

2. PAST AND PRESENT OF SPACE FARMING

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2.1 History of space farming

Approximately 60 years have passed since the United States and the former Soviet Union began space development using artificial satellites and manned spacecraft in the 1960s. In 2018, the International Space station (ISS) celebrated its 20th anniversary of operation. Humans have been trying to extend viable areas to the vicinity of the moon, the lunar surface, and Mars. Food production is the key technology needed to make long-term manned space activities sustainable with environmental control, automation, and recycling. The Controlled Ecological Life Support System (CELSS) encompasses these techniques, and the Bioregenerative Life Support System (BLSS) utilize organisms for life support systems.

From the early stages of space development, the United States and Russia (including the former Soviet Union) have also actively addressed plant cultivation in space vessels. Plant research in space environments has been categorized into gravitational physiological research exploring gravity-sensing mechanisms and plant responses to light and gravity, space agronomic research with the goal of sustained crop production with agronomic approaches, and plant physiological research that has been mainstream in space using relatively small cultivation devices. Large-scale cultivation studies aimed at BLSSs have been conducted in laboratory facilities on earth. Terrestrial closed ecosystem cultivation facility research is being conducted by Russia, the USA, Europe, Japan, and Canada, or in cooperation. More recently, China has been actively conducting research. Fig. 2.1 shows a summary of the history of BLSS research ¹⁾. Fig. 2.1 History of bioregenerative life support system (BLSS) research

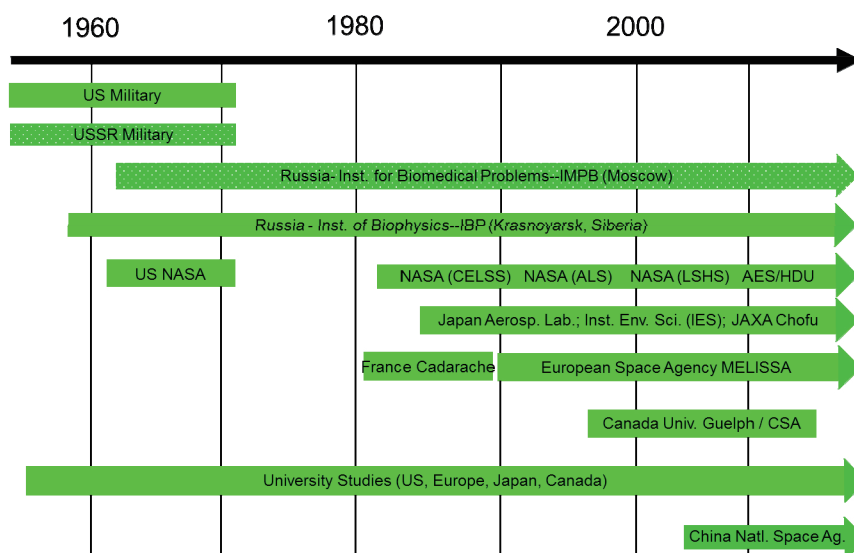


Fig. 2.1 History of bioregenerative life support system (BLSS) research ¹⁾

2.2 Use of algae

In early life support technology research in space development from the 1950s to 1960s, research on oxygen generation and carbon dioxide removal was actively conducted using algae, such as *Chlorella*. However, the weight and electric power of the photosynthetic air regeneration system were not suitable for the short-term stay

in space of the Mercury and Gemini projects, and they were not adopted. However, there is room for examination of the efficacy of utilizing algae for long-term stays, and it has been shown that it is necessary to solve problems such as edibility and the generation of volatile gases ²⁾.

2.3 Space farming by each country

2.3.1 Russia (including the former Soviet Union)

As a ground experiment regarding life support research in closed environments, Russia has conducted plant cultivation research for life support in closed environments using the ground facility BIOS, set up in Krasnoyarsk in Siberia since the 1950s (Fig. 2.2)²⁾. Studies on the recycling of gases, nutrients, and water were conducted using wheat and other species in BIOS. Studies utilizing plants for food production and oxygenation were conducted in phytotrons with a cultivated area of approximately 20 m² for more than 15 years, and algae were also studied. However, because the algae culture tank was connected to the plant cultivation area, the growth of tomatoes and potatoes ceased, the flowering of cucumbers stopped, leaves became etiolated, and an accumulation of anthocyanin was observed in the leaves of beets. Consequently, problems, such as the suspected generation of toxic volatile gas components by algae that inhibited plant cultivation became clear. Subsequently, quantitative cultivation data were obtained with a total cultivation area of 41 m² using two BIOS-3 phytotrons. Experiments using growing wheat in regenerated water from human urine revealed the need for sodium removal in water recycling, including sodium accumulation in culture water.

The BIOS-3 research team conducted ground experiments with the premise of developing bioregenerative technology as a large-scale closed life support system. However, the Russian Institute for Biomedical Problems (IMBP) began cultivation experiments in space, set on their manned space station and later on the ISS. Russia launched the first manned space station, Salyut, in 1971 and conducted plant experiments using the Oasis plant cultivation system³⁾. Russia has repeatedly improved the cultivation equipment with improvements in the space station and installed the SVET plant cultivation equipment in their next manned space station Mir (Fig. 2.3).

