Discovery of Uranium in Outer Coat of Sri Lankan Red Rain Cells

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Summary

The microbial content of the red rain that fell over central Sri Lanka in November/December 2012 shows generic similarities to that of the Kerala red rain. Light microscope examination of the Sri Lankan red rain indicates that the defining red rain cells exist in the presence of other microorganisms including diatoms. We report the results of a preliminary TEM study of the red rain cells that shows them to have outer cell walls unusually rich in uranium, and a nuclear region with a strong deficit or absence of phosphorus.

Keywords: Red rain, comets, panspermia

Introduction

Reports of red rain – rain of "blood" from the sky – have an antiquity that stretches back to biblical times (McCaffarty, 2001). Such events were often perceived as having a supernatural provenance, and very much feared in the ancient world. After naturalistic explanations of such phenomena began to be sought in the seventeenth and eighteen centuries, the causative agent of

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the red colour was variously considered to be either fine dust or algae lofted from the surface. In some instances red dust is an explanation of the phenomenon, but a significant number of instances defy any such a simple explanation.

Kerala Red Rain

The most publicized red rain event of recent times is one that took place in Kerala, India during May to August 2001. The first red rain fall in Kerala was preceded by a sonic boom in the sky, presumably caused by an exploding In this event over 50,000 tones of red particulate cometary meteoroid. material fell over an area of a few thousand square kilometers. Samples of the red rain of Kerala were first collected by Godfrey Louis and analysed using a variety of laboratory techniques. It was clearly shown that the red colour arose from red coloured living cells (Louis and Kumar, 2003, 2006). However, to this day the identity of the cells and their origin remains obscure. Sampath et al. (2001) claimed that the red rain cells were spores of algae belonging to the Trentepohlia genus but this has not been borne out in later investigations by Gangappa (2011) and Kumar, Wickramasinghe and Louis (2013). Although it is conceivable that such algal spores were present as relatively minor contaminants we dispute that they were the bulk of the red rain cells (Miyake, 2011).

The Kerala red rain cells (RRCs) remain to this day unidentified and it is not even clear whether they are eukaryotes (cells with nuclei) or prokaryotes (Gangappa, 2011; Miyake, 2011). Louis and Kumar (2003, 2006) have controversially maintained that the red cells could replicate at temperatures as high as 450C under high pressure, and furthermore that no DNA could be unequivocally identified. Although these claims are still open to challenge, the case is rapidly growing for the involvement of an unknown or alien microorganism of presumed extraterrestrial origin. Gangappa *et al* (2010) have verified the replication of red rain cells at 121°C under pressure in an autoclave, thus making the red rain cells perhaps the most thermophillic living cells yet known.

Sri Lankan Red Rain – Light microscope studies

Several episodes of red rain were reported in the Central and Southern Provinces of Sri Lanka during late November and December 2012. These events were preceded by fireball sightings and a meteor fall that happened approximately 10 days earlier. Samples were collected under the direction of Dr. Anil Samaranayake, Director, MRI Sri Lanka, and made available to NCW for the purpose of the present research.

Under the light microscope, Sri Lankan RRCs appear generally similar to Indian RRCs but are mixed with more microbial contaminants than were present in the latter (Figs.1, 2). The contaminants were almost certainly acquired near ground level and may have included small amounts of terrestrial bacteria. The RRCs were however clearly separable in TEM analysis of centrifuged samples of the rain.

Both the Sri Lankan and Indian RRC's have a reddish-brown colour and oval shapes with sizes in the range $2\sim10\mu m$. They are clearly distinguishable from light microscope images of *Trentopohlia* since RRC's red colour is visible throughout the entirety of the cell including the outer wall, and it is not caused by localized carotenoids (Kumar et al, 2013).

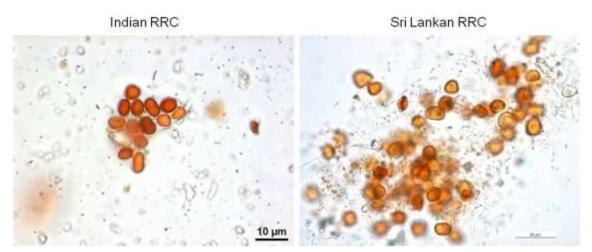


Figure 1: Indian and Sri Lankan Red Rain Cells (RRCs) under light microscopy. [left] Indian RRCs. They have a reddish-brown colour and oval shapes with sizes in the range 2~10μm. [right] Sri Lankan RRCs. Their morphology is similar to Indian RRCs. bar – 20μm.

