Investigation of hypomagnesaemia prevalence and underlying aetiology in a hospitalised cohort of dogs with ionised hypocalcaemia

Citation for published version:
Woods, GA, Oikonomidis, IL, Gow, AG, Tørnqvist-Johnsen, C, Boyé, P, Chng, Y & Mellanby, RJ 2021, 'Investigation of hypomagnesaemia prevalence and underlying aetiology in a hospitalised cohort of dogs with ionised hypocalcaemia', *The Veterinary record*. https://doi.org/10.1002/vetr.301

Digital Object Identifier (DOI):
10.1002/vetr.301

Link:
Link to publication record in Edinburgh Research Explorer

Document Version:
Peer reviewed version

Published In:
The Veterinary record

Investigation of hypomagnesaemia prevalence and underlying aetiology in a hospitalised cohort of dogs with ionised hypocalcaemia

Glynn A.Woods[1*], Ioannis L. Oikonomidis[1], Adam G Gow[1], Camilla Tørnqvist-Johnsen[1], Pierre Boyé[1], Yuru Chng[1], Richard J Mellanby[1]

[1] The Royal (Dick) School of Veterinary Studies and The Roslin Institute, Division of Veterinary Clinical Studies, The University of Edinburgh, Hospital for Small Animals, Easter Bush Veterinary Centre, Roslin, Midlothian, United Kingdom

[*]Corresponding author: Dr. Glynn A Woods

E-mail: gwoods2@ed.ac.uk

Contributor statement: GW was the main author involved in conceptualising, planning, conduct of research, reporting of the work described in the article and the named guarantor. RM contributed toward conceptualising, planning and editing of drafts. IO contributed toward statistical analysis and editing of drafts. AG, CJ and PB contributed to planning and editing of drafts. YC contributed by planning and conducting research.

Abstract

Objectives

Calcium is the most abundant mineral in the body and plays a critical role in a wide range of physiological processes. Low concentrations of ionised calcium, the most metabolically available form of calcium, have been linked to an increased risk of adverse clinical outcomes in dogs. Magnesium plays an important role in parathyroid hormone function. The objective of this study was to define the prevalence and aetiology of hypomagnesaemia in a hospitalised cohort of dogs with ionised hypocalcaemia.

Methods

A total magnesium reference interval was established using serum biochemistry results from 346 clinically healthy dogs. The clinical records of dogs with ionised hypocalcaemia were reviewed and concurrent serum magnesium concentrations were recorded alongside clinical signs and underlying aetiology. The prevalence, clinical presentation and aetiology of hypomagnesaemia were examined in the ionised hypocalcaemic population.

Results

295 ionised hypocalcaemia dogs were identified. Hypomagnesaemia was identified in 22%. Total magnesium concentration was significantly higher in dogs with renal disease. The most common cause of concurrent hypomagnesaemia and ionised hypocalcaemia was gastrointestinal diseases.

Conclusion

Low concentrations of serum magnesium occur in approximately one fifth of all dogs with ionised hypocalcaemia. Further work is required to clarify the link between magnesium status, ionised hypocalcaemia and clinical outcome.

Introduction

Calcium is an important mineral, required for cellular functions such as enzyme activity, membrane stability and transport, as well as playing a critical role in skeletal development and health (1). Calcium circulates in three forms; ionised calcium (iCa), protein bound and bound to non-protein anions (e.g. citrates, phosphates, lactate, and other small, diffusible anions). Ionised calcium is the most metabolically active component (1). Under normal physiological conditions there are three main hormones that control calcium homeostasis; parathyroid hormone (PTH), calcitonin and 1,25 dihydroxycholecalciferol. Parathyroid hormone is released in response to decreased ionised calcium and has a wide range of physiological effects on several different organs, with the overall impact of increasing serum ionised calcium. Parathyroid hormone also acts to upregulate 1-alpha-hydroxylase in renal peritubular cells, which converts 25-hydroxycholecalciferol (25(OH)D) to 1,25 dihydroxycholecalciferol, leading to increased intestinal absorption of calcium (1,2).

