test, and between more than 2 groups using the Kruskal-Wallis equality-of-populations rank test. Correlation was assessed by calculation of the Spearman rank correlation coefficient (rho). Comparisons in proportional distributions of categorical data were conducted using the chi-square test of independence, or Fisher's exact test when more than 25% of expected frequencies were <5. In analyses, values of $P < 0.05$ were considered statistically significant. Statistical analyses were conducted using commercially available software.[b]

Results

During the 5-year span investigated, 23,384 complete blood count tests were performed on 9,901 dogs at the Purdue University Veterinary Teaching Hospital. Of those, 1,254 (5.4%) tests revealed platelet counts of $>500 \times 10^3/\mu L$. These 1,254 CBCs were performed on 715 dogs, with a median of 1 platelet count per dog (range 1–23 platelet counts). The incidence of thrombocytosis in this population of dogs was 7.2%. The mean age was 9.16 ± 3.56 years. Sex distribution was 344 (48.1%) spayed females, 272 (38.0%) castrated males, 51 (7.1%) intact males, and 48 (6.7%) intact females. There were 79 breeds represented by the 715 dogs in this study with the most common being the mixed breed dog (154, 21.5%), Yorkshire Terrier (43, 6.0%), Shih Tzu (38, 5.3%), Dachshund (33, 4.6%), Golden Retriever (32, 4.5%), Beagle (27, 3.8%), Chihuahua (26,

3.6%), Labrador Retriever (24, 3.4%), Boston Terrier (23, 3.2%), and Cocker Spaniel (23, 3.2%). No other breed represented greater than 3 percent of the population. Demographic information and selected hematological and biochemical variables for each diagnostic category are displayed (Table 1).

Primary, or idiopathic, thrombocytosis was not diagnosed in any case. Secondary causes identified at the initial visit included a diagnosis of neoplasia in 315 dogs (44.1%), endocrine disease in 87 dogs (12.2%), inflammatory disease in 378 dogs (52.9%), and a miscellaneous diagnosis was made in 9 dogs (1.3%). There were 115 dogs that were assigned multiple diagnoses (16.1%). Median platelet counts were similar between all diagnostic groups (Fig 1). There was no platelet count at a dog's initial visit that could reliably predict the diagnosis.

Of the 1,254 tests documenting thrombocytosis, the median platelet count was 582 $\times$ 10^3/μL (range 500–1,810 $\times$ 10^3/μL). Fifty-five of these tests (4.4%) had platelet clumping noted, so the automated count could have underestimated the total. A total of 560 of these tests had a concurrent anemia (44.7%; RBC RI 5.50–8.50 M/μL) and 10 had a concurrent erythrocytosis (0.8%; RBC RI 5.50–8.50 M/μL). Of the 560 tests with concurrent anemia, reticulocyte counts were available in 63. Regenerative anemia was present in 42 of 63 cases (66.7%; RI > 100 $\times$ 10^3/μL reticulocytes). There was no significant association identified between severity of thrombocytosis and the presence of a regenerative anemia ($P = 0.72$). Concurrent leukopenia was noted in 112/1,254 tests (8.9%; RI 6.0–17.0 $\times$ 10^3/μL), and leukocytosis was a concurrent finding in 263 tests (21.0%; RI 6.0–17.0 $\times$ 10^3/μL).

Table 1. Demographic information and selected hematological and biochemical variables of 715 dogs from the first documentation of thrombocytosis and 1,430 control dogs. Age is reported as mean ± SD. Hematological and biochemical variables are reported as median and range. Distribution of diagnoses for the thrombocytosis and control dogs differed significantly ($P < 0.001$).

