Reviewer’s report

Title: Clinical implications of thymidylate synthetase, dihydropyrimidine dehydrogenase and orotate phosphoribosyl transferase activity levels in colorectal carcinoma following radical resection and administration of adjuvant 5-FU chemotherapy

Version: 4 Date: 10 July 2007

Reviewer: Silke Lassmann

Reviewer’s report:

General

In the submitted study the authors addressed the question of whether or not measurement of the activity levels of thymidylate synthase, dihydropyrimidine dehydrogenase and orotate phosphoribosyl transferase are of “prognostic value” for colorectal cancer patients receiving adjuvant 5FU.

The included 18 patients with Dukes C and 22 patients with Dukes B colorectal cancer, all of whom had received adjuvant oral uracil-tegafur (UFT) therapy. The study cohort is quite small and hence - upon taking into account the clinico-pathological characteristics – conclusions about the “prognostic value” of the measured enzymes are a bit adventurous. Moreover, the differences of enzyme activity observed in the “tumor” samples may be simply due to differences in the content of tumor cells within the “tumor sample”, as the “tumor” may contain various percentages of tumor and stromal (e.g. lymphocytes) cells. Other specific queries arise due to misleading values in e.g Table 2, as specified below.

Except some mistakes and/or misleading sentences and data as well as rather long Materials and Methods as well as Discussion sections, the manuscript is well written and generally presented in adequate Tables and Figures.

Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)

To strengthen the results of enzyme levels in “normal” and “tumor” samples and their clinical impact, several aspects should be addressed:

1) Investigation of an increased number of patients, with Duke’s B and Duke’s C cancer, so as to be able to analyse the impact of enzyme levels within these two (known) prognostic patient sub-groups is highly encouraged. At present, the authors have mixed Duke B and C cases for survival analysis.

Moreover, the selection of the cut-offs for “low” and “high” enzyme levels need to be clarified for correlation of enzyme levels with patient follow up.
Also, why is it in Table 2 that “N(-)” and “N(+)” cases are similar in numbers (n= 22 and 18, respectively) to those listed under “Duke B” and “Duke C”, but that enzyme activities are different (TS, i.e. probably just a typing error? Please check values)?

2) Correlation of enzyme levels to the number of tumor cells within the “tumor” samples, to rule out that enzyme activities simply reflect the number of tumor versus stromal cells present in the harvested tissues.

3) With the above points, a multivariate analysis including known prognostic markers should be performed to conclude on the value of the quite laborious analysis of TS, DPD and OPRT enzyme activities for the clinical setting.

Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

Materials and Methods:

3) Why using both “refrigerated” and “fresh frozen” tissues? Does this impact enzyme activities? The paragraph describing this is confusing with “Tissue was taken from….and immediately stored in a refrigerator …and stored in -80°C. TS, DPD and OPRT activity in refrigerated tumor and healthy tissues was examined. TS, DPD and OPRT activity in fresh-frozen specimens from tumor and non-tumor tissues were examined”. Also, strictly speaking, if the tissues were refrigerated before freezing, they are not “fresh-frozen”.

4) In the description of OPRT activity measurements, there is a redundant sentence “the filter paper was placed….[3H]-fluorouridine monophosphate (FUMP) formed was quantified to determine the concentration of (FUMP) formed was quantified to determine th concentration of FUMP. The reaction rate was obtained …”

5) Please specify how cut-offs for “high” and “low” activity groups were selected.

Discussion:

6) Last sentence of the second paragraph (“These findings suggest that if OPRT activity were examined preoperatively a determination could be made as to whether lymphadenectomy is an appropriate surgical intervention”) is highly speculative (see 1 above) and should be toned done or left out completely.

7) Please shorten and focus the discussion.

Discretionary Revisions (which the author can choose to ignore)

none
What next?: Unable to decide on acceptance or rejection until the authors have responded to the major compulsory revisions

Level of interest: An article of limited interest

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:

Please note that I do not wish my name to be given to the authors.

I declare that I have no competing interests.