

## Use of cyanobacteria in a closed ecosystem in space

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Cyanobacteria are the first organisms that acquired the ability to evolve O<sub>2</sub> by photosynthesis. Thus evolved O<sub>2</sub> has made the environment of the Earth oxygenic. Cyanobacteria has been surviving through serious changes in natural environment, and now they are distributing all over the world. *Spirulina*, a kind of cyanobacteria, is commercially available as a food supplement. Cyanobacteria are very useful organisms when we human beings are going to expand to space. The International Space Station (ISS) is one of the closed ecosystem in space. It is composed of astronauts and microorganisms that are brought into space ship by astronauts. Incidentally, no cyanobacteria have been detected in ISS. It is necessary to keep the life of astronauts, inside of the machine-full space ship, safe and comfortable. We are trying to grow plants or microalgae in the space ship, because their photosynthetic O<sub>2</sub> evolution and CO<sub>2</sub> fixation activity would make the intra space ship environment clean and, in addition, their green color would surely make astronauts feel easy. However, the effects of space environment, such as micro gravity and cosmic ray, on the fundamental photosynthetic mechanisms have not yet been determined. Now, we are going to grow cyanobacteria in the satellite which will be launched in the Indian-Japan Space Corroboration Experiments and to estimate their photosynthetic activity in space. We have selected two filamentous cyanobacteria, *Spirulina (Arthrospira) platensis* NIES-39 and *Nostoc sp.* as the test materials. The *Spirulina* is edible and its full genome sequence has been determined. We have made full automatic on board culture chamber. The size of the chamber is 20cm depth x 20cm width x 10cm height covered with an aluminium box. The *Spirulina* cells grown under laboratory conditions were washed by centrifugation and then re-suspended in a sterile culture medium containing 5 atom % of H<sub>2</sub><sup>18</sup>O and 4atom % of NaH<sup>13</sup>CO<sub>3</sub>. Each 10 mL of cell suspension was inoculated into 6 transparent plastic bags that were placed between LED panels. The light intensity was adjusted at 20 μmoles m<sup>-2</sup> sec<sup>-1</sup> at the surface of a bag and bags were continually illuminated. After appropriate time intervals, each 10mL of pure ethanol was introduced to the bags by a diaphragm pump to stop the reaction. After 2 weeks experiment, the volume of gas phase of each bag was measured and then concentrations of O<sub>2</sub> and CO<sub>2</sub> were measured by newly developed GC/MS system (Shimadzu GCMS-QP2010 Plus) equipped with micro volume gas sampler. O<sub>2</sub> was evolved constantly under the experimental conditions though the values are fluctuated by sampling error. The isotope ratio of the evolved gas was increased as the incubation prolonged and reached at the value which is calculated from 5 atom % H<sub>2</sub><sup>18</sup>O. Thus, effectiveness to use a stable isotope in measuring O<sub>2</sub> evolution was established. No CO<sub>2</sub> was detected under the illumination. Incorporation of <sup>13</sup>C into the cells was increased linearly with time and its value was well correlated to that of O<sub>2</sub> evolution. In another experiment, a terrestrial cyanobacterium, *Nostoc sp.* harvested from the field, was once dried and then put into a plastic bag (6 x 5 cm). The cells were wetted by a small amount of water and then illuminated by LED (660nm) light. After appropriate time intervals, O<sub>2</sub> and CO<sub>2</sub> concentration in the bag were measured using the GC/MS system. In the dark, O<sub>2</sub> was consumed and CO<sub>2</sub> was evolved, conversely in the light, CO<sub>2</sub> was consumed and O<sub>2</sub> was evolved. It is concluded that O<sub>2</sub> evolution and CO<sub>2</sub> fixation were precisely measured by this experimental system.

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