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Azolla as a component of the space diet during habitation on Mars

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Abstract

We evaluate a candidate diet and specify its space agricultural requirements for habitation on Mars. Rice, soybean, sweet potato and a green-yellow vegetable have been selected as the basic vegetarian menu. The addition of silkworm pupa, loach, and *Azolla* to that basic menu was found to meet human nutritional requirements. Co-culture of rice, *Azolla*, and loach is proposed for developing bio-regenerative life support capability with high efficiency of the usage of habitation and agriculture area. Agriculture designed under the severe constraints of limited materials resources in space would make a positive contribution toward solving the food shortages and environmental problems facing humans on Earth, and may provide an effective sustainable solution for our civilization.

Keywords: Space agriculture; Space diet

1. Introduction

Mars is a sister planet of Earth. Mars might be the only planet, other than Earth, where either extant or extinct living organisms could be found. This expectation drives us to plan a manned mission to Mars for astrobiology exploration. For habitation on extraterrestrial planets, such as Mars, bio-regenerative space agriculture [1–3] is required to support human life. By recycling bio-materials, the launch mass from Earth and landing mass on Mars could be reduced for missions with large crews and long operational periods. At the same time, design of space diet is important for making manned activities in outer space more creative and productive [4–6].

We evaluated a candidate menu designed for space agriculture. A combination of rice (*Oryza sativa*), soybean (*Glycine max*), sweet potato (*Ipomoea batatas*), green-yellow vegetable, silkworm (*Bombyx mori*) pupa [7–9], loach (*Misgurnus anguillicaudatus*) [10], and *Azolla* was examined to determine whether it is an appropriate diet that fulfills human nutritional requirements. The potential use of *Azolla* (an aquatic fern) and its life support capability in space has been studied [11,12]. Rice, *Azolla*, and loach will be cocultured in rice paddies in order to enhance productivity

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under limited available resources [13–15]. Symbiotic cyanobacteria in *Azolla* can quickly fix nitrogen when there is a shortage of nitrogen fertilizer. *Azolla* is an effective green manure, and also suppresses the growth of weeds by covering the water surface [16,17]. Coculture of fish is further advantageous, by converting *Azolla* to animal meat, and upgrading biomass to edible food. Action of fish or bird in rice paddies brings other positive effects on rice production as well. In this study, we examined the feasibility of adding *Azolla* to the human space habitation diet, and the nutritional value of the *Azolla* menu.

2. Outline of space agriculture

Space agriculture is engineered to support life by synthesizing an artificial ecosystem, which consists of plants, microorganisms and animals. Materials are recycled in a closed loop among those members. We define the requirements for space agriculture to initiate our design of life support system. For the Mars habitation system, we assume a population size of 100 people for a duration of 20 years. One hundred is the typical number of people on an Antarctic science expedition team. Although the crew members may not be fixed for the full 20-year period, due to change-out of crews at certain intervals, the crew size present at any time should remain fixed. Team of 100 crew members will be organized with scientist, facility engineer, medical staff, farmer, and so on.

Our space agriculture concept employs plants to (per person per day) regenerate 100 kg of water, revitalize 0.5 kg of oxygen from carbon dioxide, and produce 2 kg of food materials from human metabolic waste and inedible biomass. Composted inorganic components of waste are fed to plants in order to achieve the higher degree of closure in the materials recycle. However, we do not intend to close the loop 100%. We will conduct a trade-off to find the optimum closure level. Solar light is the energy source to drive space agricultural production. On-site planetary resources, such as bio-elements in Martian regolith, a dilute atmosphere, and subsurface water, are utilized for expanding space agriculture on Mars.

There are three loops of materials circulation in space agriculture. Waste water is irrigated to the plant farming yard. Based on the ratio between photosynthetic production and water respiration of ordinary plant species, 100 kg of water would be recovered from water vapor evaporated from plant leaves, during the production of food materials. The psychological problem of drinking recycled water could be eased by "natural" filtering of water through plants. Stoichiometry of the photosynthetic reaction produces oxygen at an amount equivalent to food biomass. Cultivation of trees is a core concept of our space agriculture. Tree leaves support sericulture and produce excess oxygen. Wooden lumber is a byproduct used to furnish the interior of the habitation module. Although the Martian atmosphere is diffuse, it contains carbon dioxide with trace nitrogen. A pressurized greenhouse dome creates living environment with air inside at 10 kPa partial pressure of oxygen, verified as the lowest pressure required for driving the life cycle of plants, balanced by nitrogen and minor H_2O and CO_2 to keep total pressure at 20 kPa [3]. We should note that the day length of Mars is close to 24 h, even though solar light intensity is about half.