Additionally, the Russians continuously developed and used space cultivation equipment, such as the Lada plant cultivation equipment (Fig. 2.4) installed in the Russian module of the ISS. The cultivation of various plants, including wheat, barley, soybeans, and mizuna, has been attempted, and data on water and gas environments have been acquired. At the beginning of the experiments in Salyut and Mir, growth inhibition and fruiting failure were observed. It was finally concluded that it was because of the accumulation of ethylene, suggesting the necessity of gas exchange and an ethylene removal filter inside and outside the cultivation equipment. The goal of IMBP is to produce food in a microgravity environment during the period of travel to Mars by advancing research at the space station²⁾.



Fig. 2.2 BIOS-3 by Russia



Fig. 2.3 Plant cultivation on the Russian Space station Mir (provided by NASA)



Fig. 2.4 Plant cultivation in the ISS Russia module (provided by NASA)

2.3.2 United States

(1) Overall

In 1958, just before NASA was launched, the Biologistics Symposium was held in Ohio to create a list of crops that would be needed to support people in space²⁾. The criteria of growing even at low-light levels, with compact size, high yield, and resistance to salt (sodium), which is a problem in recycled urine, were organized and 13 types of crops, such as sweet potatoes, lettuce, Chinese cabbage, cabbage, turnips, and cauliflower were selected. In the 1960s and 1970s, the U.S. military was also heavily involved in the space program, and plant research for life support was conducted. Then, high-light output by artificial light became technically possible because of technological advances over the previous several decades, and research progressed to design cultivation spaces such as vertical farming, hydroponics, and attempts to increase photosynthetic efficiency through high CO₂ cultivation⁴⁾.

In the 1980s, NASA re-engaged in CELSS research and discussed crop cultivation during workshops. At NASA, plant-related studies were conducted in three laboratories: the Kennedy Space Center (KSC), Lyndon B. Johnson Space Center (JSC), and Ames Research Center (ARC). NASA also granted research funds to universities⁵⁾. As a result, wheat, rice, potatoes, sweet potatoes, soybeans, peanuts, and lettuce were selected as crops cultivated in CELSS. A study was conducted in which NASA and the university shared data on cultivation methods, growth measurements, and gas exchange. In addition to KSC, Purdue University, Utah State University, University of Wisconsin, and Tuskegee University conducted the experiments (Fig. 2.5)¹⁾. At the Advanced Life Support Plant/Food Production Meeting in 1997, 23 crop species were proposed in recognition of the nutritional aspects necessary for life support, crop yield index, ease of cooking, and cultivation performance in a closed

environment⁶⁾. The use of LED light sources in space and vertical/multi-stage farming were technological innovations, and space use research became a useful example of terrestrial agriculture.



Fig. 2.5 Previous closed ecological test facilities at NASA¹⁾

- A) Ames Research Center Closed Chamber System, B) Purdue University Minitrons,
C) Johnson Space Center Variable Pressure Growth Chamber,
D) Kennedy Space Center Biomass Production Chamber**

(2) NASA bio-regeneration type on-ground tests

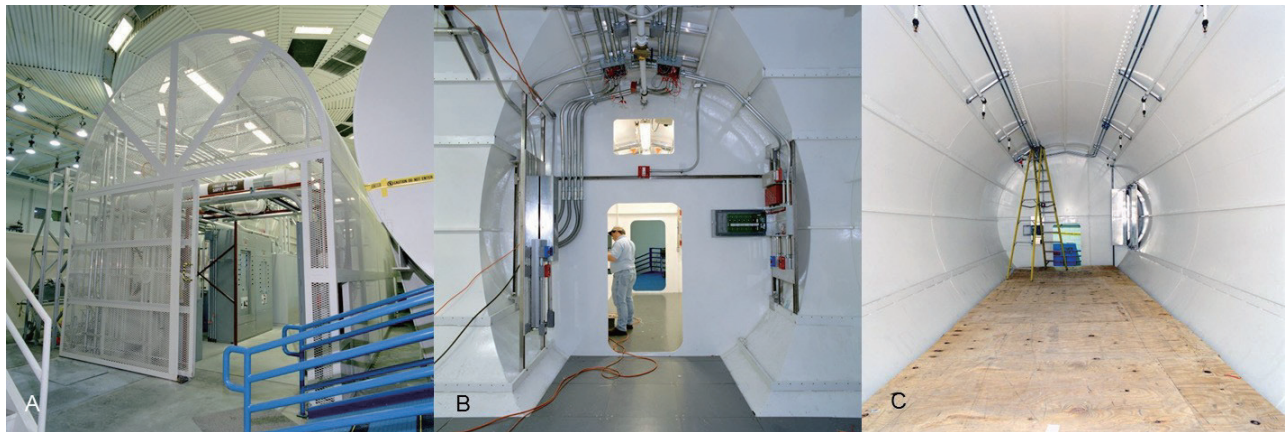
Similar to the Russian BIOS project, research by NASA also took into account the application for use in space stations and on planets, and research is being conducted to control the environment, such as water supply and drainage, with the premise that there will be gravity. Ground-based studies have been extensive for closed ecosystem life support systems, especially for bioregenerative studies involving plant cultivation, because of their considerable size. Ground-based experiments funded by NASA have been extensively studied in small-scale growth chambers with cultivated areas ranging from 1 to 4 m². Additionally, the Biomass Production Chamber (BPC), operated at KSC from 1988 to 2000, is well known as a large-scale experimental facility (Fig. 2.5D). BPC has 20 m² of cultivation area, and a vegetation cultivation study was conducted for life support in space in a closed system. The BPC utilized NFT-hydroponic cultivation, with four shelves placed in a 7.5 m high chamber. Here, wheat cultivation was conducted for 86 d, potatoes for 105 d, soybeans for 90 d, lettuce for 28 d, tomatoes for 85 d, as well as rice and radish cultivation. Data on yield changes due to carbon dioxide concentration and ethylene accumulation were collected²⁾.

