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Role of cancer-associated fibroblasts in oral squamous cell carcinomas, surgical margins, and verrucous carcinomas: An immunohistochemical study

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Handling editor:
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1st Editorial decision
02-Nov-2021

Ref.: Ms. No. JCTRes-D-21-00149
Role of Cancer-Associated Fibroblasts in Oral Squamous Cell Carcinomas, Surgical Margins, and Verrucous Carcinomas: An Immunohistochemical Study.
Journal of Clinical and Translational Research

Dear Dr. Datar,

Reviewers have now commented on your paper. You will see that they are advising that you revise your manuscript. If you are prepared to undertake the work required, I would be pleased to reconsider my decision.

For your guidance, reviewers' comments are appended below.

If you decide to revise the work, please submit a list of changes or a rebuttal against each point which is being raised when you submit the revised manuscript. Also, please ensure that the track changes function is switched on when implementing the revisions. This enables the reviewers to rapidly verify all changes made.

Your revision is due by Dec 02, 2021.

To submit a revision, go to https://www.editorialmanager.com/jctres/ and log in as an Author. You will see a menu item call Submission Needing Revision. You will find your submission record there.

Yours sincerely

Michal Heger
Editor-in-Chief
Journal of Clinical and Translational Research

Reviewers' comments:

Reviewer #1: Is it necessary to mention the code of ethics?
Using a marker in this evaluation is not enough
No new findings
The exact location of the surgical margin is not stated in the materials and methods
The type of biopsy performed (exact inclusion criteria) is not stated
Verrocus carcinoma does not have dysplasia. The introduction and purpose of comparing it with SCC are not specified.

EDITOR:

Dear authors, thank you for submitting your work to JCTR. Although the reviewer who appraised your paper recommended a reject verdict based on the comments above, the editorial board has decided to give you a chance to address the comments and revise your manuscript accordingly. In doing so, it is imperative that you do so in a point-by-point manner and elaborately. A substantial effort should be put into rebutting the concerns and incorporating the necessary changes into the manuscript where warranted.

Our experts in dentistry and myself would like you to incorporate additional representative histological images in Figures 1-3. There should be at least 1 representative panel per score (0-4) for OSCC, VC, and NMOSCC with appropriate anatomical annotation (e.g., arrows pointing to CAFs, red arrowheads pointing to tumor, blue arrowheads pointing to healthy tissue).

Thank you,

Michal Heger
Editor

Authors’ response

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Re: Revision Ms. No. JCTRes-D-21-00149

Dear, Michal Heger,

Thank you for reviewing our manuscript 'Role of Cancer-Associated Fibroblasts in Oral Squamous Cell Carcinomas, Surgical Margins, and Verrucous Carcinomas: An Immunohistochemical Study.' and allowing us to better it.

We are grateful for this opportunity and we have addressed all comments of the reviewers using the track changes function in Word. The constructive criticism has definitely refined the outcome of the manuscript. We have tried our best to incorporate all the suggested changes. We have included point to point response to the review's suggestion and the modifications made in the manuscript are highlighted with the red-colored font.

Reviewer #1:

Comment 1: Is it necessary to mention the code of ethics?

We have omitted the mention of the code of ethics in the revised manuscript.

Comment 2: Using a marker in this evaluation is not enough

We understand the limitation of the study and the concern expressed by the reviewers. However, αSMA is expressed during the differentiation of fibroblasts to CAFs so its use as only marker for identification of CAFs is widely accepted. (Page 2 Line 7)

We agree that the use of additional markers would enable better characterization of CAFs in the lesions being discussed.

Comment 3: No new findings

Although literature exists regarding myofibroblasts in precancer and cancer, our current understating of their role as cancer-associated fibroblasts has evolved over the past decade. As highlighted, the CAFs play a role in stoma modulation and metastasis hence in determining the prognosis.

Since α SMA is a well-recognized marker for the identification of CAFs we emphasize its definitive use as a screening marking for surgical margins, OSCCs, and VCs.

Comment 4: The exact location of the surgical margin is not stated in the materials and method.

Thank you for this suggestion we have made the necessary changes.

In the patient-matched OSCC and HNMOSCC tissue 26 were males and 4 females in the age range of 35-90; buccal mucosa was the predominant site (12) followed by the tongue (7), three each of alveolar mucosa, retromolar area, and gingivobuccal sulcus and two of the palate. VC had 16 males and 14 females of the age range 35-80 years. VCs were predominantly located on the buccal mucosa (18) followed by labial mucosa (4), two each of Buccal vestibule and gingivobuccal sulcus; and one each of retromolar area, tongue and alveolar mucosa and palate.

