Author’s response to reviews

Title: The synthetic peptide P111-136 derived from the C-terminal domain of heparin affin regulatory peptide inhibits tumour growth of prostate cancer PC-3 cells

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Author’s response to reviews: see over
Dear Editor,

Please, find enclosed a revised version of our previous manuscript ref 1373169867514307 untitled “The synthetic peptide P111-136 derived from the C-terminal domain of heparin affin regulatory peptide inhibits tumour growth of prostate cancer PC-3 cells”. According to your mail, we have fully addressed all of the referee 2 concerns (see below the modifications point by point) and highlight in yellow the modifications in the revised manuscript. We hope now our manuscript will be suitable for publication in the journal.

We are looking forward to hearing from you.

Yours sincerely,

Jean Delbé
Referees' 2 Specific Comments:

**Major points**

**Comment 1)** The Western blot added to Fig.1 (Fig.1A) is not so convincing.

**Answer:** According to the referee, we performed a new experiment using more PC-3 cells than previously to give a better signal from the conditioned media. The resulting Western blot was replaced in Figure 1A of the revised manuscript.

**Comment 2)** the reply to the major comment 3a is quite superficial. In this revised version, the Authors showed that the previously described P122-131 peptide is less effective on growth inhibition of PC3 than DU145 and LnCap cells but the explanation or even a comment of this data need to be more carefully documented… I haven’t found out in the literature that ALK receptor is absent in DU145 and LnCap cells. The possible explanation of different effects for the similar peptide on three different prostate tumor cell lines was not investigated at all.

**Answer:** In the discussion section of the manuscript, we speculated that the difference in efficiency of the P122-131 on PC-3 cells compare to DU145 and LnCap could be due to the absence of ALK expression in DU145 and LnCap. To validate this assumption, we mentioned a recent publication of Diamantopoulou et al. Mol Cancer 2010 (cited reference number 33) page 15 lane 21. We apology for this mistake since the absence of ALK expression in DU145 and LnCap has not been mentioned in this publication. This result was not yet published but presented below (Figure I). Therefore we have added the sentence in the discussion section page 15 lane 23 “It is interesting to mention that these cell lines do not express the ALK receptor (unpublished results).”

![Figure I. Expression of the ALK receptor in PC-3, DU145 and LNCap cells. DU145 and LNCaP were cultured in completed medium as described in Diamantopoulou et al. Mol. Cancer 2010. (cited ref. 33 in the revised manuscript). Western blot (WB) and RT-PCR](image-url)
experiments were performed with respectively lysate and total RNA from PC-3 (positive control), DU145 and LNCaP cells as described in Dos Santos et al. Int. J. Cancer 2010. (cited ref. 28 in the revised manuscript).

According to the remark of the referee concerning the absence of a full investigation of these peptides on the three different prostate cell lines, we have also modified a sentence in the revised manuscript to clarify this point on page 16 lanes 2-3 “but further investigations of both peptides on these different prostate cancer cell lines will be need to clarify this point.”

Comment 3) (Sven Christian et al, J Cell Biol 2003): this reference is cited at page 13 but it cannot be found in the list of references.

Answer: We apology for this mistake, the reference was added in the references section with the number 35.