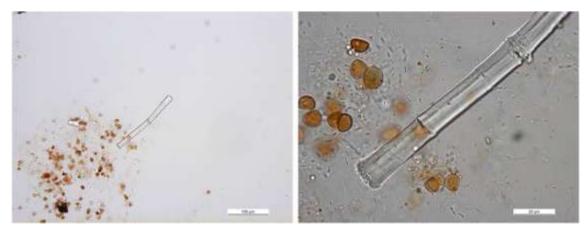


Figure 2: Light microscopic image of Sri Lankan Red Rain sample. [left] Image of a diatom with Sri Lankan RRCs. bar $-100\mu m$. [right] Magnified image showing the smaller bacterial cells surrounding RRC and diatom. bar $-20\mu m$.

Transmission Electron Microscopy

The Sri Lankan red rain samples were concentrated in Eppendorf tubes by centrifuging at 14,800g/2min. They were then fixed directly in suspension for 5 days by adding 25% v/v stock glutaraldehyde to give a final concentration of 2.5% v/v. The suspensions were concentrated by centrifuging at 15,000g for 2min at room temperature and the pellets re-suspended in molten 4% w/v agarose. When the agarose solidified, it was cut into small pieces (approx. < 1mm³) and placed in double distilled water for TEM processing. This was carried out as explained in Newman and Hobot (2001).

Ultrathin sections were cut on a LKB III Ultramicrotome and they were mounted on nickel grids. We prepared two samples – one with negatively stained using Uranyl acetate and another without *any* staining whatsoever. The sample with negative staining was used for obtaining the best contrast from electron microscopy, whereas the unstained sample is for elemental analysis.

The sections were examined using a Philips/FEI CM12 TEM (Transmission Electron Microscope) operating at 80kV. Images were recorded using a SIS MegaView III digital camera. The elemental compositions were obtained using an Oxford Instruments INCA ENERGY (EDX) x-ray analysis system

incorporated with TEM.

The cross-section of RRCs revealed unusually thick outer walls similar to the Kerala red rain cells (Fig.3A). Cells are seen to possess multiple membrane layers, and an inner protoplast that seems poorly preserved. The degradation of outer wall coating appears to have started already, and some cells were seen to have lost it completely. There are some cells "captured" in a process of cell division, probably by binary fission, into two daughter cells within an outer cell wall (Fig.3B).

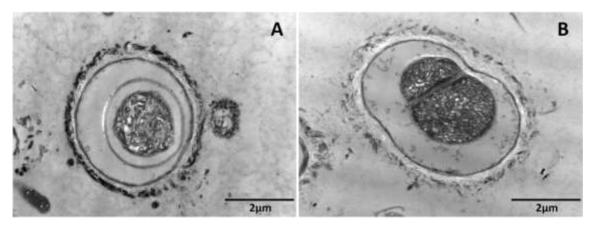


Figure 3: TEM (Transmission Electron Microscope) image showing the ultrastructure of negatively stained RRCs. [A] Cross-section of RRC with a thick outer wall and multiple layers of cellular membrane. There are huge gaps between the wall and the cellular membranes. [B] This image shows the cell multiplying into two daughter cells, probably by a binary fission, within the outer wall.

Excluding C, N, which shows high peaks at the lowest energies, the distribution of atomic nuclei in the outer coatings of the cell and the entire cell interior show distinct differences. Most striking is the high abundance of uranium in the outer cell wall and the lack of a phosphorus peak in the cell interior (Fig.4). Note that the elemental composition was analyzed without using any negative staining. This has been proved by not detecting uranium from the cell interior. (If the sample was UA stained, uranium should be detected from entire cell and also from the control.) We are therefore compelled to conclude that the Sri Lankan RRC's have a high concentration of uranium, that is not characteristic of a normal terrestrial cell. We note from the lower frames of Fig.4 that there is a conspicuous absence of mass peaks corresponding to the element P. The lack of phosphorous also points to an unusual organism, and if confirmed supports the contention of Louis and Kumar for a lack (or dearth) of DNA. All this can be taken as arguable evidence of a meteorite/cometary origin of the cells.