In particular, NASA had a Lunar Mars Life Support Project (LMLSTP), an example of simultaneous food production and residence at a ground experiment facility. LMLSTP was a project conducted at NASA JSC from 1995 to 1998⁷⁾. It was conducted in a four-stage closure experiment. Phase I was an experiment involving one person for 15 d, while phase III was an experiment involving four people over 91 d. In particular, in Phase III, air and water were implemented as a completely closed system, and food and waste treatment were implemented as a partially closed system (Table 2.1)⁸⁾.

Table 2.1 Lunar Mars Life Support Project (LMLSTP) ⁸⁾

Test	Phase I	Phase II	Phase IIA	Phase III
Duration	15 days	30 days	60 days	91 days
Crew	1	4	4	4
Types of Systems	Biological (Wheat)	Physicochemical (Advanced)	Physicochemical (ISS Regenerative ECLSS)	Integrated Physicochemical & Biological (Advanced)
Full Closure	Air	Air & Water	Air & Water	Air & Water
Partial Closure				Food & Waste
Open Loop	Water, Food & Waste	Food & Waste	Food & Waste	

NASA also built BIO-Plex (Bioregenerative Planetary Life Support Systems Test Complex) at JSC. BIO-Plex was equipped with two large agricultural modules (Biomass Production System), each 80 m²⁹⁾; however, the plan was canceled in the 2000s. The reason for the cancellation was to concentrate funds on NASA's constellation plan, and a full-scale experiment as a closed life support system with biological regeneration was terminated at a stage where only the structure (Fig. 2.6)¹⁰⁾ was completed because of policy changes caused in part by a lack of planned funds. The Obama administration also canceled the constellation plan in 2010.

**Fig. 2. 6 Bioregenerative Life Support Systems Test Complex (BIO-Plex) ¹⁰⁾**

Constructed as a large manned facility for four people living within for 1 year and constructed at the NASA Johnson Space Center.

A) View of exterior, B) View of interior, C)View of interior

(3) Space experiment

NASA has been developing cultivation element technology that can function even in a microgravity environment simultaneously with the CELSS research assuming a planet where gravity exists. Many studies have been conducted on water supply systems that are easily affected by gravity and photosynthetic efficiency because of the light environment. NASA provided research funds to universities and others to research water supply systems that use porous tubes and membranes. These achievements led to the system design of a salad machine that provides astronauts with fresh vegetables. Although the size of the experiment was small, the plant

cultivation equipment Astroculture (ASC) and Advanced Astroculture (ADVASC) were installed on the Space Shuttle and ISS from the 1990s to the 2000s, and cultivation experiments were conducted. Later, plant cultivation equipment, such as the Plant Generic Bioprocessing Apparatus (PGBA), Biomass Production System (BPS), and more recently Veggie (Fig. 2.7), were developed by NASA and NASA-funded research. Most were used in gravitational biological studies and were small experiments for food supplies.

In an example of an experiment that focused on space agriculture using space equipment, Russia and NASA jointly grew wheat in the plant cultivation equipment SVET on the space station Mir¹¹⁾, potatoes in ASC¹²⁾, and wheat in BPS¹³⁾ measuring photosynthetic efficiency. The Veggie, which has been installed on the ISS since 2014, has a simple design with an LED light source, a fan that moves the air, and a water supply system operated by the crew. It has also been used in plant physiology experiments and experiments on the cultivation of lettuce and other plants. Bacterial examinations were also conducted with an awareness of HACCP in space, and the cultivated samples were frozen and examined on earth, followed by the first formal astronaut-eating event of lettuce cultivated in August 2015, the second time it was cultivated (Fig. 2.7C). Next, Zinnias were harvested in 2016, and Tokyo Bekana was harvested in 2017. Tomato cultivation was planned for 2018¹⁴⁾. Additionally, the United States launched the Advanced Plant Habitat (APH) on the ISS in 2017 and installed it in the Japanese Experiment Module "Kibo" to conduct the first cultivation experiment (Fig. 2.8).



