SURVIVAL OF NANNOCHLOROPSIS PHYTOPLANKTON IN HYPERVELOCITY IMPACT EVENTS UP TO VELOCITIES OF 4 KM S^{-1} **D. L. S. Pasini[1],** M. C. Price[1], M. J. Burchell[1], and M. J. Cole[1].
[1]School of Physical Sciences, University of Kent, Canterbury, Kent, CT2 7NH, UK (corresponding author: dp335@kent.ac.uk).

Introduction:

Studies have previously been conducted to verify the survivability of living cells during hypervelocity impact events to test the panspermia and litho-panspermia hypothesis [1], [2]. It has been demonstrated that bacteria survive impacts up to 5.4 km s^{-1} (approx. shock pressure 30 GPa) – albeit with a low probability of survival [1] whilst larger more complex objects (such as seeds) break up at ~1 km s^{-1} [2]. The survivability of yeast spores in impacts up to 7.4 km s^{-1} has also recently been shown [3]. We demonstrate here the survivability of Nannochloropsis Phytoplankton, a eukaryotic photosynthesizing autotroph found in the 'euphotic zone'(sunlit surface layers of oceans) [4] at impact velocities up to 4 km s^{-1}. Phytoplankton from a culture sample was frozen and then fired into water (to simulate oceanic impacts, as described in [5]) using a light gas gun (LGG) [6]. The water was then retrieved and placed into a sealed culture vessel and left under a constant light source to check the viability of any remnant organisms.

Methodology:

Table 1. LGG shot parameters of shot programme.

Shot ID	Intended velocity (km s^{-1})	Measured velocity (km s^{-1})	Approx. shock pressure (GPa)
G220312#1	1.25	1.257	2.90
G041012#1	1.25	1.246	2.90
G101012#2	2.50	2.600	8.70
G251012#2	2.50	2.330	7.30
G311012#1	3.50	3.280	12.6
G221112#3†	1.25	1.316	3.10
G281112#2	4.00	3.930	17.0

†Control shot performed using ice, not Phytoplankton.

Two sterile sealed glass bottles were prepared for each shot, one for the control sample, the other for the retrieved fired sample. 700 ml of HPLC grade water was mixed with 3.5 ml of nutrient, 400 ml of this was then placed directly into one of the sealed glass bottles as a control, the remaining 300 ml was used for the target. A sterile, hollowed out projectile was filled with a mixture of water and phytoplankton, then frozen overnight to approx. -20°C (Fig. 1). This projectile was then transferred to the LGG a few minutes prior to firing. Table 1 gives details of the shot programme including the intended and measured impact velocity, and the approximate shock pressure of the impact.

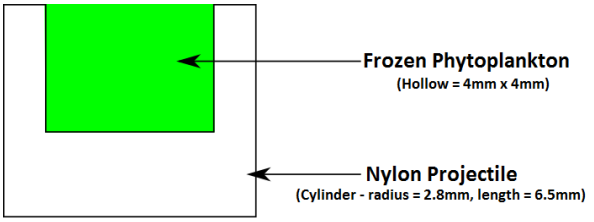

Fig. 1. Schematic showing Projectile structure.

The target was a sterilised polythene bag (50 microns thick) filled with approximately 300 ml of the target fluid. The bag was mounted in a specially designed target holder previously autoclaved at 120 °C at 1 bar for 30 minutes prior to each shot. Sterilisation of the immediate surroundings was performed by soaking in isopropyl alcohol. The launch tube was cooled overnight to approx. -140°C, and coolant (approx. -28°C) was used to cool the gun. The pressure in the target chamber was lowered to 50 mBar and the shot performed. Immediately after the shot, the target chamber was returned to atmospheric pressure, the target holder removed, and the remaining water in the target holder was funneled into a glass bottle which was then immediately sealed and left to culture under constant lamp light. A temperature reading was taken at the original projectile location to verify the projectile was still frozen during the shot, these readings were always <-10°C. After the 'live' shots, a control shot was also performed. The target (and holder) were prepared in an identical fashion to the 'live' shots, but the projectile was a sterile hollowed out projectile filled with HPLC grade water and frozen. The control and recovered water was placed into glass bottles in an identical manner to the live phytoplankton shots.

Preparation of Nannochloropsis Phytoplankton:

The phytoplankton was first cultured in plastic bottles using a mixture of water and F/2 medium nutrient [7]. The culture was then split and recultured, to verify the culturing technique was repeatable. Then the culture was split in two halves, one half was recultured and the other was frozen to -20°C. The frozen sample was left frozen for one week, then it was cultured to see if the organism was able to survive the freezing process, as any transfer between planetry bodies would involve encountering the freezing temperatures of space. The frozen sample was successfully recultured, albeit at a slower rate than before, suggesting that not all of the organisms survive the process. This culture was then

used as the source for all the 'live' shots in the programme.

