Reviewer's report

**Title:** Inhibitory effects of prostaglandin E2 on collagen synthesis and cell proliferation in human stellate cells from pancreatic head adenocarcinoma.

**Version:** 4  **Date:** 10 September 2013

**Reviewer:** Max Bachem

**Reviewer's report:**

This reviewer has not seen the original manuscript, he has only seen the revised version.

Present knowledge: In contrast to COX-1, which is expressed constitutively in several cell types in normal tissue, COX-2 is an inducible enzyme producing primarily prostaglandin E2 (PGE2) at sites of inflammation. Increased COX-2 expression is frequently found in several cancers including 60 – 70 % of cases of pancreatic adenocarcinomas. Furthermore it was shown, that COX-2 correlates with the invasiveness of pancreatic tumors and a shorter overall survival. COX-2 inhibition reduces progression of the disease in mouse models of pancreatic cancer. The product of COX-2 PGE2 plays a role in proliferation, invasion, angiogenesis, chemoresistance and metastasis. As shown by the group of S. Friedman, almost 10 years ago, PGE2 inhibits both basal and TGF# induced collagen synthesis of hepatic stellate cells (Hui AY et al., J Hepatol 41: 251-258, 2004). Interestingly in a human pancreatic stellate cell line PGE2 stimulated cell proliferation, migration and invasion. Furthermore in this study also expression of extracellular matrix proteins and MMPs were stimulated by PGE2 (Charo C et al., Pancreas 42: 467-474, 2013). These effects were mediated via the EP4-receptor.

The present study of Pomianowska et al. investigated the expression of COX-2 in pancreas cancer cells and in stromal cells surrounding pancreas cancer. In addition the authors explored the role PGE2 in stellate cell proliferation and collagen synthesis. As shown by immunohistochemistry COX-2 was expressed in pancreatic cancer cells but not in stromal cells. However, PSCs obtained by the outgrowth method, expressed COX-2 in culture. The COX-2 expression could be induced further by IL-1#, EGF, thrombin and PGE2. Furthermore indirect coculture of PSC with the pancreas cancer cell line BxPC-3 (but not Panc-1) increased COX-2 expression in PSCs. It is known that Panc-1 cells do not express COX-2. Addition of PGE2 to cultured PSCs stimulated cAMP, phosphorylation of ERK and reduced TGF# stimulation of collagen synthesis and PDGF induced cell proliferation. Pretreatment of PSC with indomethacin had no effect on TGF# induced collagen synthesis or PDGF induced proliferation. As shown by specific inhibitors the effect of PGE2 was mediated via EP2 receptors.

The authors conclude from their data that PGE2 is mainly produced by COX-2 expressing pancreatic cancer cells and PGE2 exerts a suppressive effect on proliferation and fibrogenesis in PSC via EP2 receptors. Because PGE2 inhibits
collagen synthesis and PSC proliferation therapeutic inhibition of COX-2 in cancer cells might accelerate fibrosis progression in pancreatic cancer.

The obtained data have value for a better understanding of the interaction between pancreas cancer cells and PSC and the role of COX-2 and of PGE2 hereby.

In the near future in vivo-studies with animal models (e.g. KPC-mice, orthotopic injection of COX-2 pos. pancreas carcinoma cell lines together with PSCs) are urgently needed to elucidate the effect of COX-2 inhibition on tumor desmoplasia.

**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