Fig. 2.7 ISS NASA cultivation equipment Veggie (provided by NASA and Food Production for Space Exploration¹⁴⁾)

A) Veggie appearance (photographed on earth), B) Astronaut Steve Swanson harvesting lettuce on the ISS (June 2014), C) Astronauts Kjell Lindgren, Scott Kelly, and Kimiya Yui growing lettuce on the ISS, (August 2015) and D) Zinnias blooming on the ISS (February 2016)



Fig. 2. 8 NASA Plant Cultivation Device “Advanced Plant Habitat” (APH)

A) Wheat growing inside APH, B) Science Carrier removed from APH with wheat, and C) Astronaut Norishige Kanai removing the Science Carrier where the wheat grew from APH (provided by NASA)

(4) Non-NASA space farming-like terrestrial experiment equipment, “Biosphere 2”

Biosphere 2 is a large privately sponsored closed environmental facility built in Oracle, Arizona, from the 1980s to the early 1990s. It was planned that 2,720 m³ of soil would be brought into the vast 2,000 m² agricultural area to provide 80% of the food for eight people to live for 2 years. In a complex system containing a wide variety of plants and animals, the goal was to challenge life support and biological regeneration techniques in a closed environment with a view for space applications. The closure experiment was conducted twice for 2 years from 1991 to 1993 and for the following 6 months; however, the first 2-year test failed to maintain a completely closed environment, and in the 16th month, the oxygen concentration became 14%, and the closed system was forced to open. Insects and birds also died because of fluctuations in carbon dioxide concentrations¹⁵⁾, and there were also food shortages. Changes in the gas environment speculated to have been caused by a lack of sunshine and soil bacteria respiring beyond expectations¹⁶⁾. In addition to these factors, it has been noted that there were psychological problems between the crew and operation staff.

(5) Mars Simulation Experimental Facility MDRS

The Mars Desert Research Station (MDRS) is a Mars simulation facility owned by the Mars Society, a U.S. nonprofit organization in Utah, the United States. Approximately six teams stayed for 2–3 weeks and experimented with simulating residence on Mars. From Japan, Mars Society Japan dispatched eight participants in 2014 and 2015. A food-producing and plant-science laboratory called GreenHab has also been established to study the type and quantity of food required for future Mars manned missions. Recently, Yusuke Murakami of the Mars Society Japan participated in a long-stay mission with a total stay of 160 d in both Utah and the Arctic.

2.3.3 Europe

The European Space Agency (ESA) has been conducting a biological environment maintenance research project, including material circulation, since 1987, known as the Micro-Ecological Life Support System Alternative (MELiSSA) project. The early MELiSSA project was a waste treatment concept using microorganisms and cyanobacteria for biomass production; however, it is now expanding to including plant cultivation. MELiSSA also involves crop monitoring by remote sensing and has acquired data on the cultivation

conditions of plants, such as wheat, beets, and soybeans, in a closed hydroponic environment²⁾.

ESA has developed the European Modular Cultivation System (EMCS), a cultivation device for space, and it has been using it on the ISS since 2006 for plant research in space¹⁹⁾. Additionally, the German Aerospace Center (DLR) has led the development and operation of the EDEN ISS in Antarctica to research crop cultivation that will serve as food for the manned exploration era (Fig. 2.9). The EDEN ISS also has significance as a spin-off of space utilization²⁰⁾. It is a multi-stage cultivation module for supplying fresh vegetables in the polar regions and is the only one with space utilization. The EDEN ISS container was shipped to Antarctica in 2018, operates 400 m from the German Antarctic base Neumayer-Station III, and is currently conducting cultivation experiments²¹⁾.



Fig. 2. 9 ESA EDEN ISS plant cultivation container

- A) Cultivation shelves in EDEN ISS containers,**
 - B) Tomatoes, C) Cucumbers, D) Basil, and E) Radish grown in EDEN ISS,**
 - F) Operation control room in Bremen,**
 - G) EDEN ISS appearance set up in Antarctica and transport boxes with harvested vegetables carried by sled to the Neumayer Station III**
- Source: Facebook, @spaceedeniss**

2.3.4 Japan

(1) Japanese CELSS study

The study of space agriculture in Japan is aimed at application on Mars as a destination for manned activities and was began approximately 30 years ago by Prof. Masamichi Yamashita. Activities at ISAS and the "Space Agriculture Salon" included the cultivation of highly salt-tolerant plants, honeybee flight under low gravity, and examination of menus using insect foods, such as silk moths.

However, as in CELSS research, the National Aerospace Laboratory of Japan (NAL) has been developing bioregeneration technology for closed ecosystem life support systems since the latter half of the 1980s. The

Closed Ecology Experimental Facilities (CEEF) were constructed as a facility where two people could live for 120 d in a completely closed environment with plant cultivation²²⁾. Built between 1994 and 1999 at the Institute for Environmental Science and Technology in Rokkasho Village, Aomori Prefecture, this facility was created to simulate radioactive nuclear dynamics in the ecosystem. This was the experimental facility that Keiji Nitta of the same institute had fostered since the NAL era. It was a regeneration device that extracted C and O₂ from CO₂ via hydrogen using a wet oxidation device that decomposed under high temperature, high pressure, and a Sabatier reaction. It was an advanced Environmental Control and Life Support System (ECLSS) incorporating elemental technologies leading to manned space development²³⁾. Notably, a small goat (Shiba goat) was introduced as a part of the waste treatment (ingesting the inedible parts of the plants and contributing to weight reduction and decomposition). Near the facility's completion, four researchers were selected and hired as candidates for residence since 2000 and have completed training in plant cultivation, equipment calibration, safety, and health management. In 2005, three 1-week living experiments were conducted, in 2006, 1-week living experiments were conducted six times, and in 2007, 2-week living experiments were conducted three times, followed by 4-week living experiments, after which the closed residence experiment was completed. All plant cultivation was conducted using hydroponics without soil. Rice was cultivated as a carbohydrate source in a 60 m² area, soybeans as a protein source in a 30 m² area, peanuts as a fat source in a 30 m² area, and 20 types of vegetables in a 30 m² area in anticipation of the need for other vitamins and minerals²²⁾²⁴⁾. In this experiment, the self-sufficiency rate was high (up to 95% for residents, 100% for goats). In addition to the practice of processing and feeding crops harvested in a closed system, Russia and NASA have been researching bacterial flora, monitoring the psychological state of residents, and organizing troubleshooting techniques unique to closed systems. The results were rigorously evaluated, even compared with the previous experiments²⁴⁾.

