



Interactive comment on “Experimental fossilisation of viruses from extremophilic Archaea” by F. Orange et al.

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Received and published: 11 May 2011

Thank you very much for your comments on our manuscript.

The sentence mentioned by the reviewer (p. 9, l. 11–15) was not correctly phrased, which may have lead to confusion. This will be corrected in the revised version. Fig. 2a and 2b are not control samples. Observations of the silica precipitate formed in the control experiment without viruses were made but were not included in the first version of the article. This precipitate had a similar structure to that formed in the SIRV2 and TPV1 experiment. They will be added to the revised version.

We based our identification of filaments as DNA on observations made previously on SIRV2 by the authors, or on similar virus in other studies. SIRV2 particles have an

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helical body made of a filament of a protein and DNA. The filament observed is the result of the fragmentation of the particle, as was observed by Prangishvili et al. (1999). Vestergaard et al. (2008) observed similar filaments between fragments of the *Stygiolobus* Rod-Shaped Virus (SRV), and identified these structures as 'DNA or DNA-protein fibers lacking the protein core' (Vestergaard et al., 2008 ; Fig. 2). We ruled out the possibility that these filaments may be artefacts of sample preparation. The sample preparation for electron microscopy led to the formation of artefacts, due to the drying and the staining. But similar filaments were not observed in the TPV1 and PAV1 experiments or on samples without viruses, while they were almost ubiquitously observed in fossilised SIRV2 samples, always connecting SIRV2 fragments. Thus, these filaments are definitely the results of the degradation of the virus, although they could include viral protein as well as DNA. This will be corrected in the revised version.

The figures captions will be modified for more clarity.

Mention of the chelating and binding properties of EDTA will be added when explaining the role of the medium in the particular aspect of the silica precipitate in the PAV1 experiment. EDTA is likely to have bound silica through its carboxylic functional group, and thus to have modified the aspect of the silica precipitate, although this has not been tested. The presence of salts and the difference in the ionic strengths of the media apparently did not play a role on the aspect of the silica precipitate that formed, as the silica precipitate had a similar aspect in both the TPV1 and SIRV2 experiments (in media with and without salts, respectively). It may however have influenced the rate of silica polymerisation, although this was not tested in our experiment.

The direct nucleation of silica on the outer surface of viruses was observed after one month, and led to the formation of hemispherical silica particles bound to SIRV2 particles (Fig. 2C). The increase of the size of the particles observed after two months (Fig. 2E) indicates a growth of these particles by continuous polymerisation of silica, while the particles retain a hemispherical shape. Particles that formed spontaneously in the medium and were bound afterwards to the virus particle surface do not have this

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shape (as for example the middle silica particle among the three attached to the virus in Fig. 2E, which has similar aspect to the free particles present around the virus). These details will be added to the revised version of the manuscript.

References :

Prangishvili, D., Arnold, H. P., Götz, D., Ziese, U., Holz, I., Kristjansson, J. K., and Zillig, W.: A novel virus family, the Rudiviridae: structure, virus-host like interactions and genome variability of the *Sulfolobus* viruses SIRV1 and SIRV2, *Genetics*, 152, 1387–1396, 1999.

Vestergaard, G., Shah, S. A., Bize, A., Reitberger, W., Reuteur, M., Phan, H., Brigel, A., Rachel, R., Garrett, R. A., and Prangishvili, D.: *Stygiolobus* Rod-Shaped Virus and the interplay of Crenarchaeal Rudiviruses with the CRISPR Antiviral System, *J. Bacteriol.*, 190, 6837–6845, 2008.

Interactive comment on *Biogeosciences Discuss.*, 8, 2235, 2011.

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