Hypocalcaemic disorders are clinically important in veterinary patients. Ionised hypocalcaemia (IHC) decreases the threshold potential of neuronal, cardiac and muscle cells and as a result, the majority of clinical signs associated with hypocalcaemia are a result of increased cell excitability (3). Dogs with IHC can present with a variety of signs including weakness, vomiting, diarrhoea, abdominal pain, arrhythmias and a spectrum of neurological signs including seizure activity (4–7). Importantly, the severity of IHC has been associated with all cause morbidity and mortality in dogs and associated with poor outcomes in canine critical illness (8,9).

Similar to calcium, magnesium is present in the serum in ionised, protein bound and complexed forms. Magnesium is an essential element, acting as a cofactor in a large number of vital intracellular physiological reactions(10). Total serum magnesium remains the most commonly measured form of magnesium as free magnesium requires the use of specialised ion selective electrode techniques.

Disturbances in magnesium homeostasis are increasingly recognised in veterinary patients, especially in critically ill animals (11). Magnesium is required for the function of the sodium potassium ATPase pump and is therefore vital for control of electrolyte gradients across cell membranes (12). Similar to IHC, hypomagnesaemia has been linked to exacerbating morbidity in both human and veterinary patients (13–17). For example in human patients, hypomagnesaemia has been linked to morbidity in patients with diabetic ketoacidosis and hypoparathyroidism (18,19). However, few studies have examined the relationship between magnesium and calcium in veterinary patients.

Both hypomagnesaemia and hypocalcaemia can share a similar pathogenesis in veterinary patients involving intestinal loss, malabsorption, altered distribution and abnormalities of vitamin D metabolism (20,21). Magnesium also plays a key role in regulating calcium homeostasis by modulating the production and release of PTH (22). Severely decreased magnesium concentration results in an inhibition of PTH release (23–25) by inhibiting magnesium-dependent enzymes required for PTH exocytosis (24). This functional hypoparathyroidism decreases calcium and magnesium absorption in the distal convoluted tubule of kidney. Consequently, severe hypomagnesaemia can lead to clinically relevant hypocalcaemia in people (26). In human medicine, evaluation of magnesium in patients with IHC is considered standard clinical practice (23) since resolution of hypocalcaemia can be challenging without magnesium repletion (23). Despite the improved understanding of the importance of magnesium in regulating plasma concentrations of ionised calcium in humans, the role magnesium plays in regulating canine calcium homeostasis and the prevalence of hypomagnesaemia in dogs with IHC is poorly understood. A better understanding of the relationship of magnesium status in dogs with IHC may help guide diagnostic and monitoring strategies. Consequently, the aim of this study was to define prevalence of total serum hypomagnesaemia in a population of hospitalised dogs with IHC and to establish which diseases have the highest prevalence of concurrent hypomagnesaemia and IHC.

2. Method

2.1 Design and setting

The Royal (Dick) School of Veterinary Studies clinical database was searched for dogs with IHC at admission from January 2012 and November 2018. IHC was defined as iCa ≤ 1.18 mmol/L (reference interval [RI] 1.18-1.53mmol/L) (27). Dogs younger than 12 months of age were excluded. The medical records of dogs with IHC were reviewed and the following variables were recorded: age, breed, sex, glucocorticoid administration, diuretic therapy, final clinical diagnosis and serum iCa, total calcium (tCa), albumin and magnesium concentrations. Analytes of each dog were measured at the same time point and as part of routine biochemical analysis. If dogs presented with IHC more than once, the first instance of IHC was recorded. Dogs treated with glucocorticoids or diuretics 48 hours prior to referral were excluded. Only dogs with IHC and available magnesium values were included in the study. The study was approved by R(D)SVS VERC.

Ionised calcium was measured within one hour of sample collection and the analysis of the remaining biochemistry was performed within three hours of collection. Measurement of biochemical analytes was performed on serum and measurement of iCa on lithium heparin whole blood. The iCa sample was drawn into a syringe after sampling and the syringe capped by adding a stopper in order to facilitate anaerobic storage prior to analysis. The AU480 biochemistry analyser was used for all biochemical analyses other than for iCa (Beckman Coulter, High Wycombe, Buckinghamshire, UK). The Gem 3500 analyser (Instrumentation Laboratory, Warrington, Cheshire, UK), which employs an ion-selective electrode assay, was used for measurement of the iCa.

