SURVIVAL OF THE TARDIGRADE HYPSIBIUS DUJARDINI DURING HYPERVELOCITY IMPACT EVENTS UP TO 3.23 KM S^{-1}. D. L. S. Pasini[1], M. C. Price[1], M. J. Burchell[1], and M. J. Cole[1].
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Introduction:

Studies have previously been conducted to verify the survivability of living cells during hypervelocity impact events to test the panspermia and lithopanspermia hypotheses [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]. Previous work by the authors demonstrated the survivability of Nannochloropsis Oculata Phytoplankton, a eukaryotic photosynthesizing autotroph found in the 'euphotic zone' (sunlit surface layers of oceans [4]), at impact velocities up to 6.07 km s^{-1} [5]. Other groups have also reported that lichens are able to survive shocks in similar pressure ranges [6]. However, whilst many simple single celled organisms have now been shown to survive such impacts (and the associated pressures) as those encountered during the migration of material from one planet to another [1, 3, 5], complex multicellular organisms have either largely not been tested or, those that have been, have not survived the process [2]. Hypsibius dujardini, like most species of tardigrade, are complex organisms composed of approximately 40,000 cells [7]. When humidity decreases they enter a highly dehydrated state known as a 'tun' and can survive extreme temperatures (as low as -253°C or as high as 151°C), as well as exposure to X-rays and the vacuum of space [7]. Here we test Hypsibius dujardini by firing a nylon projectile onto a frozen sample of water containing frozen tardigrades using a light gas gun (LGG) [8]. The recovered ice and water was then analysed under an optical microscope to check the viability of any remnant organisms that may have survived the impact (and the associated pressures).

Methodology:

Several original samples of tardigrades were sourced from Sciento [9]. These were first analysed to ascertain how many viable organisms were in each sample, then the samples were divided up and placed in a freezer at approx. -20°C. Two test shots were performed initally to test the target structure's durability. Next, a shot program ranging in velocity from 0.372 to 3.23 km s^{-1} was undertaken, firing a cylindrical nylon projectile (diameter 4.4 mm × length 4.5 mm) at a reinforced target of frozen water containing frozen tardigrades (51 × 51 × 10 mm). For each sample fired upon, another was also removed from the freezer and thawed, this served as the unshocked control. Tables 1 & 2 give details of the shot programme, including measured impact velocity, the approximate shock pressure of the impact, and the range of pressures felt across the target. The target was mounted in a specially designed target holder and the pressure in the target chamber was lowered to 50 mBar and at which point the gun was fired. Immediately after the shot, the target chamber was returned to atmospheric pressure, the target holder removed, and the remaining water and ice in the target holder were collected and analysed under a optical microscope to search for surviving tardigrades.

Table 1. Parameters of shot programme so far undertaken.

Shot ID.	*Velocity (km s^{-1})*	*Pressure Eqn 1. (MPa)*	*Pressure AUTODYN (MPa)*
Test 1	1.009	-	-
Test 2	1.225	-	-
Shot 1	0.372	380	4.15 – 71.1
Shot 2	1.028	1361	24.8 – 166
Shot 3	1.950	3409	99.9 – 368
Shot 4	3.230	7548	219 – 971

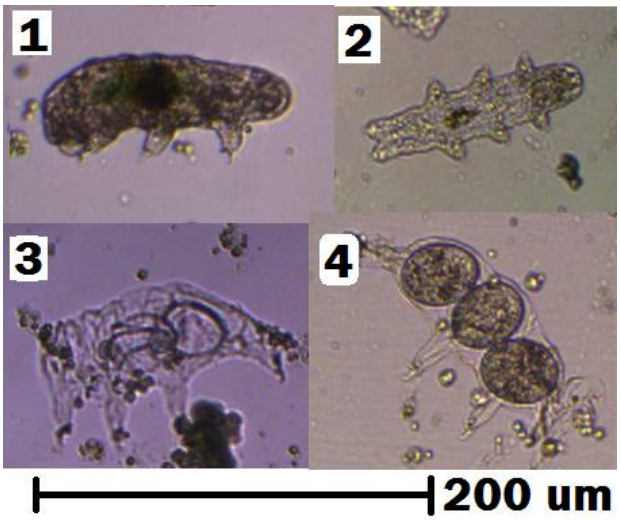

Fig. 1. Optical images of Hypsibius dujardini at 100× zoom. **1.** A living organism. **2.** A dead organism. **3.** A discarded husk. **4.** An egg laden husk.