3. Nutritional requirements

Next to water and oxygen demands, human nutritional requirements are factors that must be considered when designing space agriculture. As a nutritional reference, we chose the Standard of Foods Intake distributed by the Ministry of Health, Labour and Welfare of Japan [18]. In addition to the target level of energy intake and each nutrient, the allowance level is defined for many items. Considering that the levels depend on age, sex and physiological state, we took the standard for an adult under normal activity levels. Items analyzed in this study are energy, proteins, fat, carbohydrates, dietary fiber, vitamins, minerals, trace elements, and electrolytes. The detailed items are the same as those listed by Katayama et al. [8].

The ratio between proteins, fat, and carbohydrate (PFC ratio), for energy intake, was evaluated. The amount of essential amino acids was examined, in terms of balance of nutrients. The amino acid score is an index to examine if amino acids composition is adequate. Excess uptake of certain amino acids causes a heavier load on the nitrogen metabolic pathway.

Fat is an energy source complementary to carbohydrate. Because of the difficulty in defining requirements for fat uptake, we used standard recommendations for the ratio of energy uptake from fat. In Japan, the recommended energy intake from fats is 20–50% for adults, 45% for infants under six months, and 30–40% for pregnant and lactating women.

It is important to keep the balance of fatty acid groups within an appropriate range. Firstly, the best composition of fat intake occurs when the ratio of fat sources is mammalian meat: plant: fish = 4: 5: 1. Secondly, the ratio among saturated fatty acid (SFA), monounsaturated fatty acid (MUSFA), and polyunsaturated fatty acid (PUSFA) should ideally be SFA: MUSFA: PUSFA $= 3:4:3$. Thirdly, PUSFA is divided to two categories: *n*-6 fatty acid from plants, which are rich in linoleic acid, and *n*-3 fatty acid from fish, which are rich in linolenic acid, eicosa pentaenoic acid (EPA) or decosahexaenoic acid (DHA). The ratio between *n*-6 and *n*-3 is best at 4:1.

For cholesterol uptake, the Japanese reference defines only its daily upper limit of 600 and 750 mg/day for women and men, respectively. Cholesterol is an essential chemical family, necessary for the synthesis of bile acid, adrenal cortex hormone, and sex hormones. At the same time, its excessive intake induces hardening of arteries. The upper consumption limit is defined to avoid this kind of negative effect. On the other hand, insufficient cholesterol intake results in imperfect immune function and menstrual irregularity. Phospholipids are another important group of nutrient chemicals. Their chemical structure consists of two fatty acids bound to glycerin, and a third hydroxyl group of glycerin forms ester with phosphate. These phospholipids are the main constituent of the double lipid layer found in cell membranes. Both cholesterol and phospholipids are essential for the function of the bio-membrane.

Eighty percent of the fat in our body can be reused. The other 20% of fat must be obtained from foods. A group of PUSFAs, such as linoleic acid or linolenic acid, are essential because they cannot be synthesized in our body. These essential fatty acids are required for growth regulation of the human body. Their deficiency causes retardation of growth, pathological skin changes, and degradation of immune function. On the other hand, *n*-6 and *n*-3 fatty acids, PUSFAs with longer chains, can be synthesized in our body.

There are some items, for which the upper or lower intake limit is not defined in the reference. If excessive intake of that item never results in negative effects, then the upper limit is not defined for it. In case our designed menu cannot provide a certain nutrient at its recommended level, its impact shall be carefully evaluated at either its deficiency or excessiveness. We may also consider intake of trace elements or chemicals at their appropriate level, if they are essential for our health. Other indices may come to light, based on studies that will be conducted on human nutrition, especially those specific to the space environments.

Even though the nutritional value of a proposed space menu is high, we must take into consideration individual and cultural food preferences when designing it. When we propose exotic food materials or cooking methods, we shall take those aspects into account, in order to make it acceptable to humans.

4. Choice of elementary crop species for a basic space menu

Space agricultural production makes full use of limited resources. Inner volume and effective area of the greenhouse dome, and available flux of incident solar light are the factors that must be considered when we design the basic combination of food supply. The type of plant reproduction must be taken into account when selecting crop plants. Plant species with vegetative reproduction are easy to propagate. For species with anemophilous flowers, such as rice, wheat, and quinoa, airflow with sufficient wind speed is required at their blooming stage. Entomophilous flowers (e.g. soybean and buckwheat) depend on pollinating animals or their equivalent.