2.2 Establishing total magnesium reference interval

Total serum magnesium (tMg) concentrations were obtained using the same healthy cohort as previously reported (27). Dogs were included if no clinical signs or significant history were reported by the owner and no abnormalities were detected by an attending veterinary surgeon. All dogs were fed commercially available diets and no dog was receiving supplementation or medication. Total serum magnesium was measured on the AU480 biochemistry analyser (Beckman Coulter, High Wycombe, Buckinghamshire, UK).

Reference intervals for tMg were established using a previously described Reference Value Advisor, a non-parametric Excel software tool (28). In short, the 95% coverage reference interval was defined by the 2.5 and 97.5 percentiles from the observed data with percentiles then obtained by linear interpolation when the data did not fall within exact percentiles.

2.3 Ionised hypocalcaemia diagnostic groups

The cases were grouped, according to a previously published article on IHC (29), into the following aetiologies (1,30);gastrointestinal disease (GI), renal disease (REN), pancreatitis (PAN), immune mediated disease (IMM), endocrine disease (END), neoplasia (NEO), hepatic disease (HEP) and miscellaneous conditions (MIS).

In dogs with comorbidities, the diagnosis leading to the most significant clinical signs and which would most likely account for the IHC was selected as the categorising diagnosis. Cases in which the final diagnosis was not covered in the seven major groupings were classified as miscellaneous.

2.4 Stratification of IHC severity

Dogs were grouped in three categories according to the iCa concentration and stratified, as mild (1.00 – 1.17 mmol/L), moderate (0.8 – 0.99 mmol/L) and severe (< 0.8 mmol/L) (31).

2.5 Statistical analysis

The data distribution was evaluated using the Shapiro-Wilk test. The chi-square test was used to examine the association between the levels of two categorical variables. The Wilcoxon rank sum test with continuity correction was used to compare continuous data. The Kruskal-Wallis test was used to compare three or more independent groups. When statistically significant differences were observed using the Kruskal-Wallis test, post-hoc tests were conducted employing Dunn's test in order to control the family-wise error rate and define which groups presented statistically significant differences. The correlation between two variables was examined with Spearman's correlation coefficient. All the statistical analyses were performed using the statistical language R (R Foundation for Statistical Computing, Vienna, Austria). For all tests applied, a P value <0.05 was considered significant.

Results

Ionised calcium and total calcium

The distribution of all variables was non-Gaussian. A total of 295 dogs met the inclusion criteria. The median iCa concentration was 1.13 mmol/L (range: 0.71 - 1.17 mmol/L). There was nineteen entire male dogs (6%), one hundred and fifty neutered males (51%), six entire females (2%) and one hundred and twenty (41%) females were neutered. The median age in the IHC cohort was 7 years (range: 1 to 14 years). The most common breeds with IHC were Labradors (n=29, 10%), cocker spaniels (n=20, 7%) and cross breeds (n=14, 5%).

Two-hundred and sixty five (90%) dogs had mild IHC (1.00 - 1.17 mmol/L), twenty-five (8%) dogs had moderate IHC (0.8 – 0.99 mmol/L) and five (2%) with severe IHC (< 0.8 mmol/L). No statistically significant difference ($P = 0.596$) in the distribution of sex was noted between the dogs with mild IHC (149 males and 116 females) and dogs with moderate and severe IHC (20 males, 5 females). Dogs with severe IHC were combined with moderate IHC to facilitate statistical analysis. There was no statistical difference in breed ($P = 0.466$), sex ($P = 0.598$) or age ($P = 0.141$) between all groups either when compared individually or when the mild IHC group was compared with the moderate and severe IHC group combined.

All 295 IHC dogs had a serum tCa concentration recorded. Two hundred (68%) dogs had a decreased tCa (RI: 2.24 – 2.85 mmol/l). The median tCa was 2.12 mmol/L (0.81-2.80 mmol/L). One hundred and seven (36%) of dogs with IHC had a tCa within the RI. There was a statistically significant, weak positive correlation (rho = 0.287, $P<0.001$) between tCa and iCa.