	Control Dogs (N = 1,430)	Thrombocytosis Dogs (N = 715)				Serum Potassium (mmol/L)
Diagnosis	Number (%)	Number (%)	Sex	Age (years)	Platelets (×10^3/μL)	N (% of Category)
Neoplasia	472 (33.0%)	315 (44.1%)	166 FS, 10F, 125 MC, 14 M	10.2 ± 2.6	568 (500–1,313)	4.9 (3.7–6.4) 201; 66.7%
Endocrine	54 (3.8%)	87 (12.2%)	48 FS, 4 F, 33 MC, 2 M	10.2 ± 2.8	573 (500–1,313)	5.0 (2.6–6.7) 79; 90.8%
Inflammatory	606 (42.4%)	378 (52.9%)	174 FS, 32 F, 138 MC, 34 M	8.5 ± 4.0	571.5 (500–1,810)	4.9 (2.6–7.1) 325; 86.0%
Miscellaneous	261 (18.3%)	9 (1.3%)	1 FS, 1 F, 6 MC, 1 M	10.1 ± 3.0	562 (537–710)	4.65 (3.8–5.0) 8; 88.9%
Multiple	37 (2.6%)	115 (16.1%)	61 FS, 5 F, 46 MC, 3 M	10.5 ± 2.8	576 (500–1,313)	5.0 (2.6–6.7) 98; 84.5%

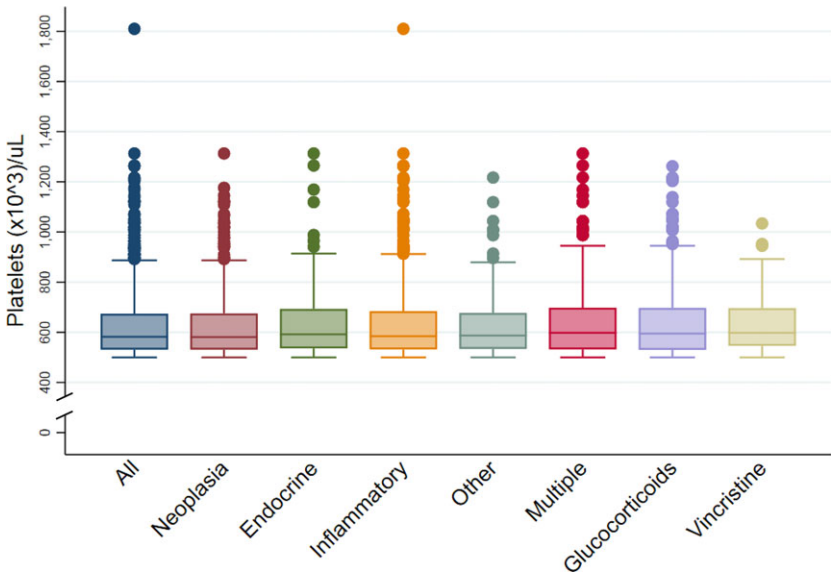

Fig. 1. Box-and-whisker plots showing the median (horizontal line inside the box), 25th and 75th quartiles (top and bottom of box), and minimum and maximum canine platelet counts in each major category.

When stratified by severity of thrombocytosis, 701 (55.9%) tests were classified as mild, 429 (34.2%) as moderate, 91 (7.3%) as marked, and 33 (2.6%) as extreme thrombocytosis. Neoplasia was the most common diagnosis identified for mild, moderate, and extreme thrombocytosis. Inflammatory disease was the most common diagnosis identified for those tests characterized as a marked thrombocytosis. There was no statistical difference between severity of thrombocytosis and diagnostic category ($P = 0.74$; Table 2).

A diagnosis of neoplasia was identified in 315 of 715 dogs at their initial visit (44.1%). When evaluating all tests (n = 1,254), 699 tests were associated with a diagnosis of neoplasia (55.7%). Carcinoma was the most common tumor type identified (365/699, 52.2%; Table 3). There was no significant difference between the median platelet counts of dogs with any specific carcinoma diagnosis ($P = 0.23$); 247 tests were associated with a diagnosis of a round cell tumor (35.3%), and 87 tests were associated with a sarcoma diagnosis (12.5%). The most common neoplastic diagnoses included transitional cell carcinoma (247/699, 35.3%), lymphoma (190/699, 27.2%), mast cell tumor (38/699, 5.4%), and hemangiosarcoma (37/699, 5.3%). There was no statistical difference in the median platelet count between carcinoma, sarcoma, or round cell tumor diagnoses ($P = 0.79$).

