Reviewer’s report

Title: Overexpression of miR-9 in mast cells is associated with invasive behavior and spontaneous metastasis

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Reviewer: michele mortarino

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The study is well designed and the results are clearly illustrated.

Discretionary Revisions

1. Methods - Quantitative reverse-transcription-PCR profiling of mature miRNA expression in MCT biopsies, first paragraph:

Normalizer gene for miRNA expression analysis. The authors selected snRNA U6 as a normalizer gene after miRNA array due to its stable expression in the sample set. In the past years, small RNAs other than miRNAs, like U6, were reported to be unsuitable reference genes for miRNA expression data in different human and canine tumours (e.g. see Peltier and Latham, 2008. Normalization of miRNA expression levels in quantitative RT-PCR assays: Identification of suitable reference RNA targets in normal and cancerous human solid tissues. RNA 14, 844-852; Mortarino et al., 2010. Identification of suitable endogenous controls and differentially expressed miRNAs in canine fresh frozen and FFPE lymphoma samples. Leukemia Research 34: 1070-1078). A short statement discussing the result from the present study in comparison with abovementioned studies should be added to the manuscript.

2. Methods – RNA isolation and quantitative real-time PCR, second paragraph:

Normalizer gene for mRNA expression analysis. The authors should explain the rationale of selecting 18S gene as reference gene.

3. Results – miR9 is overexpressed in biologically high-grade canine MCTs

Case H11, corresponding to a dog with biologically high-grade MCT, clustered with the biologically low-grade tumors after miRNA array (Figure 1) and expressed miR-9 as lowly as low-grade tumors except L12 (Figure 2A). This unexpected result should be discussed in the manuscript as done for case L12.

Minor essential revisions
Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:

I declare that I have no competing interests