PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Characterization of the fibro-inflammatory process involved in progression from acute to chronic pancreatitis: study protocol for a multicentre, prospective cohort study
AUTHORS	Novovic, Srdan; Borch, Anders; Werge, Mikkel; Karran, David; Gluud, Lise; Schmidt, Palle; Hansen, Erik; Nøjgaard, Camilla; Jensen, Annette; Jensen, Frank; Frøkjær, Jens Brøndum; Hansen, Mark; Jørgensen, Lars; Drewes, Asbjørn; Olesen, Søren

VERSION 1 – REVIEW

REVIEWER	Cosimo sperti Department of Surgery, Oncology and Gastroenterology, University of Padua, Padua, Italy
REVIEW RETURNED	05-Feb-2019

GENERAL COMMENTS	The topic of this manuscript is interesting and the protocol is well-described. However, statistical analysis is not reported; so, it is not clear how evaluate the change of biomarkers, pain perception, nutritional status and imaging features at every event. Moreover I am not sure that 50 patients in each cohort is a valid sample size.

REVIEWER	A/Professor Max Petrov University of Auckland
REVIEW RETURNED	26-Feb-2019

GENERAL COMMENTS	The following comments may help to improve the manuscript: - The sample size calculations are based on dubious assumptions or studies published a long time ago. The use of up-to-date epidemiological estimates, as reported in PMID 30482911, would be more appropriate. - The authors do themselves a disfavour by not covering in the introduction the HPP (holistic prevention of pancreatitis) framework - PMID 30482911. - A protocol for a another longitudinal study in the field has recently been published (PMID 30325862). Please discuss the similarities and dissimilarities between the two and what new data your study could possibly add. - It is unclear what centers are going to participate in the study. And how the number of centers would relate to recruiting the desired number of patients.

REVIEWER	PRAMOD GARG A.I.I.M.S., New Delhi
REVIEW RETURNED	13-Apr-2019

GENERAL COMMENTS	I have evaluated the protocol 'Characterisation of the fibro-inflammatory process involved in progression from acute to chronic pancreatitis: study protocol for a multicentre, prospective cohort study'. It is a well conceptualised study protocol that is likely to provide new information about the pathophysiology of chronic pancreatitis. The following comments might be helpful: 1. The primary outcome is to understand the pathophysiological basis of the development of CP through a sequence of AP to RAP to CP. The likely percentage of patients developing RAP from AP is around 15-25% and from RAP to CP is around 30-50%. In that case, the number of patients developing CP amongst those with AP will be 8-10 out of 50 and 15-20 amongst those with RAP. The sample size and the number of events of interest (i.e. CP) seems to be small. In my opinion, the investigators should include more number of patients with AP and RAP, at least 100 each. 2. What advice and treatment will be given to patients with RAP and early CP during follow up? The treatment provided is likely to be an important modifier of the disease progression and thus a significant confounder when analysing the factors associated with disease progression. 3. What treatment will be offered to patients with alcohol and smoking induced RAP and early CP regarding abstinence? This will also have an effect on disease progression. The authors should include a plan of analysis a priori to compare disease progression between those who quit alcohol and smoking versus those who don't. 4. The authors should consider genetic analysis of the commonly reported mutations/ polymorphisms in certain genes such as PRSS1, SPINK1, CFTR, MORC4 etc. as important risk factors and see what is the influence of genetic mutations on disease progression.

VERSION 1 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:

Reviewer: 1
Reviewer Name: Cosimo sperti
Institution and Country: Department of Surgery, Oncology and Gastroenterology, University of Padua, Padua, Italy Please state any competing interests or state 'None declared': I have no competing interests

Please leave your comments for the authors below The topic of this manuscript is interesting and the protocol is well-described. However, statistical analysis is not reported; so, it is not clear how evaluate the change of biomarkers, pain perception, nutritional status and imaging features at every event. Moreover, I am not sure that 50 patients in each cohort is a valid sample size.

We thank the reviewer for this important comment pertaining to sample size estimation. After careful consideration, we have revised the study cohorts to better match with the recent study protocol published by the Consortium for the Study of Chronic Pancreatitis, Diabetes, and Pancreatic Cancer (CPDPC) (Yadav et al. Pancreas 2019). We report the revised inclusion criteria in the methods section. In addition, we have increased the sample size in the revised patient cohorts to 60 patients, which is in line with recently proposed estimates from a MRI based study protocol from the CPDPC (Tirkes el al. Abdominal Imaging 2019). Based on current epidemiological estimates we thus

anticipate that 20-30 patients with suspected CP will progress to definitive CP during the study period, which is generally believed to be an adequate sample size for obtaining clinical meaningful group differences for MRI based assessment parameters.

Reviewer: 2
Reviewer Name: A/Professor Max Petrov
Institution and Country: University of Auckland Please state any competing interests or state 'None declared': None declared

Please leave your comments for the authors below. The following comments may help to improve the manuscript:
- The sample size calculations are based on dubious assumptions or studies published a long time ago. The use of up-to-date epidemiological estimates, as reported in PMID 30482911, would be more appropriate.