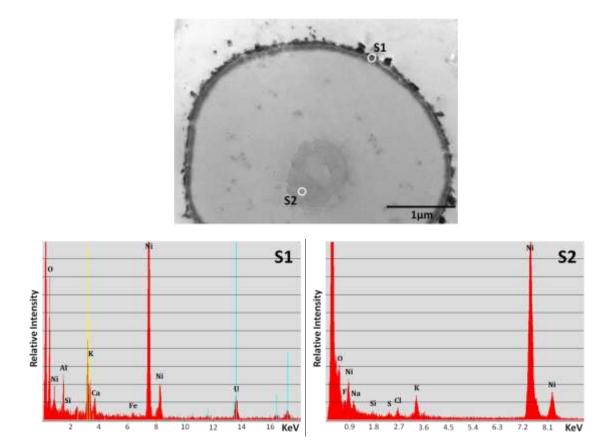


Figure 4: [Above] TEM image showing the ultrastructure of non-stained RRCs. Elemental composition of the outer wall was taken at S1 and inside the cell was taken at S2. Note that Nickel peaks are shown due to the Ni grid used. [S1] Elemental composition of the outer wall containing Aluminum and Uranium, and some Calcium, Silicon, Iron and Potassium. (blue line – primary peaks of U, yellow line – secondary peaks of U). [S2] Elemental composition of the protoplast containing Sodium, Chlorine and Potassium, and some Fluorine and Sulphur.

In the latter context it is of interest to note that structures consistent with RRC's were discovered in SEM (Scanning Electron Microscope) studies of the Polonnaruwa meteorite as shown in the left hand frame of Fig. 5

(Wickramasinghe, et al, 2013). The right hand frame is an SEM of Kerala RRC's.

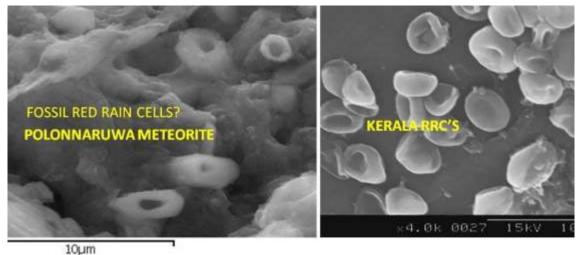


Figure 5: [left] SEM image of an interior section of the Polonnaruwa meteorite. [right] SEM of a sample of Kerala Red Rain cells.

Although these preliminary results do not prove that the red rain cells are of extraterrestrial origin, they appear to be strongly supportive of such a hypothesis. Alternatively, it has to be concluded that they represent an hitherto unrecognized form of terrestrial life.

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References

Gangappa, R., 2011. PhD dissertation, University of Glamorgan

Gangappa, R., Wickramasinghe, C., Wainwright, M., Kumar, S., Louis G., 2010. Growth and replication of red rain cells at 121°C and their red fluorescence. arXiv:1008.4960 [astro-ph.CO].

Louis, G. and Kumar, S., 2006. New red rain phenomenon of Kerala and its possible Extraterretrial Origin, *Astrophys. Sp. Sci.*, 302:175-187.

Louis, G, and Kumar, S., 2003. New biology of red rain extremophiles prove cometary panspermia, http://arxiv.org astrophysics e-print archive.

Louis, G. and Kumar, S., 2006. New red rain phenomenon of Kerala and its possible Extraterrestrial Origin, *Astrophys. Sp. Sci.*, 302:175-187.

McCafferty, P., 2008. Bloody rain again! Red rain and meteors in history and myth, *Int. J. Astrobiol.*, 7(1), 9-15. doi:10.1017/S1473550407003904.

Miyake, N., 2011. PhD dissertation, Cardiff University

Newman, G.R., and Hobot, J.A., 2001. Resin Microscopy and On-Section Immunocytochemistry, 2nd ed. Heidelberg: Springer.

Sampath, S., Abraham, T.K., Sasi, K.V., and Mohanan, C.N., 2001. Coloured Rain: A Report on the Phenomenon, *CESS-PR-114*

Wickramasinghe, N.C., Wallis, J., Wallis, D.H., Samaranayake, A., 2013a. Fossil diatoms in a new carbonaceous meteorite, *Journal of Cosmology*, **21**, 37