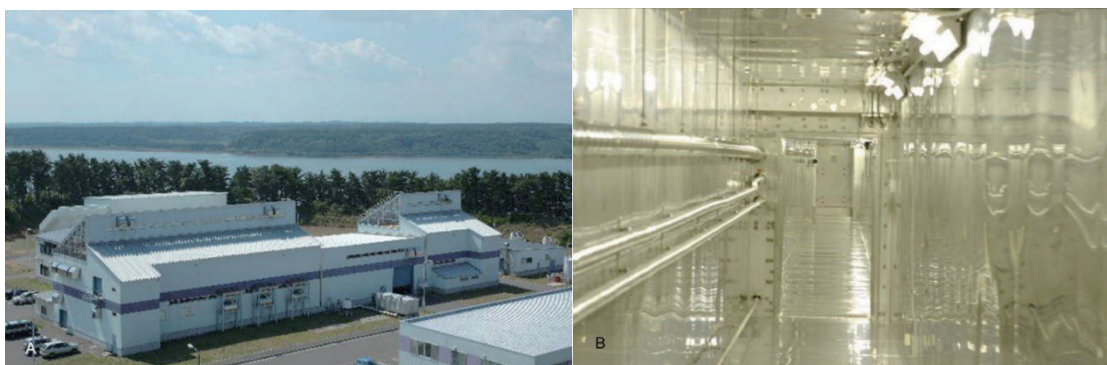


Fig. 2.10 Appearance and Interior of the Closed Ecology Experimental Facilities (CEEF) within the Institute for Environmental Sciences (IES)

A) CEEF and its support facilities were built inside a common building, B) CEEF constructed a highly airtight closed environment with stainless steel and glass without any concrete, and all crops were cultivated using hydroponics²²⁾



Fig. 2.11 Crops and Vegetables cultivated in CEEF ^{22) 24)}

A) Sequentially grown rice, B) soybeans, C) tomatoes D) cabbage, E) and sequentially grown crowndaisies, F) carrots, and G) vegetables

(2) Space experiment

Plant growth experiments in the dark for several days in space have been conducted on the Space Shuttle and the International Space Station. The results showed that, in addition to the lack of gravitropism in the microgravity environment, the constituents of the plants' cell walls changed, and the water tropism of the roots was more likely to appear. Regarding the Seed-to-Seed experiment, if the results of experiments in Russia, the United States, Europe, Japan, and other countries worldwide were combined, we can see that it is possible to complete the life cycle of plants and the growing environment can be adjusted, even in space. Japan has also succeeded in the Seed-to-Seed experiment (Space Seed experiment, Fig. 2.12) of *Arabidopsis thaliana* using the plant experiment unit (PEU) in the cell culture device on the ISS Japanese Experiment Module "Kibo"²⁵⁾.

