RESEARCH

Biology Associated With A Titanium Sphere Isolated From The Stratosphere

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A sphere of diameter 30 microns was isolated from the stratosphere at a height of between 22–27 kilometres. It was found to be mainly composed of titanium (with smaller amounts of vanadium). Nanomanipulation and EDX analysis showed that the titanium sphere contains a carbonaceous non-granular material which we suggest is a biological protoplast. Damage to the surface of the sphere revealed a carbonaceous, filamentous material, having a "knitted" appearance", which we also suggest is biological in nature. The titanium sphere produced a distinct impact crater when it impacted the carbon sampling stub. We conclude by suggesting that this largely titanium sphere contains biological elements which impacted the sampling stub at speed as it made the journey from space to the stratosphere.

Introduction

In our earlier papers we suggested that the biological entities (ranging in size from 10-300 micron) which we have isolated from the stratosphere originate from space, rather than Earth (Wainwright et al., 2013a, b). We base this conclusion on the current paradigm which states that particles greater than 5 microns radius cannot cross the tropopause to arrive at the heights in the stratosphere from where we have sampled them (Rosen, 1969, Zolensky & Mackinnon, 1985). Also, the sampling stubs on which we isolated these biomorphs were found to be remarkably free of the contaminating material (such as fungal spores, pollen grains and volcanic dust) which we would expect to find had these biological entities been elevated from Earth to the stratosphere. Furthermore, marked impact events caused by inorganic micrometeorites, together with a wide variety of cosmic dust particles, also occur on the same sampling stubs as do some of the biomorphs; while it could be argued that the biomorphs reached the stubs post-impact, we nevertheless suggest that this association remains highly noteworthy. We ask the question - if a mechanism exists which can elevate the biological entities we find, from Earth to the stratosphere, how is it able to "sieve out" only the biomorphs (which are of varying sizes and masses) from the general debris that would be carried to the stratosphere were these particles elevated from Earth to the stratosphere? It could be argued that the biomorphs in question originate

* Department of Molecular Biology and Biotechnology, University of Sheffield, U. K. from a terestrial aquatic environment, but if so, we would again expect them to be associated with other marine, or freshwater organisms and debris; we certainly would not expect a marine water spout to be elevated to a height of 22–27km, and carry with it biomorphs. We contend however, that the biomorphs isolated here formerly existed in a watery environment, namely a comet, the icy debris of which would have been largely lost during its transport to the stratosphere.

While we are convinced that the our earlier findings (Wainwright et al., 2013a, b) strongly suggest that our stratsopheric biomorphs originate from space, we continue to seek further evidence to strengthen our argument. Here we provide findings based on the use of nanomanipulation of a spherical particle which we have isolated on one of our sampling stubs. We provide evidence to strongly suggest that that this entity is biological in nature and, by using nanomanipulation, show that this biomorph, having produced a marked impact crater in the sampling stub, must have been travelling at speed (from space), when it impacted the stub. The results of this study, we assert, prove that a biological entity, originating from space, has been captured in the stratosphere en route to Earth. The implication of this finding is, we suggest obvious and profound, namely that this, and other biological entities, are continuously raining down to Earth from space.

Materials and Methods

A balloon-launched sampling device was released from Chester, NW England on 31 July 2013. The sampler included a drawer mechanism that could be opened and closed at a predetermined height. The stratosphere sampler carried a video camera by which the opening and closing of the sampling drawer could be viewed, confirmed and recorded. The sampling apparatus was

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protected from downfall of contaminating particulate matter from the balloon by a cover. Prior to launch, the inside of the draw device was scrupulously cleaned, air blasted and finally swabbed with alcohol. New scanning electron microscope stubs were placed in rows inside the drawer with their top surfaces facing outwards so that when the drawer was opened any particulate matter in the stratosphere would attach to them, and they could later be removed for examination under the scanning electron microscope. The protective layer on the surface of the stub was peeled off just before launching under a cover to prevent any particulate contamination. After sampling, the apparatus was transported to the laboratory and opened under conditions which avoided exposure of the stubs to contaminating dust and the stubs were similarly transferred under cover to the scanning E/M. The stubs were then sputter-coated with gold for 30 secs at 30 mA and then examined using a SEM (JEOL 6500F).

Balloon launch: The balloon was launched from an open field near Dunham on the Hill (near Ellesmere Port, Cheshire, England) during daylight hours and traversed to just south of Wakefield in West Yorkshire (England). The sampling drawer was opened for 17 minutes as the balloon rose from 22026m to 27008m. The sampling apparatus was returned to Earth (by parachute) undamaged and completely intact.

