Reviewer's report

**Title:** Calpain system protein expression in carcinomas of the pancreas, bile duct and ampulla

**Version:** 1  **Date:** 1 June 2012

**Reviewer:** Nigel Jamieson

**Reviewer's report:**

I greatly enjoyed reading this manuscript. It was well-written, easy to follow and presents novel findings in a reasonably well-powered cohort of patients. The calpain system is one that has undergone limited investigation in pancreatic disease so far and merits further investigation. These data in the manuscript have demonstrated that there is variation in expression of calpain-2 expression in pancreatic ductal adenocarcinoma and that this correlated with survival following resection.

The methodology was very clear and the use of the X-tile software to determine optimum cut-off points for prognostic stratification is good to see. Clearly a great deal of rigorous effort has been put into the pathological assessment of the TMA which is very encouraging and supports the findings in the manuscript.

The authors should be commended for following the REMARK criteria which is an important step towards enhancing pancreatic cancer biomarker translational research. This is an issue this has recently been discussed in pancreatic cancer (Jamieson et al, 2011, Clinical cancer research).

The limitations of the work have been acknowledged and the title and abstract accurately convey the findings of the manuscript.

**Major Essential Revisions**

My principal comment is regarding the grouping together of ampullary and bile duct tumours. Based on the differing clinical behavior of Cholangiocarcinoma and Ampullary adenocarcinoma I wonder if the expression pattern of the calpains could be further elucidated by splitting the 2nd TMA set analysis into Ampullary and Cholangiocarcinoma.

This strategy may yield further interesting insight into the variation of calpain expression in the HPB disease.

In particular with regard to the Ampullary cohort is data available subcategorizing patients into intestinal and pancreaticobiliary subtype tumours.

**Minor Essential Revisions**

On page 10 “Relationship with clinical outcome” I believe that on the second last
line the text should read - (Table 3 panel A).

Discretionary Revisions

1) Would it be possible to describe the operative interventions for the cohorts, in particular were patients resected with curative intent, were all patients resected by pancreaticoduodenectomy or were distal pancreatectomies included?

2) Was resection margin status available for this cohort? Resection margin status is an important prognostic factor and should ideally be included although the relatively long duration of the cohort study will impact on the quality of pathology reporting over this time.

3) Did chemotherapy impact significantly on survival for the pancreatic, ampullary or cholangiocarcinoma patients? Did the use of chemotherapy alter the prognostic impact of calpain2/ calstatin

**Level of interest:** An article whose findings are important to those with closely related research interests

**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