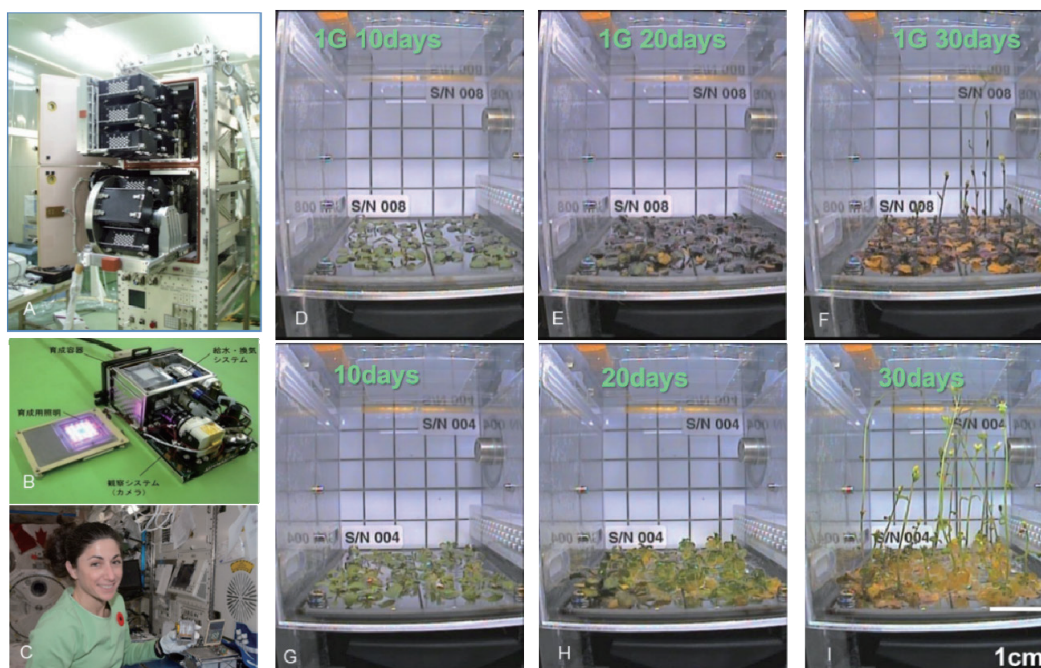


Fig. 2.12 Japanese ISS-plant cultivation experiment (Space Seed)

- A)** Cell Biology Experiment Facility (CBEF) is used cell culture and plant cultivation hardware in the ISS Japanese Experiment Module “Kibo”,
B) Plant experiment unit used in the cell culture device, **C)** NASA astronaut Nicole Stott conducting *Arabidopsis* cultivation experiments, **D–F)** *Arabidopsis thaliana* in PEU installed in the artificial gravity part of CBEF, and **G–I)** *Arabidopsis thaliana* in PEU installed in the microgravity portion of CBEF (provided by JAXA)

(3) Ground experiment

Because constant gravity exists on the planet's surface, it is speculated that problems specific to microgravity will not be applicable for cultivation on the planet surface. However, a microgravity environment was present while moving to a station near the moon (deep space gateway²⁶⁾) and planets, including Mars. During ground research focused on space plant cultivation conducted in Japan, research teams from five universities (Tohoku University, Tokyo University, Osaka Prefecture University, Utsunomiya University, and Tokai University) conducted cultivation experiments, including plant cultivation box design, ground product production, and aircraft experiments, such as "research on the life cycle of plants under microgravity and development of microgravitational field plant experiment equipment for that purpose." This research clarified that the design and verification of the lighting system, air conditioning system, airflow control, and nutrient water supply system, which are not easily affected by gravity, were important issues among the elemental technologies to realize plant cultivation in space²⁷⁾. Based on these achievements, the research progressed to technical element extraction work in the Lunar Farm Working Group of the Space Exploration Innovation Hub.

(4) Expert Committee on the Science of the Use of the Space Environment

One of the academic community support activities led by ISAS is the Space Environment Use Scientific Expert Committee. Since 2017, this expert committee has positioned plant cultivation research at the forefront of research and has provided support.

(5) Research on Elemental Technologies for Crop Culturing to Secure Stable Food during Manned Planetary Exploration

In 2015, research on elemental technologies for crop cultivation to secure stable food during manned planetary exploration was noted as technological development necessary for manned planetary exploration, and research at the JAXA Tsukuba Space Center began. They conducted research to cultivate crops to secure food for planetary bases, provide fresh vegetables for astronauts en route to the planet, and cultivate plants that contribute to mental health support. Utilizing existing plant factory technology, a system was created to collaborate with research institutes, such as universities and industries with research experience. This activity is being conducted simultaneously with the Lunar Farm Working Group activity of the Space Exploration Innovation Hub, and members of the Lunar Farm Working Group are participating in activities such as in the Advisory Committee.

2.3.5 China

Lunar palace-1 is a 160 m² (500 m³) enclosed residential experimental facility for four people built at Beihang University in Beijing, China. In the 105 d closed-living experiment from 2013 to 2014, a closed-living experiment was conducted with three people and the plant cultivation module half-operated [100 m² (300 m³)], and the productivity of 1.6 people was confirmed. At the time of this experiment, wheat was cultivated in 30 m², tiger nuts (Chufa) in 10 m², soybeans in 5.6 m², and eight species, such as leafy vegetables and strawberries in 12.7 m². The cultivation shelves were multi-stage with red and blue LEDs. One of the unique points was that animal breeding was not conducted, mealworms were bred for the treatment of non-edible parts and food, and feeding during the life of the actual experiment was also conducted (Fig. 2.13)²⁸⁾. From 2017 to 2018, the system was operated continuously for 370 d, two teams of four people with different metabolic rates alternately lived within 110, 200, and 60 d, and the reports indicated that the experiment was completed²⁹⁾. Researchers at Peking University have made most academic reports concerning the 105 d experiment in 2014. Academic reports have been made in various fields, including space development, ecological engineering, agriculture/plant physiology, and flora. Closed circulation has not been completed in the previous reports, and the details regarding waste treatment have not been clarified. Treatment by microorganisms was assumed, and the report of the results of this 370 d experiment is awaited.