Control flight: A separate control flight was made to the stratosphere prior to the sampling flight, when the drawer was not opened, but all other sampling procedures were observed. No particulate matter was found (using the SEM) on any of the unexposed microscope stubs. This shows that the drawer remained airtight and that none of the stubs was exposed to particles at, or near, groundlevel or at any height up to the stratosphere. These results also show that no particles contaminated the stubs during any of the sample processing procedures, thereby demonstrating that the scrupulous procedures used to prevent ground level contamination proved effective and that no such contamination occurred.

Nanomanipulation

The adhesive carbon tabs on which stratospheric particles had landed during sampling were introduced into a modified Jeol 6500 SEM set-up complete with two nano-manipulation probes. Nickel-chromium nano-tips were prepared and introduced to the terminus of probes in order to directly manipulate the particle on the stub surface. Probes were hand controlled by an operator in order to both derive a visual understanding of the structural resilience of objects and to reveal previously hidden details by moving objects. Dynamic manipulations were video documented in real time and scanning electron micrographs taken to image finer detail. Although probe load data are not directly quantifiable, object resilience can be gauged through visual assessment of probe flexure when attempting to move the objects.

The stubs used (adhesive carbon tabs, sometimes referred to as Leit tabs) are made by Agar Scientific. In the sampling tray they were arranged essentially in a circle of around 10–11 cm diameter with the conductive sampling side facing outwards, with protective films removed shortly before launch in a controlled environment. The stubs were then transferred to 32x10mm sterile aluminium stubs for SEM analysis.

Results and Discussion

Figure 1A shows two spherical objects which we have isolated on sampling stubs exposed to the stratosphere at heights of between 22-27km; the main, or large spherical object (LSO) having on its surface a smaller, spherical object (SSO). Non biological particles which look like cosmic dust are also associated with the LSO, notably an amorphous inorganic mass located at the top of the large particle (Fig. 1). The central surface of the LSO is shown by EDX analysis to be mainly made up of titanium, with smaller amounts of vanadium, carbon and nitrogen (Fig. **1B**). The surface of the LSO shows distinct fault lines (Y) presumably caused by impact. In Fig. 2A, these appear as hexagonal plates, while Fig. 2B shows a long fault-line which suggests that hexagonal stress-fracture plates are formed towards its centre. Note also a number of small features at the bottom of Figure 3B (marked with an X) which may be biological in nature.

On the top right-hand surface of the LSO (Fig. 1) can be seen smaller spheres which are about 1micron in diameter and which appear to be bacteria (shown in detail in Fig. 3A). Two of these spheres show depressions which are presumably caused by the low pressure of the stratosphere or by SEM visualisation, showing that they are not solid mineral particles; no further evidence is however, available to confirm that they are bacteria. Bacteria have indeed been isolated from the stratosphere by a number of workers (Wainwright et al., 2006); if these spheres are bacteria then they may have attached to the LSO at its origination, or they may have adhered to the LSO as it passed through the upper stratosphere. The presumptive biological features associated with the surface of the LSO, while of interest, are not germane to our argument that this particle originated from space. The evidence in support of this claim is shown in the remaining Figures. Figure 4 for example, shows the LSO after it has been moved across the surface of the graphite stub by the nanosampler (which appears, in a damaged form, in the lower foreground of the image (labelled X). The contents of the LSO are clearly seen being dragged out of the main body of the particle, while the end of the inner matrix stream is attached to an impact crater.

A close-up of the LSO after it has been dragged across the surface of the stub is shown in **Fig. 5A**. It clearly shows that the SSO is attached to the LSO, but the larger sphere has rotated during nanomanipulation and the SSO seen in Fig. 1 is now seen at the rear of the larger particle. The contents of the LSO are clearly seen issuing from what appears to be a tear in its mid-side. **Figure 5B** shows that the bottom part of the LSO is made up of titanium, smaller amounts of vanadium and carbon and oxygen.

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Fig. 1: A) Large spherical object (LSO) isolated from the stratosphere showing a smaller sphere attached (marked X) and surface debris; Y and Z indicate respectively surface cracking and putative bacteria (see below). **B)** EDX analysis of the LSO.



Fig. 2: A) Hexagonal plates on the surface of the LSO, presumably resulting from impact; marked Y on Fig. 1 and B) a long fault line on the surface of the LSO, enlarged from Fig. 5. Note presumptive biological entities.