Total serum magnesium reference interval

Three hundred and forty-six clinically healthy adult dogs were used to establish the total tMg RI. The median age of these dogs was 5.9 years (range: 1 to 14 years). There were 161 spayed females (47%), 24 entire females (7%), 113 neutered males (33%) and 48 entire males (14%). Dogs in the healthy cohort were younger (P = 0.018) and more likely to be entire (P = 0.008) than the IHC dogs. The most common breeds were cross breed dogs (n = 91, 26%), Labrador retrievers (n = 49, 14%), cocker spaniels (n = 29, 8%), springer spaniels (n = 15, 4%) and golden retrievers (n = 14, 4%). There was significant correlation between tMg and tCa (rho=0.275, P <0.001) and between tMg and serum albumin (rho=0.339, P <0.001) in the healthy dog. There was no correlation between tMg and iCa (rho = -0.094, P = 0.105).

TMg, IHC, tCa and albumin

Of the 295 dogs with IHC, sixty-four (22%) ionised hypocalcaemic dogs had a tMg concentration below the reference interval, 221 (75%) had a tMg concentration within the reference range and ten dogs (3%) had IHC and hypermagnaesemia. The median age of dogs with IHC and hypomagnesaemia was 7 years (range: 1 to 14 years). There were two entire females (3%), 25 neutered females (39%), 7 entire males (10%) and 30 neutered males (46%). The most common breeds with hypomagnesaemia and IHC were Labradors (n = 9), cocker spaniel (n = 6) and labradoodle (n = 4).

Fifty-five out of 64 (86%) dogs had mild IHC, 7 (11%) dogs had moderate IHC and 2 (3%) dogs had severe IHC. No significant correlation was found between tMg concentration and iCa concentration (rho = -0.094, P = 0.105). Serum tMg concentration was weakly positively correlated with tCa concentration (rho = 0.275, P <0.001) and with serum albumin concentration (rho = 0.339, P = <0.001).There were no statistical differences in serum magnesium concentration between patients with mild, moderate and severe IHC (P = 0.279) (Figure 1).

Diagnostic categories

Eighty-two (22%) IHC dogs were categorized with GI, 53 (18%) with NEO, 42 (14%) with MIS, 35 (12%) with REN, 33 (11%) with IM, 20 (7%) with HEP, 19 (6%) with END and 11 (4%) with PAN (table 1). The Kruskal-Wallis test revealed a statistically significant difference in median tMg concentration between the different disease groups (P = 0.002). Specifically, the multiple pairwise comparisons using Dunn's test showed that serum magnesium concentration was significantly higher in dogs with renal disease (median: 0.99, range: 0.30-1.70 mmol/L) compared to those with endocrine diseases (median: 0.79, range: 0.45-1.12 mmol/L, P = 0.010) and gastrointestinal disease (median: 0.78, range: 0.25-1.66 mmol/L, P = 0.002] (Figure 2). The tMg concentration of all IHC dogs within each diagnostic category are summarised in Table 1. There was no significant difference between the prevalence of the IHC disease categories between the different Mg concentrations (P = 0.180). The number of dogs within each IHC diagnostic category remained statistically non-significant even when hypomagneaemia was compared to a combined normomagnesaemia and hypermagnesaemia group (P = 0.671). The number of IHC dogs categorized as gastrointestinal was numerically greater representing 38 % and 23 % of all IHC dogs with hypomagnesaemia. Comparison of disease frequencies between dogs with hypomagnesaemia and dogs with normo-, and hypermagnesaemia were not significant (P = 0.311).

Magnesium concentration	IHC Diagnostic category								
	Total	IM	END	GI	HEP	NEO	PAN	REN	MIS

Hypomagnesaemia	64	6	5	24	6	11	2	6	4
Normomagnesaemia	218	26	14	56	14	40	7	25	36
Hypermagnesaemia	13	1	0	2	0	2	2	4	2

Table 1: Number of IHC dogs with hypo-, normo- and hypermagnesaemia within each IHC diagnostic category. IMM, immune mediated, END, endocrine disease; GI, gastrointestinal/dietary disease; HEP, hepatic disease; PAN, pancreatitis; PHP, primary hypoparathyroidism; REN, renal disease; MIS, miscellaneous causes.