The growth media

The target fluid was a mixture of HPLC grade water (700 ml) and 'Phyto Nutrient - Modified F/2 Medium'(3.5 ml) purchased from Reefphyto [7]. The formula is based on the Guillard F/2 medium [8] and has exactly the same N, P, trace element, and vitamin content as the original F/2 medium.

Results:

To test the viability of the Phytoplankton collected after impact, the sample was placed in a glass bottle and left to culture under a constant lamp light. All samples were left to culture under the same light. Growth was witnessed in all shot samples (except the ice control shot) soon after culturing began (usually 7-10 days). The physiological characteristics of the Phytoplankton were checked under both a light microscope and a scanning electron microscope, and were found to be the same as the unfired samples, thus demonstrating that the organisms had indeed survived. The fired samples continued to show an increase in growth with time indicating healthy reproduction of the organisms, as shown in Fig. 2. These results lead to the conclusion that the Phytoplankton collected were indeed survivors from the shots. However, the rate of growth was significantly lower than for the unshocked Phytoplankton. The appearance of the recovered samples showed no green colouration immediatly after the shot, but once it began to grow it was not significantly different in appearance to the unshocked samples. No significant growth was witnessed in the control samples. Being agents for primary production (the creation of organic compounds from carbon dioxide dissolved in the water, a process that sustains the aquatic food web) [4], Phytoplankton are excellent candidates for survival on another world, transforming its waters (and atmosphere, via the release of oxygen) into an enviroment conducive to life as we know it. They are the base of a food chain on Earth and could serve the same function on another world to lifeforms that subsequently evolve from them.

Survivability probability.

No quantitive measurements have yet been performed on the survivability probability as a function of impact velocity for the phytoplankton. A qualitative observation is that as the impact velocity increased, the cultures took longer to establish the same level of observed growth. This constraint is not very tight as the number of organisms delivered to the target, and subsequently recovered, is unknown. Work is currently in hand to quantify this and produce a survival rate.

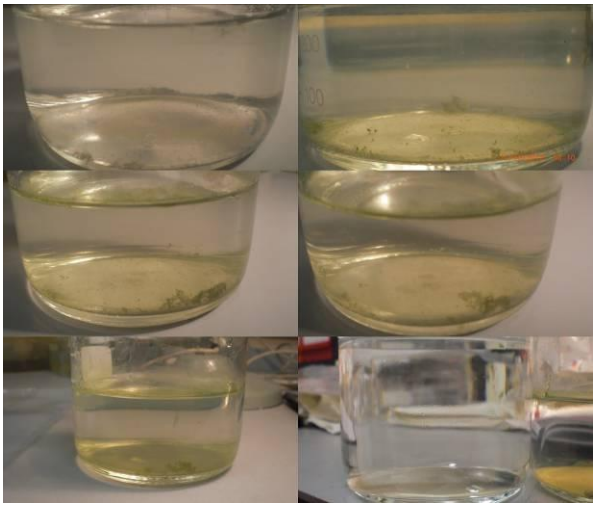

Fig. 2. Growth of Phytoplankton for shot G041010#1. Left to right, top to bottom, 2, 9, 13, 26, 56 days, and control sample for comparison.

Shock pressure experienced during impact:

The approximate maximum shock pressure, *P*, for each impact was calculated using Eqn. (1) from [1] which allows for a finite projectile impacting a flat target:

$$P = \frac{mv}{2V_p}\left(C + \frac{sv}{2}\right) \qquad \text{Eqn. (1)}$$

where *v*, V_p, and *m*, are the projectile velocity, volume, and mass respectively, *C* (1.48 km s^{-1}) and *s* (1.60) are the linear shock wave speed parameters for water [9]. These calculations (Table 1) show that the plankton have survived shock pressures up to ~17 GPa.

Conclusions:

We have extended the range of organisms that survive hypervelocity impacts to include an ocean dwelling photosynthesizing micro-organism. Other groups have also reported that lichens are able to survive shocks in similar pressure ranges[10]. This demonstrates that in addition to bacteria, and yeast, life forms that can serve as the base of a food chain and transform an environment making it suitable to life as we know it, could survive the ejection and re-impact onto a planetary body (Mars or the Moon for example), thus giving a foothold to life on another world.

References:

[1] Burchell M. J. et. al. (2004). *MNRAS*, 352; 1273. **[2]** Jerling A. et. al. (2008). *Int. J. Astrobiology*, 7; 217. **[3]** Price M. C. et al. (2013). *Icarus, 222*, 263. **[4]** Ghosal S. et. al. (2002). *NASA/TM-2001-210935*, 88. **[5]** Milner D. J. et. al. (2006). *Int. J. Astrobiology*, 5; 261. **[6]** Burchell M. J. et al. (1999). *Meas. Sci & Tech.*, 10; 41. **[7]** http://www.reefphyto.co.uk/ accessed Jan. 2013. **[8]** Guillard R, et. Al. (1963). *Can. J. Microbiol*, 8; 229. **[9]** Melosh H. J., (1989). *Oxford Uni. Press*. **[10]** Horneck G., et al. (2008) *Astrobiology, 8*, 17.