We are very grateful to the reviewer for stressing this important point. The epidemiological estimates and references have been updated as suggested. In particular, the limited progression rate of patients with a single episode of acute pancreatitis is of great relevance to this study. After careful consideration, we have decided to take the study cohort comprising of patients with a single episode of acute pancreatitis out of the protocol. In addition, we have revised the remaining study cohorts to better match with the recent study protocol published by the Consortium for the Study of Chronic Pancreatitis, Diabetes, and Pancreatic Cancer (CPDPC) (Yadav et al. Pancreas 2019). We report the revised inclusion criteria in the methods section. In addition, we have increased the sample size in the revised patient cohorts to 60 patients, which is in line with recently proposed estimates from a MRI based study protocol from the Consortium for the CPDPC (Tirkes el al. Abdominal Imaging 2019).

- The authors do themselves a disfavour by not covering in the introduction the HPP (holistic prevention of pancreatitis) framework - PMID 30482911.

Thank you very much for this suggestion. We have read the proposed paper, which we find very interesting and relevant in this context. We have added a paragraph and reference in the introduction as suggested.

- A protocol for another longitudinal study in the field has recently been published (PMID 30325862). Please discuss the similarities and dissimilarities between the two and what new data your study could possibly add.

We have read with great interest the study proposals from our colleagues from the US (Yadav et al. Pancreas 2019 and Tirkes et al. Abdominal Radiology 2019) and included a discussion on the (dis)similarities between study protocols. Additionally, and as outlined above, we have revised the study cohort of our study to better match with the study cohorts presented in the PROCEED study.

- It is unclear what centers are going to participate in the study. And how the number of centers would relate to recruiting the desired number of patients.

We have clarified this in the methods section.

Reviewer: 3
Reviewer Name: PRAMOD GARG
Institution and Country: A.I.I.M.S., New Delhi Please state any competing interests or state 'None declared': None

Please leave your comments for the authors below I have evaluated the protocol 'Characterisation of the fibro-inflammatory process involved in progression from acute to chronic pancreatitis: study protocol for a multicentre, prospective cohort study'. It is a well conceptualised study protocol that is likely to provide new information about the pathophysiology of chronic pancreatitis. The following comments might be helpful:

1. The primary outcome is to understand the pathophysiological basis of the development of CP through a sequence of AP to RAP to CP. The likely percentage of patients developing RAP from AP is around 15-25% and from RAP to CP is around 30-50%. In that case, the number of patients developing CP amongst those with AP will be 8-10 out of 50 and 15-20 amongst those with RAP. The sample size and the number of events of interest (i.e. CP) seems to be small. In my opinion, the investigators should include more number of patients with AP and RAP, at least 100 each.

We thank the reviewer for this important comment. As also stated for reviewer #1 and reviewer #2: "…the limited progression rate of patients with a single episode of acute pancreatitis is of great relevance to this study and after careful consideration, we have decided to take the study cohort comprising of patients with a single episode acute pancreatitis out of the protocol. In addition, we have revised the remaining study cohorts to better match with the recent study protocol published by the Consortium for the Study of Chronic Pancreatitis, Diabetes, and Pancreatic Cancer (CPDPC) (Yadav et al. Pancreas 2019). We report the revised inclusion criteria in the methods section. In addition, we have increased the sample size in the revised patient cohorts to 60 patients, which is in line with recently proposed estimates from a MRi based study protocol from the CPDPC (Tirkes el al. Abdominal Imaging 2019). Based on current epidemiological estimates we thus anticipate that 20-30 patients with suspected CP will progress to definitive CP during the study period, which is generally believed to be an adequate sample size for obtaining clinical meaningful group differences for MRI based assessment parameters".

2. What advice and treatment will be given to patients with RAP and early CP during follow up? The treatment provided is likely to be an important modifier of the disease progression and thus a significant confounder when analysing the factors associated with disease progression.

This is an observational study and patients are counselled and treated according to best clinical practice, including advice on alcohol and smoking cessation. We have included a paragraph addressing this in the methods section.

3. What treatment will be offered to patients with alcohol and smoking induced RAP and early CP regarding abstinence? This will also have an effect on disease progression. The authors should include a plan of analysis a priori to compare disease progression between those who quit alcohol and smoking versus those who don't.

The reviewer raises an important point, which we have already considered in the study design. Accordingly, detailed information on alcohol and smoking consumption is registered at baseline and annually during the follow-up visits. The retrieved information will be included in analyses of assessment parameters to account for the influence of e.g. continued smoking. This has been further detailed in the methods section.

The participating centres all provide counselling against alcohol misuse and smoking as part of routine clinical practice as outlined in our response above.

4. The authors should consider genetic analysis of the commonly reported mutations/ polymorphisms in certain genes such as PRSS1, SPINK1, CFTR, MORC4 etc. as important risk factors and see what is the influence of genetic mutations on disease progression.

This is an excellent suggestion for which we thank the reviewer. We have decided to collect suitable blood samples in order to have the possibility to perform genetic analyses on a later stage if deemed relevant and following approval from our Ethics committee. This information is now added in the methods section.

VERSION 2 – REVIEW

REVIEWER	Cosimo Sperti Department of Surgery, Oncology and Gastroenterology, 3rd Surgical Clinic, University of Padua, Via Giustiniani 2, 35128 Padua, Italy
REVIEW RETURNED	20-Jun-2019

GENERAL COMMENTS	The manuscript has been improved in this revised form

REVIEWER	Pramod Kumar Garg All India Institute of Medical Sciences, New Delhi
REVIEW RETURNED	18-Jun-2019

GENERAL COMMENTS	I think the revised version is OK