Discussion

This study reports the prevalence of tMg abnormalities in a large population of dogs with IHC. We found that 22 % of dogs with IHC were hypomagnesaemic. This study documented that there was a significant difference in serum tMg concentration between IHC disease categories. Results of this study found no significant difference between the prevalence of hypo-, normo- and hypermagnesemia between different IHC disease categories. Furthermore, tMg concentrations did not differ significantly between IHC severity groups.

Although tMg's role in contributing to IHC has been postulated in veterinary patients, this is the first study to assess the relationship between tMg and iCa in a large number of IHC dogs. Our study did not find a relationship between tMg and severity of IHC despite both moderate and severe IHC being combined for statistical appraisal.

Results of our study agree with a previous canine experimental study (32), providing corroborating clinical evidence that roughly one fifth of IHC dogs are hypomagnesaemic. The results are consistent with findings in humans with IHC where 2 - 65 % of patients are likely to be hypomagnesaemic (33).

In our study, the most common disease grouping of dogs with hypomagnesaemic and IHC was gastrointestinal diseases. Ionised hypocalcaemia is a well-recognised complication of gastrointestinal disease, particularly in dogs with a protein losing enteropathies (PLE)(34–37). Hypomagnesaemia has also been documented in conjunction with IHC in canine PLE. Future studies investigating the relationship of calcium, albumin and magnesium in different gastrointestinal conditions are warranted.

Results showed that over a third of dogs with IHC had a total calcium within the reference interval. Discrepancies between total calcium and ionised calcium have been reported in both human and veterinary literature (27,38–40). A previous study documented that approximately one third of dogs with ionised hypercalcaemia had a total calcium within the reference interval (27). Results of this paper highlight the value of measuring both total and ionised calcium when assessing calcium homeostasis in ill dogs.

The measurement of total magnesium was a major limitation of this study. Extracellular magnesium represents only 1% of total body magnesium. Ionised magnesium represents 55 – 70% of extracellular magnesium and is considered to be the most biologically active constituent (10). Measurement of total serum magnesium does not adequately correlate with whole body magnesium levels because as in human patients only 0.3% of total body magnesium is contained in serum(41). The correlation between iMg and tMg is highly variable in human studies, affected not only by the underlying disease process but also the degree of hypoalbuminaemia (42–44). Similarly this measurement does not predict the intracellular magnesium level, which is responsible for vital cellular reactions (45).

In summary, this study demonstrated that 22% of IHC dogs were hypomagnesaemic. The most common disease causing concurrent IHC and hypomagnesaemia was gastrointestinal disease. Further studies are required to explore the impact of treating hypomagnesaemia on calcium homeostasis in dogs with IHC.

