Reviewer's report

Title: Fibroblast activation protein-alpha-expressing fibroblasts promote the progression of pancreatic ductal adenocarcinoma

Version: 1
Date: 20 April 2015
Reviewer: Marianne Sinn

Reviewer's report:

Thank you for the opportunity to review this interesting manuscript. The authors present data about cancer-associated fibroblasts (CAF) and the role of fibroblast activation protein-alpha (FAP) in pancreatic adenocarcinoma (PDAC). This is a scientifically relevant topic as former investigations suggest an association between the expression of FAP in the peritumoral stroma of PDAC and a worsened prognosis. Furthermore, the peritumoral reaction seems to play a key role in general in the still dismal prognosis of the disease, but is not completely understood so far. To target the peritumoral stroma seems actually to be a promising therapeutic strategy in PDAC.

For the current paper, archival tissue from 48 patients with resected PDAC (from a single centre) were analysed retrospectively for FAP expression. Immunohistochemical staining was performed and correlated with survival data. In addition, preclinical analysis (invasion assay, co-culture, western blot) were done to demonstrate a correlation between FAP expression and invasiveness activation/progression of cell cycle in PDAC cells.

The presented data can contribute to a better understanding of the peritumoral stroma and stromal-tumor cell interactions, especially the role of FAP in PDAC.

Several issues should be addressed or clarified (Minor Essential Revisions):

1, you named a 5-year survival rate for PDAC of 10% in the introduction (page 3, line 21), please specify for which stage.

2, how do you explain the untypical results for the factors associated with overall survival in in your study population (uni-/multivariate analysis): no correlation for post-operative chemotherapy, tumor size, lymph node involvement, but for alcohol intake?

3, Figure 1: survival on the x-axis should be quoted in weeks or months

4, please explain: experiments are carried out with only one pancreatic cancer cell line and one embryologic mouse cell line of fibroblasts. How do you ensure that the individual differences of the tumours are sufficiently represented by this set up? I am particularly critical of the point that you used fibroblasts of another species. Fibroblasts generated from human pancreatic tissue would allow a clearer statement. How can you exclude confounding factors resulting from the use of cells of different species?
5, please specify the method of co-culture further: you said you were using six well culture plates and then culture inserts. Did you use the same culture inserts as for the invasion essay? If so, was there any direct contact of the fibroblasts and the pancreatic cancer cells? If there was any direct contact, did you expect any effect of that? If you use the same methodical set up in invasion essay and coculture please revise the corresponding paragraph.

6, please add: how often did you repeat the experiments on invasiveness?

7, please explain: why did you choose retinoblastoma (Rb) for your analysis? Did you expect any interaction and when yes, why?

**Level of interest:** An article of importance in its field

**Quality of written English:** Acceptable

**Statistical review:** Yes, and I have assessed the statistics in my report.

**Declaration of competing interests:**

I declare that I have no competing interests