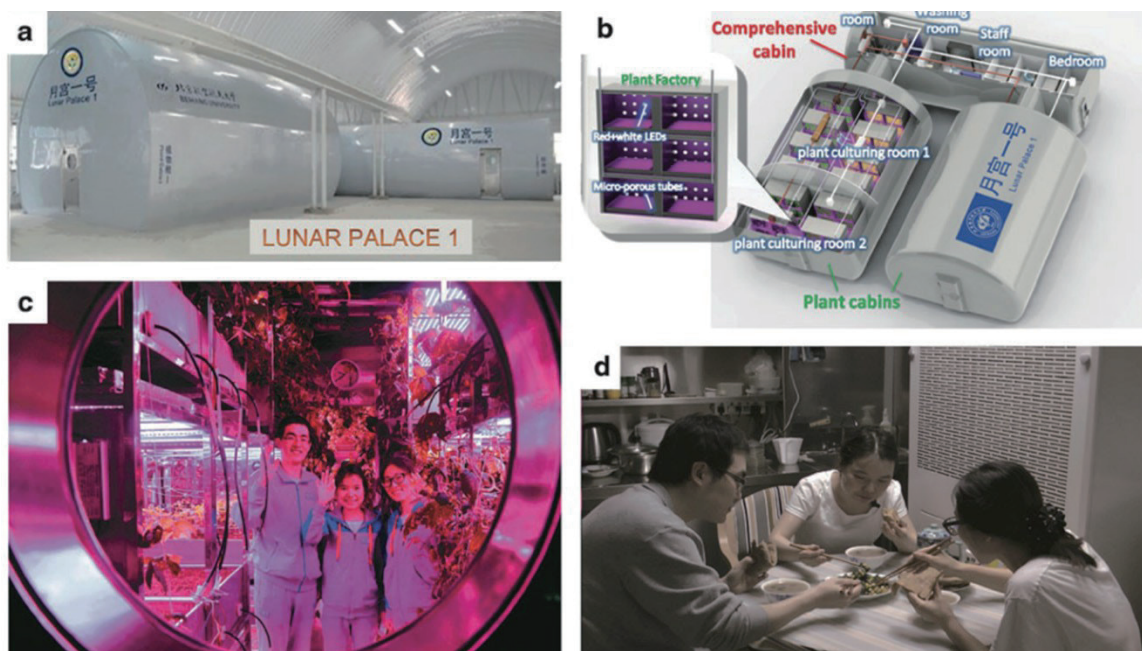


Fig. 2.13 Lunar Palace-1²⁸⁾

Lunar Palace-1 a) Appearance, b) Illustration of the internal appearance, c) Three crew members during the 105 d experiment, and d) Mealtime during experiments

2.3.6 Others

NASA has collaborated with researchers at the University of Arizona's School of Agriculture to develop a prototype of an inflatable greenhouse for farming in space, according to information from May 5, 2017³⁰⁾. Additionally, Russia reported a proposal for a device based on a unique idea, a six-sided cultivation device premised on space use³¹⁾.

2.4 Plant physiology in low-gravity environments

Plants acquire a mechanism that accepts and responds to the surrounding environmental stimuli; in particular, they manage their life cycle while utilizing gravity³³⁾. Previous studies on plant growth in the space environment have revealed characteristics, such as changes in plant cell wall constituents and the tendency for root water tropism to appear. It is also known that it is possible to complete the plant life cycle by adjusting the growth environment²⁷⁾. However, changes in plant physiology and cultivation conditions under microgravity and gravity environments have attracted attention, and there is no record of long-term cultivation under low gravity because of restrictions in the cultivation equipment and centrifuges.

Regarding the alternation of generations of plants in space, there was a problem forming fertile seeds in the space station in the early stages of space development. However, it has been found that plants can form seeds, even in the space environment, by control of the environmental factors affected by microgravity. However, abnormalities in plant aging and the fertility and shape of seeds have been observed in space, and they differ depending on the plant species.

Various plant growth phenomena affected by the gravitational response, such as water tropism, phototropism, apical dominance, and rotational turning movement, have been found in plants. Among them, research is progressing on the anti-gravity reaction, which is the reaction that conditions the body against gravity, and the direct bearer of this reaction is the cell wall. Plants use more than 90% of their fixed energy to build sturdy cell walls, and when low gravity reduces the need for anti-gravity reactions, plants will divert that energy to other needs³³⁾. It is interesting to observe the plant physiology that occurs when growing crops on the moon.

According to Kiss et al.³⁴⁾, the results of an experiment on phototropism (TOROPI-2) using *Arabidopsis thaliana*, which is widely used in plant research as a model for higher plants, indicated that the phototropism threshold for red and blue light is approximately 0.1 to 0.3 G. In low-gravity experiments using algae, the intracellular statoliths (amyloplasts) play a role regarding weights that settle at approximately 0.1 G to 0.2 G, depending on the direction of gravity. Consequently, it can be assumed that the low gravity of the Moon and Mars is closer to 1 G, which is the same as on the ground, rather than the microgravity environment for plants.

However, low gravity may affect the cultivation equipment and environment. From the results of a short-term microgravity experiment using aircraft flights, convection disappeared because of the absence of gravity, and the temperature increase may lead to a decrease in fruit set by suppressing the heat exchange between the plant and the surrounding environment³⁵⁾. Additionally, according to a recent short-term gravity fluctuation experiment using aircraft flights, in which the gravity condition was partially changed to a gravity of 1 G or less, it was suggested that low gravity might have a considerable effect on the water distribution in the medium and the environmental control of the cultivation system³⁶⁾. It is preferable to partially verify the effect of low gravity on the cultivation system.