Figure 6A shows a close up of the impact crater and the attachment of the matrix material to the crater and its emergence through the hole in the LSO; it is not clear whether this is a morphological, pre-formed, pore or merely a pressure-rip in the surface of the LSO caused by the particle having been dragged away from the impact crater. **Figure 6B** shows that the material issuing from the LSO is made up of carbon and oxygen and lacks titanium and vanadium. The video made of the nonomanipulation clearly shows that the matrix material, attached to the impact crater, emerged from the LSO as it was dragged across the surface of the stub by the nanomanipulator.

Figure 7A shows a close-up of the surface covering present at the lower end of the LSO; EDX analysis shows the presence of large amounts of carbon, oxygen and titanium (**Fig. 7B**). Finally, **Fig. 8A** shows a close-up image of the surface damage caused to the right lower part of the LSO. Below a thin amorphous surface layer can be clearly seen



Fig. 3: A) Partially collapsed bacteria-like spheres on the LSO surface marked on Fig. 1. with a Z; B) shows a detail of the SSO and filamentous bacteria-like filaments on the right hand side.



Fig. 4: Nanomanipultor needle (X) returned to foreground after removing the large spherical object from its impact resting place, showing impact site and material issuing from the LSO.

a distinct "knitted" pattern of filaments, some of which bifurcate and appear biological in nature (**Fig. 8B**).

The biological nature of the particle

We base our claim that the LSO contains biology on the fact that:

- (a) its sub-surface layers contain carbonaceous material (i.e. dominated by carbon and oxygen), and
- (b) that this is present in the form of a fibrous "knitted" layer made up of individual filaments which bifurcate in a manner typical of biological forms, e.g. of fungi and the filamentous matrices of



Fig. 5: A) Detail of large spherical object and material issuing from it at point where the object was dragged from its impact site. Note damage to surface layer and underlying subsurface and smaller sphere at rear of the LSO, following rotation of the LSO under micromanipulation. **B)** EDX analysis of the LSO.



Fig. 6: A) Detail of the carbonaceous material issuing from the LSO and the impact crater. B) EDX analysis of the LSO.

algae. The inner content, which is seen oozing out of the LSO during nanomanipulation are also composed predominantly of carbon and oxygen, indicating carbonaceous material which we consider makes up the main protoplast of this biological entity.

Titanium and vanadium are common on Earth and throughout the cosmos, and have also been found on

Mars and in comets. Titanium also exists on Earth, as man-made titanium dioxide microspheres and in coalderived soot (Lida et al., 1998, Shabtai & Fleminger, 1994). A remarkable feature of the LSO shell, however, is that it is made up of only two metals titanium and, to a lesser extent, vanadium - a fact that rules out its origin as an Earth-derived pollution particle. Although some of the latter may superficially look similar to the LSO, unlike our particle, they are generally rich in alumino-silicates (Del



Fig. 7: A) Surface coating of the bottom of the LSO. B) EDX analysis of the LSO.



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Fig. 8: A) Filamentous, "knitted" fibrous material underneath the surface of the LSO (taken from point marked X on Fig. 5A) which was broken on impact or during micromanipulation. B) Detail of filament showing bifurcations typical of biology.

Monte & Sabbioni, 1984). The particles of terrestrial origin also do not possess the "knitted" layer of the LSO and any carbon they might contain would also likely be in the form of solid, granular soot particles which is not consistent with the organic ooze released from the LSO. Coal soot-based titanium balls of a terrestrial origin could not of course be carried up (by any known mechanism) to the heights from which the LSO was isolated. Nor could they have impacted the sampling stub to form a distinct crater as did the LSO.

The two biological entities seen in the LSO may be part of a single organism which formed the titanium ball, with

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the mycelial "knitted" material forming a subsurface structure, and the biological material which streams out from the sphere at the point of impact being the main protoplast of the organism. This would be analogous, for example, to the behaviour of an Earth-based algal protoplast within a siliceous shell (which can sometimes be associated with an organic cell wall matrix), or for example, a terrestrial testate amoeba, having a calcium and iron shell. No organisms which produce titanium shells are however, known on Earth.