During the study period, there were 87 dogs and 151 complete blood counts displaying thrombocytosis that were associated with a diagnosis of endocrine disease; 71 of 151 had a diagnosis of hyperadrenocorticism (47.0%). An adrenal tumor was identified in 6 dogs (8.5%), pituitary-dependent hyperadrenocorticism was confirmed in 29 dogs (40.9%), and the cause for hyperadrenocorticism was unable to be determined in the remaining cases (36/71, 50.7%). There was no statistical difference in median platelet count depending on the type of hyperadrencorticism identified ($P = 0.31$). Other endocrine diagnoses included diabetes mellitus (55/151, 36.4%) and hypothyroidism (48/151, 31.8%).

A total of 378 dogs and 585 complete blood counts displaying thrombocytosis were associated with a diagnosis of inflammatory disease (585/1,254, 46.7%). The most common cause of inflammation identified was immune-mediated disease (130/585, 22.2%). Other common inflammatory processes included renal/urinary disease, gastrointestinal disease, and hepatobiliary disease. There were 205 tests associated with inflammatory disease due to multiple processes (35.0%). Those associated with immune-mediated disease had the highest median platelet count ($636 \times 10^3/\mu L$; range 500–$1,262 \times 10^3/\mu L$), and this was statistically significant ($P < 0.0001$) when compared to the other processes within inflammatory disease. A breakdown of tests

Table 2. Distribution of 1,254 cases of thrombocytosis by severity in each of the major diagnosis categories. There was no significant different noted between severity of thrombocytosis and diagnostic category ($P = 0.75$).

Thrombocytosis ($\times 10^3/\mu L$)	All	Neoplasia	Endocrine	Inflammatory	Miscellaneous	Multiple
Mild	701	393	82	317	13	123
(500–600)	55.9%	56.2%	54.3%	54.2%	54.1%	50.6%
Moderate	429	248	54	196	10	95
(601–800)	34.2%	35.5%	35.8%	33.5%	41.7%	39.1%
Marked	91	40	11	49	1	17
(801–1,000)	7.3%	5.7%	7.3%	8.4%	4.2%	7.0%
Extreme	33	18	4	23	0	8
(>1,000)	2.6%	2.6%	2.6%	3.9%	0%	3.3%

Table 3. Distribution of cases associated with carcinoma. Platelet counts represent the median and range. The distribution of carcinoma diagnoses differed significantly between the thrombocytosis dogs and control dogs ($P < 0.001$). Additional carcinoma diagnoses were made within the control population that are not displayed here.

	Thrombocytosis Dogs (N = 365)		Control Dogs (N = 222)
Carcinoma Type	Platelets ($\times 10^3/\mu L$)	Frequency N (% of Total)	Frequency N (% of Total)
Transitional Cell	580 (500–1,176)	247 (67.7%)	139 (62.6%)
Squamous Cell	587.5 (501–815)	22 (6.0%)	8 (3.6%)
Nasal	564 (506–741)	18 (4.9%)	6 (2.7%)
Hepatobiliary	602.5 (501–782)	16 (4.4%)	8 (3.6%)
Mammary	606 (514–754)	16 (4.4%)	7 (3.2%)
Anal Gland	585.5 (500–726)	12 (3.2%)	16 (7.2%)
Neuroendocrine	519.5 (502–620)	12 (3.2%)	3 (1.4%)
Bronchogenic	602 (580–629)	7 (1.9%)	3 (1.4%)
Prostatic	591 (513–794)	6 (1.6%)	1 (0.4%)
Gastric	547.5 (528–618)	4 (1.1%)	2 (0.9%)
Thyroid	923 (506–1,176)	4 (1.1%)	8 (3.6%)
Pancreatic	593	1 (0.2%)	0 (0.0%)

associated with a diagnosis of inflammatory disease is displayed in Table 4. Sources of inflammation classified as "Other" included dermatologic, dental, cardiovascular, and ophthalmic diseases.