1. DiBartola SP. Fluid, Electrolyte, and Acid-Base Disorders in Small Animal Practice. Fluid, Electrolyte, and Acid-Base Disorders in Small Animal Practice. Elsevier Inc.; 2012.
2. Harjes LM, Parker VJ, Dembek K, Young GS, Giovaninni LH, Kogika MM, et al. Fibroblast Growth Factor-23 Concentration in Dogs with Chronic Kidney Disease. J Vet Intern Med. 2017;31(3):784–90.
3. Barrett KE, Ganong WF. Ganong's review of medical physiology. McGraw-Hill Medical; 2010. 714 p.
4. Dhupa N, Proulx J. Hypocalcemia and hypomagnesemia. Vet Clin North Am Small Anim Pract [Internet]. 1998 May [cited 2019 Aug 14];28(3):587–608. Available from: http://www.ncbi.nlm.nih.gov/pubmed/9597716
5. Drobatz KJ, Casey KK. Eclampsia in dogs: 31 cases (1995-1998). J Am Vet Med Assoc [Internet]. 2000 Jul 15 [cited 2020 Mar 16];217(2):216–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/10909462
6. Russell N, Bond K, Robertson I, Parry B, Irwin R. Primary Hypoparathyroidism in Dogs: A Retrospective Study of 17 Cases. Aust Vet J [Internet]. 2006 [cited 2020 May 27];84(8). Available from: https://pubmed.ncbi.nlm.nih.gov/16911229/
7. Brauer C, Jambroszyk M, Tipold A. Metabolic and toxic causes of canine seizure disorders: A retrospective study of 96 cases. Vet J [Internet]. 2011 Feb [cited 2020 Mar 16];187(2):272–5. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19939714
8. Goggs R, De Rosa S, Fletcher DJ. Electrolyte Disturbances Are Associated with Non-Survival in Dogs—A Multivariable Analysis. Front Vet Sci [Internet]. 2017 Aug 18 [cited 2019 Aug 14];4:135. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28868302
9. Luschini MA, Fletcher DJ, Schoeffler GL. Incidence of ionized hypocalcemia in septic dogs and its association with morbidity and mortality: 58 cases (2006-2007). J Vet Emerg Crit Care [Internet]. 2010 Aug [cited 2020 Mar 16];20(4):406–12. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20731806
10. Jahnen-Dechent W, Ketteler M. Magnesium basics. Clin Kidney J [Internet]. 2012 Feb [cited 2020 Oct 31];5(Suppl 1):i3–14. Available from: http://www.ncbi.nlm.nih.gov/pubmed/26069819
11. Schulz N, Güssow A, Bauer N, Moritz A. Magnesium in dogs and cats - Physiology, analysis, and magnesium disorders. Tierarztl Prax Ausgabe K Kleintiere - Heimtiere. 2018;46(1):21–32.
12. Martin LG, Wingfield WE, Van Pelt DR, Hackett TB. Magnesium in the 1990's: Implications for Veterinary Critical Care. J Vet Emerg Crit Care [Internet]. 1993 Jul 1 [cited 2020 Mar 16];3(2):106–14. Available from:

