

June 5, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 2992-review.doc).

Title: Biliary phosphatidylcholine and lysophosphatidylcholine profiles in sclerosing cholangitis

Authors: Annika Gauss, Robert Ehehalt, Wolf-Dieter Lehmann, Gerhard Erben, Karl-Heinz Weiss, Yvonne Schaefer, Petra Kloeters-Plachky, Adolf Stiehl, Wolfgang Stremmel, Peter Sauer, Daniel Nils Gotthardt

Name of Journal: *World Journal of Gastroenterology*

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The manuscript has been improved according to the suggestions of the reviewers.

Also, a native English speaker has revised the manuscript. Changes are marked in yellow.

A. Concerning major issues

1. We feel the reviewer is right about his critique on potential dilution effects on the results of total biliary PC and LPC concentrations. Unfortunately, it was not noted for the patients whether the bile samples could be obtained right away before any manipulation or whether contrast medium was first applied to confirm cannulation of the common hepatic duct before sample acquisition. Thus we are not able to provide these data in the study. Our common experience is, though, that primary bile acquisition is possible in about 90% of patients, and to be more certain about potential dilution effects, we removed about the same volume of contrast medium that was injected before bile acquisition in cases that it was not possible right away.

We think it is appropriate to acknowledge the reviewer's concerns and to adapt the discussion and conclusion of the manuscript in a way that our study does not give enough evidence of changed total PC and LPC concentrations in bile from patients with CCC and SSC (article was changed in this respect). After all, one of the reasons why we chose to calculate relations of PC and LPC contents to other bile compounds such as bile acids was that we apprehended false data interpretation by unknown effects of dilution. Concerning the two bile samples in the group of patients with CCC which had to be excluded due to their colorlessness, the reason for this was most probably not dilution by contrast medium, but extreme cholestasis with serum bilirubin levels of up to 30 mg/dl in these patients, which usually results in colorless bile. As can be viewed in Table 1, serum bilirubin levels in the included patients did

not differ significantly between the groups, so that we think that data interpretation should not be significantly affected by this circumstance.

2. In response to the second major point of critique, we removed the section *"Intra-individual variability of bile phospholipids in two patients with PSC"* as well as the related Table 5 from the manuscript.

3. Concerning the detailed information about bacterial culture results, the first paragraph of the section *"LPC concentration and LPC/PC ratio"* was taken out, and the section *"Bacterial cultures and biliary phospholipids"* was shortened.

B. Concerning minor issues:

1. As indicated by the reviewer, we shortened the *"Materials and Methods"* as well as the *"Results"* section in the abstract, even though we had understood that the number of words we used for the different sections of the abstract was recommended in this way by the journal instructions to authors.

2. The term "subjects" was changed into "patients" in the whole manuscript.

3. The indicated sentence was removed from the manuscript.

4. Some information on the indicated subject was added in the introduction of the manuscript.

5. Concerning sample numbers, our goal was to include up to 14 patients in every group, depending on availability. Most samples were available for PSC patients, as the Department has much expertise for this disease. Availability also depended to a high degree on how much of the sample was left for all the measurements needed.

concerning the control group, it was not easy to find patients meeting the criteria of having bile ducts stones without signs of inflammation. As far as the CCC and SSC groups are concerned, samples were very precious as they were rare in the sample bank, and as they were also needed for other studies. This information was introduced into the *"Materials and Methods"* section of the manuscript.

Comments to Reviewer 00070628:

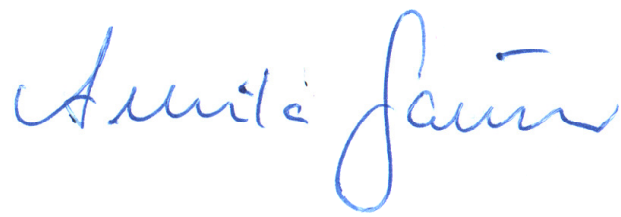
1. The problem of potential UDCA intake was discussed in the manuscript. Unfortunately, practically all patients with PSC being treated in our Department take UDCA, so that reasonable formation of subgroups was not possible in this study.

2. Analyzing subgroups of IBD and non-IBD patients would be an interesting point, as our group has previously shown that intestinal mucus PC is reduced in patients with ulcerative colitis versus controls. There might also be a change in bile from these patients. Yet, in the present study, 10 out of 14 patients with PSC and both patients with CCC/PSC suffered from IBD, so that subgroup analysis was again not performed due to sample sizes being too small.

3. Unfortunately, we did not have sufficient information on the intake of statins in this study.

We hope that all comments were adequately responded to and are open to any other questions or comments.

Yours sincerely,



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