Ten tests from 8 dogs revealed a microcytosis (MCV < 60.0 fL) and hypochromasia (MCHC < 32.0 g/dL), indicating a potential iron deficiency. Of the 8 dogs, 3 were noted to be anemic. Diagnoses in these dogs included 4 with transitional cell carcinoma, 3 with inflammatory disease of the gastrointestinal tract, and 1 with immune-mediated hemolytic anemia. In 7 of these 8 dogs, the cause of these erythroid changes could be attributed to chronic blood loss via hematuria in the dogs with transitional cell carcinoma, or gastrointestinal blood loss in the dogs with inflammatory disease. The final dog was diagnosed with an immune-mediated hemolytic anemia, with an unclear explanation for the morphologic changes in the erythrocytes. The median platelet count among these dogs was $585.5 \times 10^3/\mu L$ (Range 519–862 $\times 10^3/\mu L$) and was not significantly different when compared to the median platelet counts of the other major disease categories ($P = 0.15$).

Glucocorticoids were commonly administered in dogs with thrombocytosis, with 355 of 1,254 complete blood counts performed at a time the dog was receiving a glucocorticoid medication (28.3%). The most common glucocorticoid administered was prednisone/prednisolone (310/355; 87.3%). The median platelet count of the tests run on dogs receiving glucocorticoids was $595 \times 10^3/\mu L$ (range 500–1,262 $\times 10^3/\mu L$), which was not significantly different when compared to the major disease categories ($P = 0.10$). Glucocorticoid use was most common in tests associated with a diagnosis with inflammatory disease (196/355, 55.2%) and neoplasia (170/355, 47.9%). Within inflammatory diseases, glucocorticoid use was most common in those tests associated with a diagnosis of immune-mediated disease (105/196, 53.6%). Of the 33 tests associated with an extreme thrombocytosis, 19 of those were performed in dogs receiving concurrent glucocorticoid administration (57.6%), which is higher than the other categories of thrombocytosis severity.

Chemotherapeutic agents were commonly used in those dogs with a diagnosis of neoplasia and, to a lesser extent, immune-mediated disease. A total of 586 complete blood counts were performed at a time the dog was receiving at least 1 chemotherapeutic agent (46.7%). Vincristine was temporally associated with thrombocytosis on 84 occasions (82/586, 14.0%). The median platelet count of the tests run on dogs receiving vincristine was $598 \times 10^3/\mu L$ (range 500–1,034 $\times 10^3/\mu L$), which was not significant when compared to the major disease categories ($P = 0.32$).

Concurrent serum biochemical data were available for 874 tests. Of those, 314 tests had a serum hyperkalemia concurrent with a thrombocytosis (35.9%). There was a weak, but positive correlation between platelet count and serum potassium concentration (rho = 0.17, $P < 0.0001$) (Fig 2). Plasma potassium concentration was evaluated in 19 cases, with a plasma hyperkalemia noted on one occasion (5.3%). There was no significant correlation noted between platelet count and plasma potassium (rho = −0.22, $P = 0.34$). Serum potassium concentration was available concurrently with plasma potassium in all 19 instances. Serum hyperkalemia was noted in 9 of 19 cases (47.4%), and in 8 of those 9 cases (88.9%), plasma potassium concentration was within normal limits (88.9%). The median difference between serum potassium and plasma potassium concentrations was 0.8 mmol/L (range 0–2.0 mmol/L).

The search of medical records for control dogs with a normal platelet count (300–500 $\times 10^3/\mu L$) yielded 2,246 dogs, and after age-matching, 1,430 dogs were included in the study. The mean age of control dogs was 9.11 ± 3.51 years, which was not significantly different when compared to the thrombocytosis population ($P = 0.87$). The breakdown of diagnoses is displayed in Table 1. The distribution of diagnoses among the broad categories differed significantly between the dogs with thrombocytosis and the control dogs ($P < 0.001$), with inflammatory disease being the most common diagnosis in both groups. Subcategories of inflammatory disease differed significantly between dogs with thrombocytosis and control dogs ($P < 0.001$), with significantly more

Table 4. Distribution of cases diagnosed with inflammatory disease by primary inflammatory process. Platelet counts expressed as median and range. Distribution of diagnoses differed significantly between thrombocytosis dogs and control dogs ($P < 0.001$).

