Reviewer’s report

Title: Membrane-bound exosomal HSP70 as a biomarker for detection and monitoring of malignant solid tumours: a pilot study

Version: 0 Date: 11 Nov 2019

Reviewer: Jun Thorsteinsdottir

Reviewer's report:

Comments to the author:

The authors present a study design for evaluating membrane-bound exosomal HSP70 as a biomarker for detecting and monitoring malignant solid tumours. The study background is well done and overall aim is of interest. However, the study design could be further strengthened by additional information given in "Comments to the Author". General and specific comments are enumerated below:

In general, pages are missing within the manuscript.

Background section:

- In the sentence "It is well known that development of efficient diagnosis methods is crucial to reduce the cancer morbidity", it should be considered if the sentence should be changed in ".. to reduce cancer-related mortality" (page 4, line 20-22).

- The word "contrarily" should be changed into "in contrast to" (page 5, line 3).

- The word "Contrary" should be changed into "In contrast to.." (page 5, line 26).

- The wording of the sentence "We have demonstrated that an important pathway through which HSP70 can be secreted is via exosomes." should be reevaluated (page 5, line 25-27).
Methods section:

- The authors state that "30 women with breast cancer, 10 women with ovarian cancer, 10 men and 10 women with non-small cell lung cancer will be included" (page 6, line 25-27) and "additionally 20 healthy volunteers" (page 6, line 25-27); The total number of included patients is too low to receive significant results, especially when different tumour entities are compared. It should be considered if a matched-pair analysis would be more precise.

- The authors state that secondary objectives include that "determine whether the concentration of HSP70-exosomes varies with the nature of the primary tumour or the effectiveness of the treatment used" (page 6, line 57-59); The total number of included patients is too low to receive significant results, especially when treatment modalities are compared.

- Furthermore, the authors plan to analyze "the concentration of blood HSP70-exosomes in patients having a complete response, partial response, stable disease or progressive disease (RECIST criteria)"; the subgroups will be too small to receive any significant results. (page 7, line 13-17)

- The word "ancillary" is not very common (page 7, line 24 and in the following text).

- The time schedule to collect blood samples after the first visit after surgery should be specified, e.g. within 12 hours after surgery (page 8, line 16-18).

- The time schedule to collect blood samples should be the same in the patient (page 8, line 21-23) and healthy subject group (page 8, line 44-46), e.g. three months.

- Do healthy subjects receive a cancer screening before they are included into the study?

- In the section "sample size and statistical analysis" (page 9, line 34-46) the authors state that breast cancer patients should be classified into HER2-positive and -negative groups and with or without metastatic evolution. The sample size n=5 for the subgroups are too small.

- On page 9, line 46-48, the authors declare that "No formal sample size calculation was performed regarding the exploratory design of the study"; a sample size calculation by a statistician should be definitely performed.
Discussion section:

- What is meant by the sentence "Current research focuses on liquid biopsies that enable non-invasive sampling of tumours"? Do the authors mean "non-invasive detection of tumours"? (page 10, line 39-41).

- The word "contrarily" should be changed into "in contrast to" (page 10, line 50).

Level of interest
Please indicate how interesting you found the manuscript:

An article of importance in its field

Quality of written English
Please indicate the quality of language in the manuscript:

Needs some language corrections before being published

Declaration of competing interests
Please complete a declaration of competing interests, considering the following questions:

1. Have you in the past five years received reimbursements, fees, funding, or salary from an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

2. Do you hold any stocks or shares in an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

3. Do you hold or are you currently applying for any patents relating to the content of the manuscript?

4. Have you received reimbursements, fees, funding, or salary from an organization that holds or has applied for patents relating to the content of the manuscript?

5. Do you have any other financial competing interests?

6. Do you have any non-financial competing interests in relation to this paper?

If you can answer no to all of the above, write 'I declare that I have no competing interests' below. If your reply is yes to any, please give details below.

I declare that I have no competing interests.
I agree to the open peer review policy of the journal. I understand that my name will be included on my report to the authors and, if the manuscript is accepted for publication, my named report including any attachments I upload will be posted on the website along with the authors' responses. I agree for my report to be made available under an Open Access Creative Commons CC-BY license (http://creativecommons.org/licenses/by/4.0/). I understand that any comments which I do not wish to be included in my named report can be included as confidential comments to the editors, which will not be published.

I agree to the open peer review policy of the journal

Were you mentored through this peer review?

No