It is also possible that the LSO organism is capable of autotrophically using reduced forms of titanium minerals (in the presence of low intensity UV) as an energy source with the titanium shell it produces serving to protect the organism from high levels of cosmic UVC. Titanium, in its labile forms (particularly as nanoparticles of titanium oxide) is toxic to Earth microorganisms so, while the titanium sphere itself may not be poisonous, if it is to use titanium ions as a component of its biochemistry this stratosphere-isolated organism would have had to develop resistance to the element's toxic effects. In the past two decades microorganisms have been discovered in the most unusual and unexpected locations on the Earth and nearly every available source of chemical energy has been seen to be harnessed to drive microbial metabolism. Relevant to the present discussion it has recently been discovered that the bacterium, Delftia acidovorans produces solid gold by catalysing exothermic reactions in the presence of the element (Johnson et al., 2013). In the process Delftia acidovorans can also protect itself against toxicity from soluble gold, metallic gold being biologically inert. We postulate that a similar process may be operating in the formation of a titanium shell in the LSO seen here. Other instances of biological concentrations of rare elements may include the case of red rain cells, where recently a high uranium enhancement was found in the outer cell walls (Miyake et al., 2013).

Alternatively, the titanium sphere (LSO) isolated here may have originally been a purely inorganic entity, formed by a chemical process under astronomical conditions, and the mycelial mat and inner protoplast material might belong to unconnected organisms; a biological entity(s) may thus have used a pre-formed titanium sphere as a ready-made home, rather than itself playing a role in its formation, i.e. analogous to, for example, a hermit crab occupying a pre-formed shell; the borrowed titanium sphere would again protect the organism(s) from the damaging effects of UV light, we consider this to be far-fetched however.

The origin of the LSO

The fundamental point regarding the findings presented here is that a biological entity has impacted a sampling stub at sufficient speed to cause a marked impact event. This impact event, we argue, could only be caused by the entity arriving at speeds of ~ 1 km/s from above the stratosphere, inside a cometary bolide. The particle we claim is too large to have been elevated to 22–27km by a volcano (no major volcanic eruption occurred in the three years preceding the sampling event and no typical volcanic dust is associated with any of our isolated biomorphs). As we have discussed at length elsewhere (Wainwright et al., 2013a, b), there is no known mechanism which could elevate this 30 micron LSO found to the stratosphere, certainly no conceivable mechanism could elevate a particle of this size from Earth to a height of between 22-27 km at a speed sufficient for it to cause the marked impact event we observe on the graphite sampling stub. It could be argued that the particle might have been elevated to a great height from the Earth (say 100km), by an unknown mechanism, and then fell from this height, gaining sufficient speed to cause the observed impact crater; again we ask -what mechanism is available to account for such an extreme elevation of a 30 micron particle? Perhaps the particle was originally an Earth-derived contaminant of space debris, a satellite or an orbiting space station. This possibility would appear to have an extremely low probability of occurrence, especially since our earlier reported calculations suggest that large quantities of biological particles are incoming to the stratosphere from space- far too much to be derived from "space junk". Algae are reported to have been ejected from Earth to the stratosphere during a super volcano eruption some 2.4 thousand years ago (Van Eaton, 2013), but these particles will have obviously long since returned to Earth, unless there were in some way captured and amplified in number elsewhere.

We conclude therefore that the LSO, studied here contains elements of a likely biological origin and was incoming at speed to the stratosphere from space (probably inside a small cometary bolide) when it impacted the graphite sampling stub and caused the observed impact crater.

The heresy that biological complexity arrives to Earth from space

From observations based on our recent sampling studies of the stratosphere, we argue that biological complexity is presently continually arriving to Earth from space, mainly from comets. Since no fundamental change has occurred in the relationship between the cosmos and Earth for aeons, we maintain that biological complexity must have always been arriving in this process and played a crucial role in the disposition and evolution of terrestrial life. The rate of input will obviously have changed and will have been maximal during the mass cometary impacts which, we assert, brought biology of varying levels of complexity to the prebiotic Earth, biology which continues to arrive to Earth from space. Despite the mind-set of most orthodox biologists, Earth has always been open to the cosmos and there has never been an "Eden Project-style dome" disconnecting the Earth biome from a potential space biome. It may be that the biological entities which arrive from space have always been in the main dead, although their DNA might still be capable of being integrated with Earth biology (Hoyle and Wickramasinghe, 1982, Wickramasinghe, 2011, Wesson, 2010). Orthodox biologists will of course rail against such suggestions and point to the apparent certainties presented by evolutionary clocks and the fossil record. The critics of our work will of course simply claim that our assertions are mere sophisms, and that there must be an, as yet unknown, force by which particles of 30 microns and larger can be elevated to 22–27 km, without carrying contaminating Earth debris, and at a sufficient speed to cause an impact crater on a graphite stub. We await news of such a mechanism, the discovery of which will at enable us to claim that our work has, at least, led to the discovery of a remarkable force which is new to atmospheric physics.

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