	Thrombocytosis Dogs (n = 585)			
Inflammatory Process	Platelets ($\times 10^3/\mu L$)	Frequency N (% of Total)	Glucocorticoid use N (% of category)	Control Dogs (n = 606) Frequency N (% of Total)
Allergic	537 (500–844)	41 (7.0%)	14 (34.1%)	29 (4.8%)
Gastrointestinal	611 (500–1,217)	83 (14.2%)	27 (32.5%)	127 (21.0%)
Hepatobiliary	582.5 (502–1,265)	72 (12.3%)	9 (12.5%)	54 (8.9%)
Immune-Mediated	636 (500–1,262)	130 (22.2%)	105 (80.8%)	35 (5.8%)
Infectious	547 (501–987)	31 (5.3%)	12 (38.7%)	15 (2.5%)
Neurologic	561 (501–1,119)	56 (9.6%)	22 (39.3%)	115 (19.0%)
Orthopedic	565 (500–933)	58 (9.9%)	10 (17.2%)	94 (15.5%)
Respiratory	541.5 (502–1,810)	52 (8.9%)	15 (28.8%)	58 (9.6%)
Renal/Urinary	569.5 (500–1,169)	112 (19.1%)	25 (22.3%)	79 (13.0%)
Other	594 (501–1,313)	155 (26.5%)	32 (20.6%)	37 (6.1%)

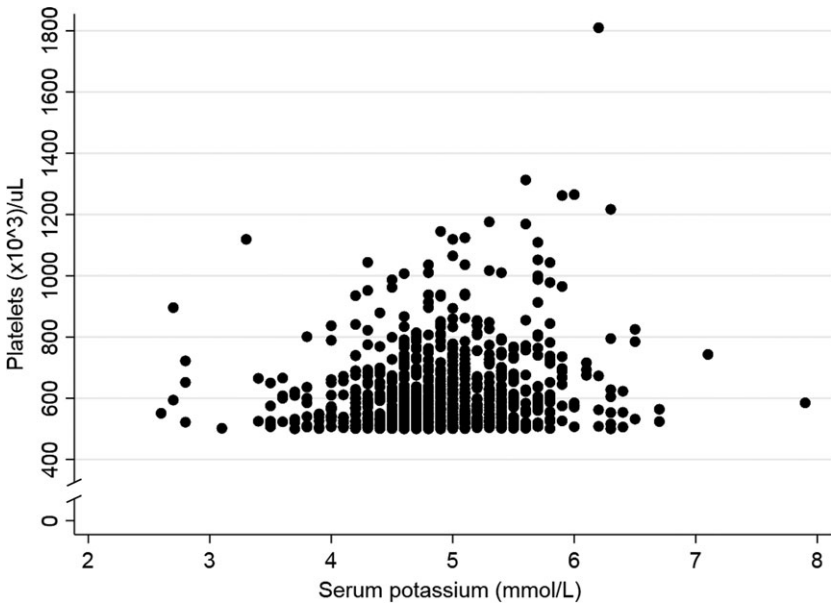

Fig 2. Scatter plot depicting the serum potassium concentration and its correlation with platelet count (rho = 0.17; $P < 0.0001$).

thromobocytosis dogs being diagnosed with immune-mediated disease (Table 4). For control dogs diagnosed with neoplasia (472/1,430, 33.0%), the most common diagnoses were carcinoma (222/472, 47.0%) and round cell tumor (187/472, 39.6%). This distribution was not significantly different from the dogs with thrombocytosis ($P = 0.072$). Within the diagnosis of carcinoma, both groups were most commonly diagnosed with transitional cell carcinoma (TCC). If the dogs with TCC were eliminated from the analysis, there were significantly more thrombocytosis dogs diagnosed with carcinoma (118/225, 52.4%) when compared to the control dogs (83/333, 24.9%) ($P < 0.001$). Other common carcinoma diagnoses differed significantly between groups, as displayed in Table 3 ($P < 0.0001$).