Results:

To test the viability of the shocked samples, the ice and water collected after impact were left to thaw overnight, and then analysed under an optical microscope. In all of the shocked samples surviving organisms were found. These surviving organisms were found to be in an active state, moving around and eating algea, just as the unshocked samples showed.

Table 2.

Shot ID.	*Time Frozen (Days)*	*Living-to-dead ratio (shocked Sample)*	*Living-to-dead ratio (unshocked control)*
Origin	N/A	N/A	1.26
Shot 1	14	0.120	0.28
Shot 2	07	0.115	0.32
Shot 3	21	0.111	0.31
Shot 4	01	0.076	0.37

*Origin sample = unfrozen, and unshocked.

Survivability ratio:

The original sample of tardigrades showed a ratio of living-to-dead organisms of 1.26. After freezing four samples for varying time intervals this drops significantly (Table 2). However, the survival rate appears to be constant regardless of the length of time the sample is frozen, with a mean of living-to-dead ratio of 0.322 ± 0.034. The four shocked samples show a decreased survival rate with a clear trend such that as the impact velocity increases (and thus, the shock pressure increases), the living-to-dead ratio of the organisms drops significantly (Table 2 & Fig. 2). Work is ongoing to perform shots at increased velocities to add high speed data to the data presented here in order to give a more accurate trend and define the point of total lethality of the organism (as well as obtain some repeatability statistics).

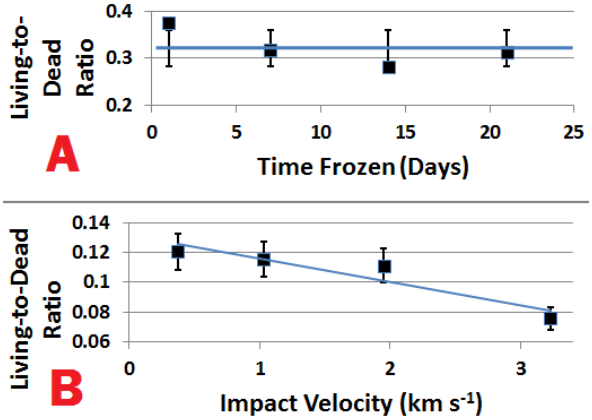

Fig. 2. A. Graph of freeze time vs. living-to-dead ratio (unshocked). **B.** Graph of impact velocity vs. living-to-dead ratio (shocked).

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 (m s^{-1}), volume (m^3), and mass (kg) respectively, C (1480 m s^{-1}) and s (1.60) are the linear shock wave speed parameters for water [10]. These calculations show the peak shock pressures at the impact point. However, to get a better understanding of the range of pressures experienced across the whole target (and thus, what the organisms experienced) a series of simulated impacts were run using Ansys' AUTODYN software using a 2-D Lagrangian mesh solverwith axial symmetry to accurately gauge the pressures involved (Table 1).

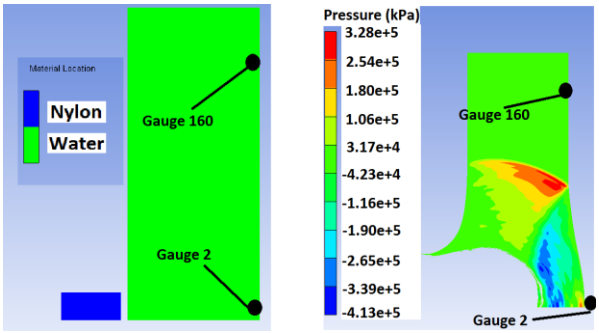

Fig 3. Ansys AUTODYN simulation showing: **Left.** Set up showing gauges #2 and #160. **Right.** Pressure contours during 3.23 km s^{-1} impact, (snapshot 6.13 µs after impact).

Conclusions:

We have extended the range of organisms that survive hypervelocity impacts to include, for the first time, a complex multi-cellular micro-animal. This demonstrates that in addition to bacteria, yeast, and phytoplankton, the complex multi-cellular life form Hypsibius dujardini could survive the ejection and re-impact onto a planetary body, such as Mars, the Moon, or Europa for example. Work is ongoing, at higher impact speeds and various freezing temperatures.

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]** Pasini D. L. S. et al. *LPSC44*, 1497. (2013). **[6]** Horneck G., et al. (2008) *Astrobiology*, *8*, 17. **[7]** Seki K. et al. *Nature*, 395. 853-854. (1998). **[8]** Burchell M. J. et al. (1999). *Meas. Sci & Tech.*, 10; 41. **[9]** www.sciento.co.uk (accessed Nov 2013). **[10]** Melosh H. J., (1989). *Oxford Uni. Press*.