

I really thank the Authors for the work made during the revision. The manuscript appears improved thanks to these changes. I still have some doubts about the utility of publishing preliminary data from a small population of animals, because these data may be statistically biased (due to the limited sample size) and, consequently, also the scientific content of the research may be affected. Since Authors have explained that they have more samples and data from other 40 dogs, it would be more interesting to group together Golden retrievers from this manuscript (n: 10) with those that still need to be evaluated (n: 40) to obtain a final sample size of 50 dogs. Nevertheless, if Authors and Editors have a different opinion and still prefer to proceed with the publication of this pilot study, I have some additional comments that I hope could be useful for the manuscript.

Title

I suggest to introduce the term “preliminary data”. as Authors have said that they likely will publish further data on this topic from a larger sample size. Moreover, I would suggest to introduce the numbers of dogs from each breed also in the title, so that readers can easily understand that data from this study should be interpreted cautiously, as it is a pilot study providing preliminary data from a small study population. According, the final version of the title should be: “Preliminary data on echocardiographic evaluation and serum taurine concentration in healthy dogs of two breeds (10 Golden Retrievers and 12 German Shorthaired Pointers) with different predisposition to nutritional dilated cardiomyopathy: a pilot study”.

Simple summary

-Line 19: What does it mean “reduction of the cardiomyocytes hypertrophy”? Does it mean that a low VitD concentration may induce atrophy of cardiomyocytes? Please, explain it better.

-Line 21: “Because taurine is involved in the digestion and absorption of fat and liposoluble vitamins”. Is VitD included among vitamins regulated by taurine? If yes, please change this sentence to: Because taurine is involved in the digestion and absorption of fat and liposoluble vitamins, including vitamin D,”

-Lines 24-25: “Our results showed that GRs presented lower levels of serum taurine and more altered echocardiographic variables than GSPs”. Given the limits of the study (first of all, the small sample size), I suggest to rewrite this sentence as: “The preliminary findings from this small pilot study showed that GRs may present lower levels of serum taurine and more altered echocardiographic variables than GSPs”).

Abstract

-Lines 30-31: “Because taurine is involved in the digestion and absorption of fat and liposoluble vitamins, the aims of... Also in this sentence I would add the terms, “including vitamin D” (if this is scientifically correct), so that the final result would be: “Because taurine is involved in the digestion and absorption of fat and liposoluble vitamins, including vitamin D, the aims of...”

Introduction

-Lines 50-56: among the list of differential diagnosis for a DCM-like phenotype, it should be included also ischemic heart disease. Moreover, I suggest to introduce here the following additional references from manuscripts and congress abstract:

-manuscripts: Schneider SM, Coleman AE, Guo LJ, Tou S, Keene BW, Kornegay JN. Suspected acute myocardial infarction in a dystrophin-deficient dog. *Neuromuscul Disord* 2016;26:361-6. // Romito G, Cipone M. Transient deep and giant negative T waves in dogs with myocardial injury. *J Vet Cardiol* . 2021 Aug;36:131-140. // Driehuys S, Van Winkle TJ, Sammarco CD, Drobatz KJ. Myocardial infarction in dogs and cats: 37 cases (1985-1994). *J Am Vet Med Assoc* 1998;213:1444-8. // Falk T, Jönsson L. Ischaemic heart disease in the dog: a review of 65 cases. *J Small Anim Pract* 2000;41:97-103.

-congress abstracts: Lekane M, Connolly D, Smets P, Borgeat K, Casamian-Sorrosal D, Boswood A, Luis Fuentes V, Gommeren K, Merveille AC. Clinical, ECG and echocardiographic findings in a canine case series of presumptive myocardial infarction. *J Vet Intern Med* 2020;34:403-4.

-Line 69: typing error “[,11,12]”.