Discussion

Thrombocytosis in dogs had an incidence of 7.2% during the 5-year span of this study. This incidence is similar to a previous report that investigated thrombocytosis in both dogs and cats.[9] Lower incidences are reported previously in a canine (4.5%) and a feline (4.6%) restrospective study.[8,28] This difference can likely be attributed to the platelet count cutoff used to define a thrombocytosis. Both of the studies reporting a higher incidence, present included, used a platelet count cutoff of $>500 \times 10^3/\mu L$ to identify dogs with thrombocytosis. A previous study of thrombocytosis in dogs utilized a platelet cutoff of $>600 \times 10^3/\mu L$ for inclusion.[8] In a study of cats with thrombocytosis, those with a platelet count of $>700 \times 10^3/\mu L$ were included.[28] There is no current consensus in human or veterinary clinical pathology as to the most appropriate cutoff for classifying a thrombocytosis. Large retrospective studies and reviews of thrombocytosis in the human medical field have utilized a cutoff of $>450–500 \times 10^3/\mu L$ to define a thrombocytosis, but a cutoff of $>800 \times 10^3/\mu L$ to signify clinically relevant thrombocytosis.[29–31] For the present study, the cutoff value of $>500 \times 10^3/\mu L$ was extrapolated from past studies to establish disease correlation with higher platelet counts, as the clinical relevance of severity is unknown and investigating this was one of the objectives of the study.

More recent literature suggests that the use of a platelet count is not be the best way to assess overall platelet mass. Limitations of a platelet count include the fact that there is an inverse relationship between platelet number and platelet size. Therefore, lower numbers of larger platelets and higher numbers of small, mature platelets represent the same platelet mass. This has been investigated in Cavalier King Charles Spaniels due to their hereditary macrothrombocytopenia, and in these dogs, a calculated plateletcrit could be superior to a platelet count for determining platelet mass.[32] The mean platelet volume (MPV) can be a useful indication of the body's regenerative response to a reduced platelet mass; however, this has not become a routine measurement in veterinary hematologic analysis.[33,34] The current study used only platelet count and review of a blood smear, so it could have benefitted from better assessment of platelet mass.

Secondary, or reactive, thrombocytosis is more common than primary thrombocytosis in all species. No cases of primary thrombocytosis were identified in this population of dogs. Neoplasia was a common cause of reactive, or secondary, thrombocytosis in this population of dogs. Forty-four percent of the dogs in this study received a diagnosis of neoplasia, which is the highest percentage reported in any other veterinary study investigating thrombocytosis. Similar to other studies, carcinoma was the most common neoplastic diagnosis.[8,9] Carcinoma-associated thrombocytosis is well documented in humans. In these cases, the tumor

has been shown to produce granulocyte-macrophage colony-stimulating factor (GM-CSF), as well as IL-6, and TPO.[35–38] Paraneoplastic thrombocytosis is a poor pretreatment prognostic factor in a number of cancers in humans, including breast, gastric, colorectal, brain, and lung tumors.[39–41] The most common tumor diagnosed in the dogs of the present study was transitional cell carcinoma (TCC). This is different than other studies, likely because this institution is a tertiary referral hospital and has a much higher TCC caseload compared to other, similar institutions, due to targeted research by the medical oncology service. This is noted in the control population, as TCC was the most common carcinoma diagnosed in this group as well. There was no significant difference in the distribution of neoplastic diagnoses between the 2 populations. However, once the dogs with TCC were removed from analysis, a significant difference in neoplastic diagnosis exists. This demonstrates an institutional bias for the diagnosis of TCC, but also supports the association between thrombocytosis and a diagnosis of carcinoma. Round cell tumors were the second most common neoplastic diagnosis in the thrombocytosis, with lymphoma the most common round cell tumor identified. Lymphoma, in dogs and cats, is a diagnosis made frequently without investigating the bone marrow. Therefore, it is possible some dogs diagnosed with lymphoma and thrombocytosis represent leukemic dogs with megakaryoblastic proliferation. Additionally, as lymphoma is one of the most common cancers in dogs, this will bias any study investigating neoplastic diagnoses in a large population of dogs.[42] The present study was also skewed by the fact that many dogs admitted to the study had more than one complete blood count documenting thrombocytosis. Dogs undergoing chemotherapeutic treatment of lymphoma have routine CBCs performed before administration of a cytotoxic medication, and are also routinely treated with glucocorticoids and vincristine, both of which can increase platelet count.

