Author's response to reviews

Title: Expression of a novel carbonic anhydrase, CA XIII, in normal and neoplastic colorectal mucosa. Comparison to other cytosolic isozymes, CA I and II

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Author's response to reviews: see over
Dear editor,

We were grateful for the constructive critiques our manuscript (1972724108559122) received and feel that we were able to respond to questions or suggestions with changes that greatly improved the manuscript. I hope that you will find the revised version of the manuscript acceptable for publication in the BMC CANCER.
Attached is a listing of the detailed responses to each of the reviewer’s comments.

Sincerely yours,

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Response to the comments from the referee 1:

1. The reviewer asked whether there is any possibility that the produced anti-CA XIII serum would cross-react with other CA isozymes. The closest homologs are the other cytosolic isozymes, CA I, II and III. Based on our studies, we know that this antiserum shows a very distinct distribution pattern for CA XIII. It is clearly different when compared to CA I and II which were included in the present paper. It is also different from CA III which is mainly expressed in the skeletal muscle. We have added the following information (Page 5, line 5): “In immunohistochemical staining, CA XIII has shown a distinctly different distribution pattern compared to the localization of CA I or II [2]. This finding indicates that the anti-CA XIII antibody does not cross-react with CA I or II under the present staining conditions.”

2. The reviewer pointed out that the discussion was far from the data presented in the manuscript. We have modified the discussion by deleting one paragraph describing the protein-protein associations. In addition, we have shortened the third paragraph of the Results and Discussion.

3. The reviewer pointed out that it would be interesting to study chromosomal alterations related to CA genes in colorectal tumors. Even though it would be certainly very interesting, it is technically a complicated issue. What could be done is an experiment with fluorescence in situ hybridization (FISH) technique. We contacted two specialists in genetics field and both of them were suspicious for such an experiment using this material. FISH is a relatively rough method, which often gives wrong negative results. Because of this technical problem, a much larger collection of samples would be required in order to obtain reliable results. The last paragraph of the Results and Discussion has been modified and we added some new information related to the genetic alterations which have been detected earlier in the colorectal tumors. We also took into account a possibility that downregulation of CAs could result from reduced levels of a common transcription factor. This suggestion was also included in the Abstract (Page 2, line 14) and Conclusions (Page 9, line 2).

4. The introduction has been shortened as suggested by the reviewer.
5. The reviewer correctly pointed out that there was an error in counting the number of lesions in the Materials and Methods. The graphic summary (Figure 3) was already based on right numbers of the samples. We have corrected the text as follows (Page 5, the 3rd paragraph): “32 distinct areas (both normal tissue and pathological lesions) of the human colorectal mucosa were examined from 12 patients. They consisted of 11 separate samples of histologically normal human colon or rectum and 17 colorectal lesions, including 6 adenomas and 11 adenocarcinomas. In 4 samples, normal tissue was found adjacent to the neoplasm and was included in the analyses. The grade of dysplasia was moderate in 3 and grave in 3 adenomas. The group of 11 malignant colorectal tumors consisted of 4 well-differentiated and 4 moderately differentiated adenocarcinomas, and 3 adenocarcinomas with a mucinous component. It should be noted that each patient had either one or more (1-3) lesions which were analyzed.”