http://doi.wiley.com/10.1111/j.1476-4431.1993.tb00108.x
13. Limaye CS, Londhey VA, Nadkart MY, Borges NE. Hypomagnesemia in critically ill medical patients. J Assoc Physicians India [Internet]. 2011 Jan [cited 2020 Jun 16];59:19–22. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21751660
14. Fiaccadori E, Del Canale S, Coffrini E, Melej R, Vitali P, Guariglia A, et al. Muscle and serum magnesium in pulmonary intensive care unit patients. Crit Care Med [Internet]. 1988 Aug [cited 2020 Jun 16];16(8):751–60. Available from: http://www.ncbi.nlm.nih.gov/pubmed/3396369
15. Velissaris D, Karamouzos V, Pierrakos C, Aretha D, Karanikolas M. Hypomagnesemia in Critically Ill Sepsis Patients. J Clin Med Res [Internet]. 2015 Dec [cited 2020 Jun 16];7(12):911–8. Available from: http://www.jocmr.org/index.php/JOCMR/article/view/2351
16. Safavi M, Honarmand A. Admission hypomagnesemia--impact on mortality or morbidity in critically ill patients. Middle East J Anaesthesiol [Internet]. 2007 Oct [cited 2020 Jun 16];19(3):645–60. Available from: http://www.ncbi.nlm.nih.gov/pubmed/18044292
17. Humphrey S, Kirby R, Rudloff E. Magnesium physiology and clinical therapy in veterinary critical care [Internet]. Vol. 25, Journal of Veterinary Emergency and Critical Care. Blackwell Publishing Ltd; 2015 [cited 2020 Mar 16]. p. 210–25. Available from: http://www.ncbi.nlm.nih.gov/pubmed/25427407
18. Davis SM, Maddux AB, Alonso GT, Okada CR, Mourani PM, Maahs DM. Profound hypokalemia associated with severe diabetic ketoacidosis. Pediatr Diabetes. 2016 Feb 1;17(1):61–5.
19. Mutnuri S, Fernandez I, Kochar T. Suppression of Parathyroid Hormone in a Patient with Severe Magnesium Depletion. Case Reports Nephrol. 2016;2016.
20. Chernow B, Bamberger S, Stoiko M, Vadnais M, Mills S, Hoellerich V, et al. Hypomagnesemia in Patients in Postoperative Intensive Care. Chest [Internet]. 1989 Feb 1 [cited 2020 May 27];95(2):391–7. Available from: https://linkinghub.elsevier.com/retrieve/pii/S0012369216332986
21. Wester PO. Electrolyte balance in heart failure and the role for magnesium ions. Am J Cardiol [Internet]. 1992 Oct 8 [cited 2020 May 27];70(10):44–9. Available from: https://linkinghub.elsevier.com/retrieve/pii/000291499291357A
22. Vetter T, Lohse MJ. Magnesium and the parathyroid. Curr Opin Nephrol Hypertens [Internet]. 2002 Jul [cited 2020 Jun 2];11(4):403–10. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12105390
23. Cooper MS, Gittoes NJL. Diagnosis and management of hypocalcaemia. BMJ [Internet]. 2008 Jun 7 [cited 2020 Jun 28];336(7656):1298–302. Available from: http://www.ncbi.nlm.nih.gov/pubmed/18535072
24. Mahadevan S, Bharath R, Kumaravel V, Velayutham P, Muthukumaran J, Premkumar A, et al. Estimation of magnesium in patients with functional hypoparathyroidism. Indian J Endocrinol Metab [Internet]. 2014 Jan 1 [cited 2020 Jun 29];18(6):821. Available from: http://www.ijem.in/text.asp?2014/18/6/821/141365
25. Gunnarsson Ö, Indridason ÓS, Franzson L, Sigurdsson G. Factors associated with elevated or blunted PTH response in vitamin D insufficient adults. J Intern Med [Internet]. 2009 Apr 1 [cited 2020 Jun 29];265(4):488–95. Available from: http://doi.wiley.com/10.1111/j.1365-2796.2008.02044.x
26. Griffin TP, Murphy M, Coulter J, Murphy MS. Symptomatic hypocalcaemia secondary to PTH resistance associated with hypomagnesaemia after elective embolisation of uterine fibroid. BMJ Case Rep [Internet]. 2013 Jun 19 [cited 2020 Jun 29];2013:bcr2013008708. Available from:

http://www.ncbi.nlm.nih.gov/pubmed/23784757

27. Tørnqvist-Johnsen C, Schnabel T, Gow AG, Del-Pozo J, Milne E, Macklin V, et al. Investigation of the relationship between ionised and total calcium in dogs with ionised hypercalcaemia. J Small Anim Pract [Internet]. 2020 Apr 11 [cited 2020 Mar 16];61(4):247–52. Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/jsap.13109

28. Geffré A, Concordet D, Braun J-P, Trumel C. Reference Value Advisor: a new freeware set of macroinstructions to calculate reference intervals with Microsoft Excel. Vet Clin Pathol [Internet]. 2011 Mar [cited 2020 Oct 31];40(1):107–12. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21366659

29. Coady M, Fletcher DJ, Goggs R. Severity of Ionized Hypercalcemia and Hypocalcemia Is Associated With Etiology in Dogs and Cats. Front Vet Sci [Internet]. 2019 Aug 22 [cited 2020 Mar 16];6:276. Available from: http://www.ncbi.nlm.nih.gov/pubmed/31508432

30. Holowaychuk MK, Hansen BD, DeFrancesco TC, Marks SL. Ionized hypocalcemia in critically ill dogs. J Vet Intern Med [Internet]. 2009 May [cited 2020 Mar 16];23(3):509–13. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19298610

31. Coady M, Fletcher DJ, Goggs R. Severity of Ionized Hypercalcemia and Hypocalcemia Is Associated With Etiology in Dogs and Cats. Front Vet Sci [Internet]. 2019 [cited 2019 Nov 8];6:276. Available from: http://www.ncbi.nlm.nih.gov/pubmed/31508432

32. Levi J, Massry SG, Coburn JW, Llach F, Kleeman CR. Hypocalcemia in magnesium-depleted dogs: evidence for reduced responsiveness to parathyroid hormone and relative failure of parathyroid gland function. Metabolism [Internet]. 1974 Apr [cited 2020 Jun 16];23(4):323–35. Available from: http://www.ncbi.nlm.nih.gov/pubmed/4817361

