Author's response to reviews

Title: Induction of chromosome instability and stomach cancer by altering the expression pattern of mitotic checkpoint genes in mice exposed to betel-nut

Authors:

Sillarine Kurkalang (sillarine.jk@gmail.com)
Atanu Banerjee (atanu_7777@yahoo.co.in)
Nitin Ghoshal (ngbiotech@gmail.com)
Hughbert Dkhar (hdkpath@rediffmail.com)
Anupam Chatterjee (chatterjeeanupam@hotmail.com)

Version: 2 Date: 21 May 2013

Author's response to reviews:

Response to Reviewer
Title: Induction of chromosome instability and stomach cancer by altering the expression pattern of mitotic checkpoint genes in mice exposed to areca-nut.

Author: Sillarine Kurkalang, Atanu Banerjee

Response to Reviewer 1 (29th April, 2013)
We thank the referees for constructive comments on our manuscript. It has been revised to take into account all the suggestions made by the referees.

1. We have replaced betel-nut with areca-nut.
2. Necessary correction has been made.
3. The soft areca-nut was cut into pieces and then made powder with a mechanical grinder.
4. We have submitted a flow-diagram on experimental design and incorporated in the revised manuscript.
5. We apologize for this mistake. It will be 40 µg instead of 40 mg.
6. The results of RBN+lime in mouse bearing advanced tumor were described in the results section. The mean frequency of aneuploid cells was mentioned.
7. We have incorporated expt number/animal number in figure legend of figure 2. However, for the figure 3 and 4, such information was already there.
8. The description on Anisokaryosis is very much there in the legend of Figure 2.
9-10. It is true that areca-nut and lime induced cellular changes must be triggered through upstream signaling molecule like- ATM and check point kinases. However, in the present study, we did not conduct any experiment to know the status of these genes. Nevertheless, we did analyze expression profile of several
genes whose initial involvement in carcinogenesis is evident. Besides these genes, indications are there for the early involvement of p16 and Rb in such tumor development. Although, it is clear that the cell cycle check-point genes' deregulation could be an early step in areca-nut mediated carcinogenesis, however, some more studies are needed before proposing a pathway in the development of areca-nut induced carcinogenesis. Therefore, we are not incorporating such diagram here showing relationship of these genes in the process of carcinogenesis.

Response to Reviewer 2 (13th May, 2013)
1. All the minor points are considered here.
2. In fact exchange aberrations are not induced by areca-nut extract exposure. It was mentioned in the Supplemental Information section, however, now we have incorporated one line in the text.
3. Normal means untreated mice. We have modified accordingly.
4. page 9; line 2 from the bottom- an appropriate modification incorporated to erase confusion.
5. PARP is absent in all the cells of tumor. Yes, it indicates that tumor originate from a single cell. Some more studies are needed to address this issue.
6. We are sorry for not able to reduce the size of Introduction and Discussion sections of the manuscript.