Whereas there were more complete blood counts associated with a diagnosis of neoplasia in the present study, more individual dogs were diagnosed with inflammatory disease at their initial assessment. Inflammatory disease is a common cause for thrombocytosis through multiple mechanisms including cytokine interactions, especially IL-6, which stimulates TPO production.[10] Additionally, inflammatory diseases associated with chronic blood loss could lead to an iron-deficient state, stimulating thrombopoiesis through an unknown, but seemingly TPO-independent process.[5,6] Whereas this iron deficiency could stimulate thrombopoiesis, there was no correlation found with the severity of thrombocytosis when compared to other diagnoses.

The most common inflammatory diseases noted in dogs with thrombocytosis were the immune-mediated disease, gastrointestinal disease, hepatobiliary disease, and renal disease. This distribution differed significantly from the control dogs, whose inflammatory diseases were most commonly attributed to gastrointestinal, neurologic, and orthopedic diseases ($P < 0.001$). Hepatobiliary disease was a more common finding in this population of dogs with thrombocytosis, with 12.3% of inflammatory diseases classified in this category, when compared to a previous study that only attributed 2.4% of inflammatory diseases to the hepatobiliary tract.[8] The liver is a source of thrombopoietin, and thrombocytosis has been reported as a paraneoplastic syndrome and prognostic factor in hepatocellular carcinoma in people. The role of TPO in liver disease of veterinary species has not been investigated, so there could be additional mechanisms contributing to thrombocytosis.

The present study also had a higher percentage of dogs with renal disease, at 19.1%, when compared to previous studies of dogs (6.6% and 3.6%) and cats (7.8%).[8,9,28] Uremia causes abnormalities in platelet function and alters the interaction between platelets and the vessel wall. Uremia has an aspirin-like effect on platelets, causing abnormal granule content and release, abnormal arachidonic metabolism, abnormal cyclooxygenase activity, and abnormal binding of the GPIIb-IIIa receptor.[43] Additionally, changes in parathyroid production associated with chronic kidney disease can affect intracellular calcium, and thus, platelet function.[43] In people with chronic kidney disease, thrombocytosis has been shown to be prognostic, due to the suspicion it is caused by iron deficiency and malnutrition-inflammation-cachexia syndrome (MICS) and thus, indicative of more severe disease.[43,44] Not all dogs in this category in the present study were uremic, as it represented a combination of renal and lower urinary tract diseases. There are no current veterinary studies investigating thrombocytosis as a prognostic marker in renal disease.

Immune-mediated disease is a common inflammatory disease associated with thrombocytosis in people and veterinary species. Immune-mediated disease represented the largest percentage of inflammatory diseases in the present study, with a much higher frequency of diagnosis in thrombocytosis dogs when compared to the control population ($P < 0.001$). Dogs with immune-mediated disease were also the most likely to be treated with corticosteroids. Thrombocytosis is a common finding in dogs with hyperadrenocorticism, although the mechanism is unknown. Iatrogenic Cushing's (hypercortisolemia) was a common finding in the present study due to the number of dogs receiving corticosteroids. Concurrent corticosteroid administration is commonly reported for dogs with thrombocytosis, but causes minimal to slight increases in platelets in response to chronic anti-inflammatory doses of corticosteroids in healthy dogs.[15,45] Immune-mediated diseases are treated with higher doses of steroids, so it is possible these higher doses contribute to a more severe hypercortisolemia, and thus a higher likelihood of developing thrombocytosis.

Interestingly, control dogs were more commonly diagnosed with inflammatory processes that were neurologic or orthopedic in origin, when compared to dogs with thrombocytosis. This could indicate that dogs with inflammatory disease of these types are unlikely to develop thrombocytosis. This could be due to the fact that neurologic or orthopedic inflammatory processes are more localized, and do not tend to be associated

with systemic inflammation; therefore, the influence of inflammatory cytokines on thrombopoiesis is minimal. Dogs with a leukocytosis and neutrophilia were previously classified by diagnosis, and none of the dogs in this population received a diagnosis of neurologic or orthopedic disease, with the exception of one primary rub tumor.[46] This increased number of neurologic and orthopedic diseases in the control group of dogs was also likely affected by the retrospective nature of this study. Dogs with neurologic and orthopedic diseases could be more likely to have complete blood counts performed at a time when they are expected to be normal, as part of a pre-anesthetic or presurgical labwork assessment.

There was no significant difference found between the severity of the thrombocytosis and the diagnostic category. There was no platelet count cutoff above which a specific diagnosis could be predicted. This differs from a previous study in which it was found that extreme thrombocytosis was more commonly associated with neoplastic disease.[8] In the present study, inflammatory disease was the most commonly associated with extreme thrombocytosis, but the difference was slight and insignificant. It was found that a higher percentage of dogs with extreme thrombocytosis were receiving glucocorticoids. It is possible that dogs receiving glucocorticoids to treat another disease with the potential to cause thrombocytosis have a higher likelihood of developing a more severe increase in platelet count due to multiple factors affecting thrombopoiesis. Similar to prevalence, these percentages could have been affected by the platelet count cutoff utilized to define thrombocytosis.

Serum hyperkalemia is a common finding in dogs with thrombocytosis. It is likely that this is due to pseudohyperkalemia caused by the release of intracellular potassium from the increased thrombocyte mass after phlebotomy.[25,26] Assessment of concurrent plasma potassium has been described to document the presence of a pseudohyperkalemia.[25] Concurrent serum and plasma potassium concentrations were available for few cases in the current study, so no conclusions about the prevalence of pseudohyperkalemia in this population can be made. There are few veterinary studies investigating the phenomenon of pseudohyperkalemia, so its mechanism would benefit from further investigation, as the majority is extrapolated from research in people.[25]

This study represents one of few investigating thrombocytosis in large numbers of animals. It is the largest retrospective of canine thrombocytosis to date. Limitations of the study include the challenges in assessing platelet mass, as previously discussed. Additionally, this study categorized diagnoses into large categories and primary inflammatory processes, but did not, in most cases, identify the specific etiology. Because of this, it is possible disease correlations are present but not identified due to the way through which cases were classified. The control group used for comparison contributed additional limitations. For one, whereas this group was age-matched to the study population, breed matching was not possible, and this could have helped to better interpret disease associations. Review of the medical records for control dogs did not include a review of concurrent medications, so the number of control dogs on concurrent medications affecting platelet count was not investigated. In people, thrombocytosis contributes to a risk of thromboembolic events, especially in cases of primary thromobocytosis or in uremic patients. The present study did not investigate the potential for thromboembolic events in dogs during medical record review. Additionally, thrombocytosis affects prognosis and survival in many diseases in people, especially neoplasia. The present study did not investigate survival or outcomes, so this should be investigated in future studies.

In conclusion, thrombocytosis in dogs is mainly a reactive or secondary process and is most commonly associated with inflammatory diseases and neoplasia, when compared to the general hospital population. Thrombocystosis in neoplasia is most commonly associated with a diagnosis of carcinoma. Immune-mediated diseases, gastrointestinal diseases, and hepatobiliary diseases are the most common inflammatory processes associated with thrombocytosis. Concurrent glucocorticoid administration was common in this population and was more likely to be associated with an extreme thrombocytosis. Serum hyperkalemia was a common concurrent abnormality and could be falsely increased due to the increased platelet count. Further studies are warranted to investigate the role of thrombocytosis as a risk for thromboembolic events, and its effect on survival and prognosis in certain disease states.

Footnotes

[a] Abbott Laboratories; Abbott Park, IL
[b] STATA SE, v.14.1, StataCorp, College Station, TX

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Off-label Antimicrobial Use Declaration: Authors declare no use of off-label antimicrobial.

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