Comment 5: The type of biopsy performed (exact inclusion criteria) is not stated

We are yet again thankful for this suggestion and we were able to include the changes of both this and the earlier suggestion in methodology.

The patient-matched OSCC lesional tissue and HNMOSCC were obtained from the surgical excision and radical neck dissection specimens available in the archives. The HNMOSCC included were taken one cm beyond the surgical margin of OSCC; they were histologically tumor-free and the epithelium was devoid of dysplasia.

Comment 6: Verrucous carcinoma does not have dysplasia. The introduction and purpose of comparing it with SCC are not specified.

Although verrucous carcinoma is almost devoid of dysplasia and does not show a breach in the basement membrane; about 20% of these tumors harbor elements of conventional OSCC and are prone for locoregional recurrence hence have an overall have a guarded prognosis. (Mentioned in Introduction Paragraph 2 First line)

Considering the possible aggressive behavior and potential for invasion into the surrounding tissues a stringent molecular screening of these lesions is deemed essential.

Considering the known presence of CAFs in OSCC they were compared VCs.

Editor's comments:

Our experts in dentistry and myself would like you to incorporate additional representative histological images in Figures 1-3. There should be at least 1 representative panel per score (0-4) for OSCC, VC, and NMOSCC with appropriate anatomical annotation (e.g., arrows pointing to CAFs, red arrowheads pointing to the tumor, blue arrowheads pointing to healthy tissue).

We greatly appreciate this suggestion and have added the representative images of all the scores noted in the three study groups with anatomical annotations.

In addition to the above changes, we noticed that HMNOSCC and NMOSCC were used to depict the same entity, it was rectified to HMNOSCC.

2nd Editorial decision
03-Dec-2021

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Michal Heger
Editor

Authors' response

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Re: Revision Ms. No. JCTRes-D-21-00149

Dear, Michal Heger,

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1. **Comment 1:** Please go through the language again and improve to academic level English. Especially the results section is poorly written. We urge you to employ a native speaker or a proofreading service. As a final option, JCTR has in-house editorial staff that could perform a deep dive edit for a fee.
   Response: Thank you for your valuable suggestion, we have edited the manuscript and checked for grammatical errors and paraphrased the manuscript to improve the language to academic level English. I hope it meets your expectations.
2. **Comment 2**: Please indicate in the methods section that the study was exempt from institutional review board approval due to its retrospective and anonymized nature.
   Response: Thank you, we have made the necessary change in the methodology section.
   The study was exempt from the institutional ethical committee review because of its retrospective nature. (Page 2 Paragraph 3 Line 2-3)
3. **Comment 3:** Please explicitly address the limitations of the study in the Discussion.
   Response: We made the recommended modification in discussion as follows
   However, being a preliminary research, the present study is limited by the lack of follow up data. (Page 6 Discussion Paragraph 2 Line 2-3)
   We have also discussed this in discussion as ……
   Further inclusion of histological parameters like tumor thickness, depth of invasion, and neurovascular invasion are needed to elaborate on the role and interactions of CAFs with the tumor microenvironment. (Page 6 Discussion Paragraph 2 Line2 -9)
4. **Comment 4:**Explicitly indicate what the novelty is of the current study relative to previously published studies on this subject matter in the Discussion

Response: We are thankful for the insightful suggestion and have made the addition in the discussion and conclusion.

But in the present study we have proposed the possible hypothesis for transformation of CAFs even in lesions with intact basement membrane. Moreover we also hypothesize that lesions with more CAFs may have poor prognosis. (Page 5 Paragraph 3 Line 14-15)

The present report distinctly discusses the role of CAF screening in OSCCs and VCs for predicting patient prognosis and highlights the need for the development of a CAF-based targeted therapy. (Page 6 Conclusion Line 6-9)

We are grateful for the suggestion and are looking forward to your response.

Thank you,

Uma Datar

3rd Editorial decision
08-Jan-2022

Ref.: Ms. No. JCTRes-D-21-00149R2
Role of Cancer-Associated Fibroblasts in Oral Squamous Cell Carcinomas, Surgical Margins, and Verrucous Carcinomas: An Immunohistochemical Study.
Journal of Clinical and Translational Research

Dear authors,

I am pleased to inform you that your manuscript has been accepted for publication in the Journal of Clinical and Translational Research.

You will receive the proofs of your article shortly, which we kindly ask you to thoroughly review for any errors.

Thank you for submitting your work to JCTR.

Kindest regards,

Michal Heger
Editor-in-Chief
Journal of Clinical and Translational Research

Comments from the editors and reviewers: