Isolation of the *Paenibacillus phoenicis*, a Spore-Forming Bacterium

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A microorganism was isolated from the surfaces of the cleanroom facility in which the Phoenix lander was assembled. The isolated bacterial strain was subjected to a comprehensive polyphasic analysis to characterize its taxonomic position. Both phenotypic and phylogenetic analyses clearly indicate that this isolate belongs to the genus *Paenibacillus* and represents a novel species.

Bacillus spores have been utilized to assess the degree and level of microbiological contamination on spacecraft and their associated spacecraft assembly facilities. Spores of *Bacillus* species are of particular concern to planetary protection due to the extreme resistance of some members of the genus to space environmental conditions such as UV and gamma radiation, vacuum, oxidation, and temperature fluctuation. These resistive spore phenotypes have enhanced potential for transfer, and subsequent proliferation, of terrestrial microbes on another solar body. Due to decreased nutrient conditions within spacecraft assembly facility clean rooms, the vegetative cells of *Bacillus* species and other spore-forming *Paenibacillus* species are induced to sporulate, thereby enhancing their survivability of bioreduction

This work was done by James N. Benardini, Parag A. Vaishampayan, and Kasthuri J. Venkateswaran of Caltech; Shariff Osman of Lawrence Livermore National Laboratory; and Masataka Satomi of the Japanese National Research Institute for NASA's Jet Propulsion Laboratory. For more information, contact iaoffice@jpl.nasa.gov. NPO-47232

Monolithically Integrated, Mechanically Resilient Carbon-Based Probes for Scanning Probe Microscopy

These probes can be used in medical applications for bacteria or protein imaging.

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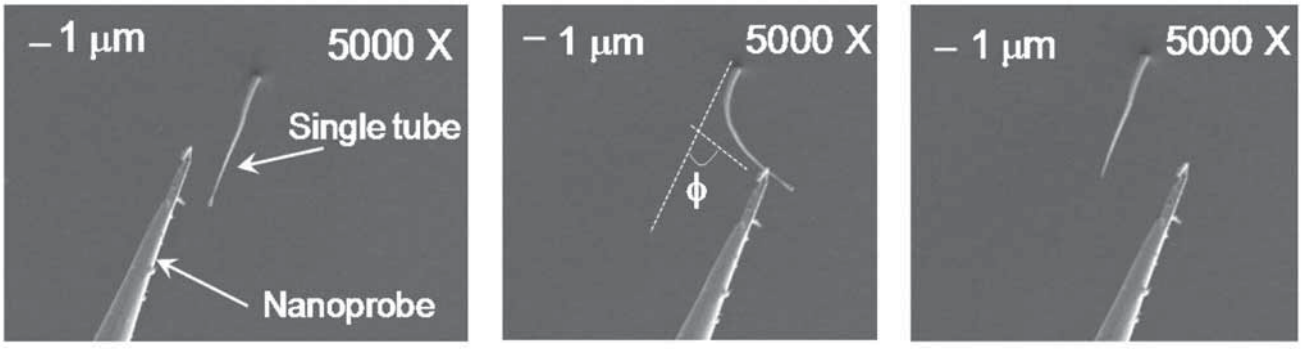
Scanning probe microscopy (SPM) is an important tool for performing measurements at the nanoscale in imaging bacteria or proteins in biology, as well as in the electronics industry. An essential element of SPM is a sharp, stable tip that possesses a small radius of curvature to enhance spatial resolution. Existing techniques for forming such tips are not ideal. High-aspect-ratio, monolithically integrated, as-grown carbon nanofibers (CNFs) have been formed that show promise for SPM applications by overcoming the limitations present in wet chemical and separate substrate etching processes.

The CNFs of this innovation have been synthesized in a load-lock-based DC PECVD (plasma-enhanced chemical vapor desposition) growth chamber, where the CNF growth was done on Si substrate with high-purity acetylene (C_2H_2) and ammonia (NH_3) at 700 °C. The ratio of C_2H_2:NH_3 = [1:4], which has been determined to minimize the amount of amorphous carbon on the substrate during growth. When the desired growth pressure was attained (3–15 Torr), a DC glow discharge was ignited, and growth was continued for a fixed duration. The PECVD growth parameters, such as growth pressure, catalyst thickness, and plasma power, were var-



The **CNFs Are Mechanically Resilient** and should enable enhanced cycling longevity for NEMS applications: (a) A nanoprobe was in close proximity to a single CNF. (b) The probe was mechanically manipulated so that it deflected the CNF to the right. The CNF accommodated large bending angle without fracture or delamination, with $\phi \approx 70^{o}$ over tens of cycles. (c) The CNF returned elastically to its initial position after the probe was removed.

ied to see their impact on the physical characteristics of the CNFs (e.g., diameter and length).

The mechanical characteristics of the CNFs were measured in a custom-built *in-situ* mechanical deformation instrument, the SEMentor, comprising a scanning electron microscope (SEM) and the nanoindenter. This instrument has generally been used to explore uniaxial deformation and defect evolution in individual, metallic pillars formed by using the focused-ion-beam (FIB), for example.