We selected rice, soybean, and sweet potato for our major plant species after consideration of the above points. The nutrient composition of each item was taken from the table issued by the Science and Technology Agency of Japan [19]. In addition to these species, Komatsuna (*Brassica rapa* var. peruviridis), Japanese mustard spinach, was included as a representative species of a required green-yellow vegetable. Composition of these four species was optimized on the basis of their contribution of carbohydrate, vitamins, minerals, and dietary fiber. It was found that a combination of 300 g rice, 120 g soybean, 200 g sweet potato and 300 g Komatsuna per person per day fills the nutritional requirement well. This combination is called a fundamental vegetable menu, and it provides energy and proteins from rice, proteins, and dietary fiber from soybean; energy, vitamins, and dietary fiber from sweet potato; and iron, calcium, vitamins, and dietary fiber from Komatsuna.

5. Demands for food of animal origin

Nutritional analysis of the basic vegetable menu consisting of rice, soybean, sweet potato, and Komatsuna reveals a shortage of Vitamins D and B_{12}, cholesterol, and sodium salt.

Since V-D deficiency causes demineralization of bone, supplementation with V-D might be critical for crew, members exposed to micro- or low gravity. V-B_{12} is essential to prevent pernicious anemia. Because the required amounts of these vitamins are minor, they could be supplied in the form of food additives, in case an appropriate set of food materials could not be composed. However, if possible we prefer to select natural food materials instead for space agriculture.

Ordinary plants are rich in potassium, but low in sodium. Under stressful situations, demand for sodium salt increases. In the Japanese standard of 2005, a daily intake of 9 g of sodium salt (NaCl) is recommended. Because ordinary food materials of our proposed daily menu contain about 2 g of sodium salt, an addition of 7 g of sodium salt is required in the form of cooking or table salt. The American standard recommendation for maximum sodium chloride intake is 5.8 g. If the American standard is used, 3.8 g of added salt is enough. When humans are exposed to low gravity on Mars, body fluid distribution shifts to the upper body, and excess water is lost as urine. When overall body fluid volume is reduced, there is increased load on the kidneys [20]. For kidney patients, salt uptake shall be reduced. In this context, we decided on the addition of 3 g of sodium salt in the form of cooking or table salt, for a total of 5 g in our space menu.

It is difficult to keep the PFC ratio in its preferred range, i.e. 15% protein, 15–20% fat, and 55–65% carbohydrate, when designing a menu with plants as the major food members. Plant materials are quite diverse in terms of nutrition, ranging from root crops characterized by their high carbohydrate content, to soybeans characterized as rich in protein. In general, vegetables are low in fat. This causes a poor PFC ratio in the vegetarian menu, and necessitates the intake of animal originated food materials to supplement fat. In terms of amino acids composition, lysine is deficient in many plant food materials. Addition of animal foods greatly increases the amino acids score.

Cholesterol is not available from plants, but can be obtained by adding appropriate animal origin food. V-D can be obtained from mushroom, egg, animal meat, or fish. Candidate source of V-B_{12} and cholesterol is mammalian meat or fish. Marine algae and shellfish, such as green string lettuce and clam, contain V-B_{12}.

6. Addition of insect to the basic space menu

In order to supplement the deficient nutrients of the core vegetarian menu, we considered insects. In our space agriculture concept, tree cultivation is implemented. Plant photosynthesis produces biomass and oxygen. If this biomass is oxidized or metabolized, oxygen is stoichiometrically consumed. By utilizing wood lumber for living space and furniture, excess oxygen can be stored and utilized. Insect rearing is also possible on tree leaves. Silkworm is our candidate insect. It has been domesticated for 5000 years. As a model animal for scientific research, the rearing protocol for silkworms is well established. In addition to this, the mulberry tree has been extensively studied for feeding silkworms. Many cultivars of mulberry are available. Furthermore, it is possible to make silkworm polytrophic. They can eat plant leaves other than mulberry. Artificial feeding materials that taste like mulberry have also been developed. Silkworms at the developmental stage of either pupa metamorphosed in cocoon or adult moth might be appropriate for utilization as a food source.

