Reviewer's report

Title: Title: Impact of KRAS, BRAF and PI3KCA mutations in rectal carcinomas treated with neoadjuvant radiochemotherapy and surgery

Version: 5 Date: 8 March 2013

Reviewer: Bridget Charbonneau

Reviewer's report:

Discretionary revisions:

Specific Comments:
1. Add a space between Materials and Methods and Results section in your abstract.
2. Page 3: Lines 5-6: Replace “metastatic spreading” with “metastases”
3. Page 3: 2nd paragraph: Line 9: Replace “as a poor prognosis factor” with “as poor prognosis factors”
4. Page 5: Remove period after Statistical Analysis (you don’t use a period in any of your other headers)

General comments:
1. When beginning a sentence with a number, spell it out (e.g. “8/98” starting the second sentence in the abstract results section, should be “Eight out of ninety-eight”)
2. Be consistent throughout the text about using radiochemotherapy (sometimes you used chemoradiotherapy). Pick one and use it throughout.

Minor Essential Revisions:

General Comments:
1. Take out anything about overall survival. I think your survival rate is too high at this time point and your numbers are too low to actually make any statements about this.
2. Throughout the document, make sure you are using PIK3CA (sometimes you have PI3KCA
3. Was DNA obtained from primary tumors for all cases? Make a statement clarifying this information in the methods section.
4. Use decimals in Table 1, not commas.
5. Add strengths paragraph or a few sentences to the discussion (generally immediately before or after your weaknesses). This paper is really lacking in power, so you are not going to be able to draw any strong conclusions about your findings. However, you need to make a convincing argument for the aspects of your study that do make it unique and/or worth publishing (e.g. although your
numbers are small, it consists of a much more homogeneous group than prior studies-at least that is what I have gathered from reading your paper). Provide references.

Specific Comments:

Page 1: Background: Replace the second sentence with the following sentence or something similar: “until recently, there have been limited predictive factors (clinical or biological) for rectal tumor response to conventional treatment.”

Page 1: Materials and Methods: Second Line: Replace “consecutive” with consecutively

Page 1: Materials and Methods: Third Line: Replace “operated” with “who received surgery”

Page 1: Results: Last sentence: PIK3CA and BRAF should be italicized and remove “and neither with overall survival”

Page 1: Conclusion: Last sentence: Remove (it implies you can rule out KRAS, BRAF, and PIK3CA mutations based on your study which lacks the power to draw any such conclusions) and replace with a statement about future studies with greater numbers, follow-up time, etc. with greater power to predict associations are necessary to fully understand this relationship.

Page 2: Line 4: Replace “in association with” with “in addition to” or “along with” or “combined with”

Page 2: Last line: Do you mean fewer recurrences? If so, replace “less” with “fewer”

Page 3: First sentence: Replace first sentence with “However, the decision to use neoadjuvant radiochemotherapy is complex.”

Page 3: 2nd to last sentence: Replace with: Currently, the best available methods to investigate improved outcomes in rectal cancer include accurate early assessment of tumor response with MRI and identifying predictive molecular tumor abnormalities. Add reference to support that these are the best current methods.

Page 3: 2nd paragraph: Lines 6-7: Replace “absence of anti-EFGR’s benefit in metastatic setting” with “poor response to EGFR in a metastatic setting”

Page 4: 1st paragraph: Line 5-7: This sentence doesn’t make sense as stated. You go on to provide a study that shows PIK3CA mutations do have prognostic value in rectal cancer specifically. I would leave it out and add it to the last sentence of the paragraph as a factor that highlights what makes your study unique from previous studies.

Page 4: 1st paragraph: last sentence: As mentioned in the last comment, the final sentence of this paragraph should describe what distinguishes your study from prior studies. Include all unique aspects (e.g. it is the first to look at all three mutations in locally advanced rectal (not colorectal) cancer in patients treated with neoadjuvant chemotherapy and surgery). My understanding is that you are
the first to look at this combination,

Page 4: 3rd paragraph, last sentence: Replace with” Prior to treatment, a history and physical exam were completed for all patients as well as assessment of performance status, complete blood counts (CBCs), liver function creatinine, and serum carcinoembryogenic antigen.”

Page 5: DNA (include space between these words) extraction and mutation analysis: Replace “PCR” with “DNA was”.
Provide references for “hot-spot” mutant selection.
Reword: “Mutation detection par sequencing.”
Replace K-RAS with KRAS and italicize

Page 5-6: Statistical methods are not consistent with what you reported in results/tables. Also, what software did you use to analyze?

Page 6: Results: Replace “Patient’s” with “Patient”
Replace “Nitty-eight” with “Ninety-eight”

Page 6: Pathologic characteristics: 1st line: Add comma after KRAS
2nd to last line: replace “in chemoradiation’s response” with “in response to chemoradiation”

Page 6: Mutations analysis: Replace “Mutations” with “Mutation”

Page 7: Survival: Replace “Survival” with “Correlation with local and metastatic recurrences”. It does not appear that you are actually doing a survival analysis (you are probably underpowered to do so). All you are showing is a correlation between the mutations and local and distant recurrences. Remove any mention of overall survival, unless you want to make a statement about how relationships with overall survival could not be calculated due to high survival rate/limited events (provide numbers).

Page 8: 1st paragraph: last sentence: replace “already described especially in the prior reports…” with “previously described in reports…”

Page 8: 2nd paragraph: 1st sentence: This sentence does not seems to be supported by the rest of the paragraph. Reword.
2nd sentence: Need references to support. Replace “seems” with “is”

Page 9: 1st Paragraph: Remove “In our study, no relation was found between KRAS, BRAF or PI3KCA mutations and local recurrence. Move to end of paragraph or leave out. You cover it in the last paragraph.

Page 9: 2nd paragraph: Replace “studies focused” with “studies have focused”.
Replace RAS with KRAS and italicize.
2nd paragraph: line 4: replace “as well as” with “and”
Lines 8-9: Reword second part of this sentence. It is also unclear which study you are referring to for the second part of the sentence-add reference.
Page 10: Last paragraph: 1st sentence: add an “or” in between BRAF and PIK3CA. Replace “for response to preoperative RT/CT or survival.” with “following preoperative RT/CT.
In general remove the word survival and use recurrence in this paragraph.
Last sentence: Replace “drawn” with “draw”

Major Compulsory Revisions:
General Comments:
The authors should provide strengths and weaknesses of the study design and offer future work that will help them address this question properly (perhaps reevaluating after a longer follow-up period).

Level of interest: An article of limited interest

Quality of written English: Needs some language corrections before being published

Statistical review: Yes, but I do not feel adequately qualified to assess the statistics.

Declaration of competing interests:
I declare that I have no competing interests