Bending tests were performed with a nanoprobe that deflected an individual CNF, and provided insight into their mechanical resilience in shear. *In-situ* electrical measurements were then conducted on individual, as-grown CNFs using a nanomanipulator probe stage mounted inside an SEM (FEI Quanta 200F) that was equipped with an electrical feed-through. Tungsten probes were used to make the two-terminal electrical measurements of individual, vertically oriented, as-grown CNFs with an HP4156C parameter analyzer.

For SPM applications, stress concentrators may exist at the CNF-to-substrate interface, as well as within the body. *In-situ* uniaxial compression tests were performed on arrays of CNFs inside the SEMentor, which provided some insight into the nature of the mechanical bond between the CNF and substrate. A Berkovich tip, which is a pyramidal, shallow-angled tip, was used to indent the fo0rest of CNFs. The SEM image taken after indentation revealed that the CNFs fractured within the tube body rather than at the CNF-to-substrate interface, where a fracture angle $\alpha_f \approx$ 25°–35° (relative to the CNF or central axis) was computed.

The significance of α_f was correlated to the structural characteristics of the CNFs, which were deciphered from transmission electron microscopy (TEM) that was performed with FEI Tecnai-F20 Scanning –(S) TEM, with a field emission source of 200 kV. The TEM analysis of the mechanically transferred CNFs grown directly on Si revealed a herringbone structure where the graphite basal planes were inclined to the central axis at a cone angle α, where $\alpha \approx 30°$. Since α_f and α did not differ appreciably, the nanomechanical measurements performed in the SEMentor confirm that the CNFs sheared from within the basal planes of the CNFs, indicating that the adhesion of the CNFs to the substrate was very strong. In addition, the image in the figure shows the CNFs can tolerate a large degree of mechanical strain where bending angles ϕ as large as 70° could be accommodated elastically, confirming the promise such carbon-based nanostructures have for SPM applications..

This work was done by Anupama B. Kaul, Krikor G. Megerian, Andrew T. Jennings, and Julia R. Greer of Caltech for NASA's Jet Propulsion Laboratory. For more information, contact iaoffice@jpl.nasa.gov. NPO-47185

Cell Radiation Experiment System

Cells can be irradiated under conditions that approximate those in living tissues.

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The cell radiation experiment system (CRES) is a perfused-cell culture apparatus, within which cells from humans or other animals can (1) be maintained in homeostasis while (2) being exposed to ionizing radiation during controlled intervals and (3) being monitored to determine the effects of radiation and the repair of radiation damage. The CRES can be used, for example, to determine effects of drug, radiation, and combined drug and radiation treatments on both normal and tumor cells. The CRES can also be used to analyze the effects of radiosensitive or radioprotectant drugs on cells subjected to radiation. The knowledge gained by use of the CRES is expected to contribute to the development of better cancer treatments and of better protection for astronauts, medical-equipment operators, and nuclear-power-plant workers, and others exposed frequently to ionizing radiation.

Traditionally, experiments to determine the effects of ionizing radiation on cells involved (1) culturing the cells in test tubes, Petri dishes, or culture flasks; (2) removing the cells from the cultures and exposing them to radiation; and (3) reculturing the cells to enable the cells to attempt to repair the radiation damage and continue to grow. The great disadvantage of the traditional approach is that cells are subjected to a succession of environments that differ radically from the precisely controlled natural environment in a human or other animal body; the effects of the succession of nonlifelike environments can alter the subtle effects of radiation damage mechanisms and intracellular repair processes, thereby introducing uncertainty into interpretation of experimental observations. By maintaining more nearly lifelike conditions, the CRES can increase the accuracy of, and confidence in, experimental observations.

The CRES (see figure) includes one or more cell-culture chambers equipped with a very thin, impermeable Mylar (or equivalent polyethylene terephthalate) membrane at one end, described in more detail below. At the opposite end of each culture chamber there is a perfusion chamber separated from a culture chamber by a permeable membrane. Through this membrane, waste and nutrients are exchanged between the culture and perfusion chambers. A circulation subsystem that includes fluid reservoirs, conduits, valves, pumps, and automated process controls provides for the slow perfusion of the nutrient medium used to culture the cells.

The system includes a source of ionizing radiation in a shielded enclosure with motor-driven shutters that are used to effect timed, selective irradiation of the cell-culture chamber(s). A process-control module exerts overall control over the circulation subsystem and other subsystems to regulate such parameters as the temperature, the rate of circulation of fresh nutrient medium, the pH of the medium, and radiation doses via the shutters. Another subsystem monitors the metabolic rates of cells by measuring ultraviolet fluorescence from the cells, the pH of the medium, and/or the concentrations of O_2 and CO_2. Still another subsystem under control by the process-control module can be made to introduce a fixative substance to preserve the cell cultures for subsequent analysis. The impermeable front membrane is thin enough to allow the ionizing radiation to pass through. Before installation, the inner face of this