2.5 Results of plant cultivation in the International Space Station

(1) Purpose of Cultivation Experiments in Space

Thus far, various plants have been tested on the Space Shuttle and ISS. Besides being a food source, plants are effective for manned space activities for environmental control in a closed environment because of carbon dioxide fixation by photosynthesis and absorption/evaporation of water. Therefore, many basic experiments and cultivation experiments have been conducted to confirm growth in space, gravity, light response, and root tropism³⁷⁾.

(2) Actual ISS experiments

Regardless of the shuttle era, the ISS has enabled continuous cultivation for an extended time. Equipment is also being developed by NASA, ESA, JAXA, and Russia and for joint missions. Approximately 80 plant experiments have been conducted on the ISS to date, and approximately half of them are experiments using the model plant *Arabidopsis thaliana*. Additionally, wheat, barley, soybeans, mizuna, tomatoes, radishes, and peas are cultivated on the ISS. There are also plans to grow bok choy, kale, and wasabi. There have been experimental examples using corn, cucumbers, rice, and lentils, but this experiment analyzed seedlings and roots and germination in the dark. Experiments have also been conducted using the legume Burr medic, the grass model plant *Brachypodium distachyon*, the spruce genus *Picea* (Pinaceae family), and *Quercus glauca*, poison ivy, and morning glory. Culture experiments with algae, such as *Chlorella* and *Chlamydomonas*, are also being conducted. Experiments have also been conducted on soils and plants such as rhizobia, *Bacillus subtilis*, staphylococci, and microorganisms closely related to health management.

Lettuce was cultivated multiple times from 2014 to 2015 by the NASA device Veggie. The first and second cultivations were frozen and recovered on the ground, and after analysis and microbiological inspection, they were provided to astronauts in orbit during the 2015 cultivation, and three astronauts, including astronaut Yui tasted them (Fig. 2.7).

In 2017, the NASA-developed Advanced Plant Habitat (APH) was installed in the Kibo inboard laboratory rack. APH has a cultivation area of 0.2 m², the largest space plant device to date, and *Arabidopsis thaliana* and wheat were cultivated for the initial verification in 2018. Fruiting of wheat has occurred (Fig. 2.8).

i) Potato cultivation

NASA is aware of the usefulness of potatoes in food production, and cultivation experiments on the Martian regolith stimulant and desert soil are being conducted in terrestrial laboratories. In space, a potato (Seed Potato) cultivation experiment was conducted in the 1995 Space Shuttle Mission STS-73, and a mid-deck rocker type cultivation device cultivated tuberous roots. The astroculture was analyzed on the ground¹²⁾. It was determined that there was no difference in tuberous root growth and starch particle size with that of the ground control group³⁸⁾. In this experiment, the flight was approximately 2 weeks; thus, the plants grown on the ground for 7 weeks were loaded. There was no example of germination or harvest from seed potatoes.

ii) Strawberry cultivation

Ground studies with the goal of space cultivation of strawberries have also been attempted by NASA and ESA (EDEN ISS), but there is no space cultivation record. There are problems regarding the transportation of seedlings, the length of the cultivation period, and pollination.

iii) Cultivation of wheat, barley, and soybeans

Much has been achieved using the NASA space cultivation equipment and Russian cultivation equipment for wheat, barley, and soybeans.

iv) Tomato cultivation

Tomatoes are described as a sample in the cultivation experiment using the Lada device installed in the Russian module, but there is no record of this in literature. Tomatoes were also planned to be grown using Veggie to test light quality and fertilizer¹⁴⁾. Long-term storage of seeds before the experiment was an issue, and the effect of pollen on astronauts was considered.

v) Cultivation of lettuce and Mizuna

There have been many achievements using NASA and Russian cultivation equipment for lettuce and Mizuna.

vi) Cultivation of sweet potato

There is no information on the cultivation of sweet potatoes, although its usefulness as a target for food production in space has been recognized^{6) 39)}.

vii) Cultivation in educational missions

Under the application conditions of space experiments where the amount of transportation and experimental space are limited, plants can be easily stored as seed samples, a large number of samples can be transported.

Seeds are relatively easy to control as biological experiment samples because experiments can be started using the water supply, and the influence of gravity on morphology can be easily observed. Therefore, many are cultivated in missions to demonstrate their cultivation and educate children and students. In 1994, Chiaki Mukai brought Kaiware daikon(Radish Sprout) to his first Space Shuttle flight and sprouted it in his cabin. Astronaut Donald Pettit of NASA used a Ziploc bag to grow zucchini without using any special equipment and published a record of flowering and fruiting with photos and explanations. Some astronauts raised sunflowers.

At the "Space Seed for Asian Future 2013" conducted by JAXA in 2013, an azuki bean germination experiment was conducted at ISS Kibo. Additionally, educational missions using peas and barley were being conducted using the Russian cultivation equipment Lada. The European Space Agency (ESA) was also conducting experiments using arugula in educational kits.

Viii) Other

Cultured Korean ginseng cells (Zhenshen, Ginseng) have been cultured in the Russian module of the ISS from 2007 to 2009. In 2016, lettuce was cultivated during an approximately 30 d manpower experiment in Tiangong-2, a space laboratory docked with Shenzhou-11, a Chinese manned spacecraft.

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