-Line 82: typing error “bredds”

-Lines 110-111: “This would increase their intraluminal concentrations with possible improvement in absorption of fat and liposoluble vitamins [21]”. A greater absorption of vitamin D due to the positive role of taurine has been demonstrated in humans? If yes, change the sentence in this way: “This would increase their intraluminal concentrations with possible improvement in absorption of fat and liposoluble vitamins, including vitamin D [21]”. Moreover, please say if a relationship between VitD and taurine has been studied also in dogs and if similar results have been found in this species (because not always what is found in humans is identical in dogs).

-Line 111: add the word “pilot” so that the final version of the sentence become: “The aim of this pilot study...”. Please, add the word “pilot” in all other part of the manuscript (e.g., line 119), so that it becomes really clear to readers the type of study.

-Lines 134-135: Please change this sentence in this way: “Furthermore, to estimate serum taurine and vitamin D3, concentration venous blood sample was collected in fastened dogs (12-8h).”

-Line 155 and 158: there are a lot of a acronyms that appear here for the first time. It is essential to provide initially the extended explanation of the abbreviation (e.g., EDVI = end-diastolic left ventricular volume indexed). Then, it is important to check before submission how many times each abbreviation is used, in order to maintain only those used sufficient times according to rules of the Journal. Lastly, I am not sure that is allowed to start a sentence with an abbreviation.

Results

-Section 3.1. named “Golden Retriever”: all this section has been written using italics.

Discussion

-Line 264-265: “In our study, this aspect is confirmed by the finding of a lower serum taurine concentration in the GRs, ...”. I don’t agree with the use of the term “is confirmed”, as a small study on 10 Golden retrievers cannot confirm a breed predisposition. Therefore, I suggest to rewrite this sentence as: “Our results appear overall in line with previous literature in light of the finding of a lower serum taurine concentration in the GRs, ...”.

-lines 269-273: at the end of this long sentence I suggest to add "..., although further studies including more dogs are needed to conclusively confirm this".

-Lines 285-286: "The results suggests that the serum and plasma taurine concentration does not differ..." . I suggest to introduce the word "may" to make less strong the statement, as it is difficult to obtain conclusive results given the small sample size. Moreover, there is a typing error. Accordingly, the final version of the sentence would be: "The results suggest that the serum and plasma taurine concentration may not differ..." .

-Lines 328-331: "The higher values of the parameters suggestive of ventricular dilation and lower values of those predictive of systolic dysfunction, associated to a low levels of serum taurine, supports the hypothesis of a possible greater predisposition of the GRs to develop DCM." I don't completely agree with this statement as the small sample size of the study does not allow to make definitive conclusions at regards. Indeed, it could be also possible that the parameters suggesting a mildly larger dimensions of the LV and a mildly "more hypokinetic" LV in GR compared to GSP, may be not related to a disease state (i.e., they may be not due to a true hypokinetic cardiomyopathy related to taurine) but they may be simply due to physiological differences concerning normal echocardiographic parameters in the two breeds. The only way to know the answer would be to make a study on ≥ 120 healthy GR and ≥ 120 GPS to create statistically reliable normality ranges for standard echocardiographic parameters in these two breeds, and then to compare the reanalyze the results of this study. Therefore, all this should be discussed and clearly explained to readers.

-lines 338-339: "The main limit of this study was the small number of subjects included." I suggest to expand this section in this way: "The main limit of this study was the small number of subjects included, which inevitably weakened our statistical analysis and results. Accordingly, findings from this small pilot study should not be read as a guide for interpretation of taurine concentration as well echocardiographic parameters in these two breeds. In contrast, they should be simply interpreted as preliminary data aimed at paving the way for further and more robust investigations in this topic."

-Additional limitation: an additional limitation that should be included is that Author have selected two breeds for which there are not well-established echocardiographic normality ranges based on statistically reliable sample sizes. This inevitably makes it more difficult to correctly interpret the echocardiographic findings from this study and makes it easier to interpret as abnormal something that maybe could be normal for that given breed. This should be explained to readers.

Conclusions

-lines 361-362: "These echocardiographic alterations/border line values could be considered as a predisposition of this breed to develop a dilatative/hypokinetic myocardial disease". This sentence should be deleted.