33. DiNicolantonio JJ, O'Keefe JH, Wilson W. Subclinical magnesium deficiency: a principal driver of cardiovascular disease and a public health crisis. Open Hear [Internet]. 2018 [cited 2020 Jun 29];5(1):e000668. Available from: http://www.ncbi.nlm.nih.gov/pubmed/29387426

34. Woods GA, Willems A, Hurst E, Mellanby RJ. Epileptic seizure in a cocker spaniel associated with hypocalcaemia, hypovitaminosis D and a protein-losing enteropathy. Vet Rec Case Reports [Internet]. 2019 Apr 11 [cited 2019 Jun 7];7(2):e000813. Available from: http://vetrecordcasereports.bmj.com/lookup/doi/10.1136/vetreccr-2019-000813

35. Allenspach K, Rizzo J, Jergens AE, Chang YM. Hypovitaminosis D is associated with negative outcome in dogs with protein losing enteropathy: a retrospective study of 43 cases. BMC Vet Res [Internet]. 2017 Apr 8 [cited 2019 Jul 4];13(1):96. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28390394

36. Mellanby RJ, Mellor PJ, Roulois A, Baines EA, Mee AP, Berry JL, et al. Hypocalcaemia associated with low serum vitamin D metabolite concentrations in two dogs with protein-losing enteropathies. J Small Anim Pract [Internet]. 2005 Jul [cited 2019 Jun 4];46(7):345–51. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16035452

37. Gow AG, Else R, Evans H, Berry JL, Herrtage ME, Mellanby RJ. Hypovitaminosis D in dogs with inflammatory bowel disease and hypoalbuminaemia. J Small Anim Pract [Internet]. 2011 Aug [cited 2020 Mar 16];52(8):411–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21797872

38. Schenck PA, Chew DJ. Prediction of serum ionized calcium concentration by use of serum total calcium concentration in dogs. Am J Vet Res [Internet]. 2005 Aug [cited

2020 Mar 16];66(8):1330–6. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16173474

39. Schenck PA, Chew DJ. Prediction of serum ionized calcium concentration by serum total calcium measurement in cats. Can J Vet Res [Internet]. 2010 Jul [cited 2020 Mar 16];74(3):209–13. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20885845

40. Gauci C, Moranne O, Fouqueray B, de la Faille R, Maruani G, Haymann J-P, et al. Pitfalls of measuring total blood calcium in patients with CKD. J Am Soc Nephrol [Internet]. 2008 Aug [cited 2020 Jun 28];19(8):1592–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/18400941

41. Swaminathan R. Magnesium metabolism and its disorders. Clin Biochem Rev [Internet]. 2003 May [cited 2020 Jun 16];24(2):47–66. Available from: http://www.ncbi.nlm.nih.gov/pubmed/18568054

42. Johansson M, Whiss PA. Weak relationship between ionized and total magnesium in serum of patients requiring magnesium status. Biol Trace Elem Res [Internet]. 2007 Jan [cited 2020 Oct 31];115(1):13–21. Available from: http://www.ncbi.nlm.nih.gov/pubmed/17406070

43. Külpmann W, Gerlach M. Relationship between ionized and total magnesium in serum. Scand J Clin Lab Invest Suppl [Internet]. 1996 [cited 2021 Jan 9];224. Available from: https://pubmed.ncbi.nlm.nih.gov/8865441/

44. Saha H, Harmoinen A, Karvonen A, Mustonen J, Pasternack A. Serum ionized versus total magnesium in patients with intestinal or liver disease. Clin Chem Lab Med [Internet]. 1998 [cited 2021 Jan 9];36(9). Available from: https://pubmed.ncbi.nlm.nih.gov/9804396/

45. Elin RJ. Magnesium metabolism in health and disease. Dis Mon [Internet]. 1988 Apr [cited 2020 Oct 31];34(4):161–218. Available from: http://www.ncbi.nlm.nih.gov/pubmed/3282851