7. Loach and *Azolla* to the basic space menu

Silkworm pupa does not meet all of the nutritional requirements of our animal meat addition to the basic vegetarian space menu. Fish contains both V-D and V-B_{12}. Herring, pacific saury, red salmon, tilapia, and loach are species that are rich in vitamins. Tilapia has been included in the space menu of NASA. The vitamins in loach are similar to those in tilapia, except that V-D is less, at 4 mg per 100 g. We selected loach based on the following reason. Loach is a traditional fish species cultured in rice paddies in East Asia. Loach is a tough fish species. In poor quality water, such as low dissolved oxygen, loach will swim up to the water surface, gulp air into the digestive tube, and expel air bubbles from the anus after making gas exchange along the gut. Thus, culture conditions are quite permissive for loach. Since loach abdominal organs are rich in vitamins, it is beneficial to eat the whole body.

We decided to add *Azolla*, and aquatic fern, to our space menu that has rice as its core grain. Simultaneous production of plant crop and animal was proposed by the International Rice Research Institute, IRRI [15]. *Azolla* is cultivated on the water surface of rice paddies for the purpose of nitrogen fixation by symbiotic cyanobacteria. This provides “green fertilizer” for rice production, and biomass other than rice is fed to water birds or fish. In our space agriculture concept, rice, *Azolla*, and loach will be co-cultured in rice paddies in order to greatly enhance productivity under limited available resources. The symbiotic cyanobacteria in *Azolla* quickly fix nitrogen if there is a shortage of nitrogen fertilizer. *Azolla* is an effective green manure, and also suppresses growth of weeds by covering the water surface. The further advantage of co-culturing fish is upgrading biomass to edible food by converting *Azolla* to animal meat. Swimming of fish in rice paddies brings other positive effects on rice production. We also examined the feasibility of adding *Azolla* to the human diet during space habitation, and analyzed the nutritional value of the *Azolla* menu.

Table 1
Nutritional Comparison of Sprouts, Algae, and *Azolla*, composition per 100 g of each food material

Nutrient	Unit/100 g	*Azolla*	Soybean sprout	Alfalfa sprout	Marine macro-alga	
			Glycine max	*Medicago sativa*	*Porphyra* sp.	*Ulva* sp.
Energy	kJ	29	155	50	23	22
Protein	g	1.1	3.7	1.6	1.3	0.9
Potassium	mg	92.8	160	43	167	131
Calcium	mg	34.6	23	14	3.2	20.0
Magnesium	mg	23.7	23	13	12.6	131
Phosphor	mg	22.5	51	37	19.7	6.54
Iron	mg	0.98	0.5	0.5	1.8	0.21
Zinc	mg	0	0.4	0.4	0.1	0.049
Copper	mg	0.04	0.12	0.09	0.01	0.03
Manganese	mg	0.98	0.3	0.1	0.06	0.69
Retinol/β-Carotene equivalent	mg	0.102	0.0	0.056	28.0	2.7
Vitamin C	mg	4	5	5	0.11	0.41
Dietary fiber	g	1.6	2.3	1.4	1.4	1.2

8. Chemical composition of *Azolla* and its cooking methods

Chemical analysis of *Azolla filiculoides*, cultivated on the IRRI medium, indicated 29 kJ, 1.1 g protein, 0.1 g lipid, 0.4 g sugar, 1.6 g dietary fiber, 35.3 mg Na, 22.5 g P, 0.98 mg Fe, 34.6 mg Ca, 92.8 mg K, 23.7 mg Mg, 0.04 mg Cu, trace Zn, 0.98 mg Mn, 0.103 mg V-A, and 4 mg V-C per 100 g of plant body. Nutritional value of *Azolla* is similar to that of alfalfa sprouts, or typical marine macro-algae, as shown in Table 1.

One important factor when designing a menu is whether visual presentation and quantity are acceptable as a meal for ordinary people. It is true that including large quantities of *Azolla* in a human diet requires certain treatment. *Azolla* were cut to leaves and roots. *Azolla* roots taste similar to Alfalfa sprouts, and the leaves resemble moss. However, the smell of *Azolla* might cause a problem of acceptance as food material. However, boiling *Azolla* reduces its smell to an acceptable level. Boiling is also important for storage of *Azolla*. *Azolla* contains 96.4% water, and its taste is crisp. Roots of *Azolla* can be cooked in many different ways, for example fried, sautéd, and baked or added to soup and salad. Leaves of *Azolla* are green or red in color. Red-colored *Azolla* leaves contain anthocyanins, which are known to show anti-oxidative function. This is good for healthy life in space.

9. Model menu and its nutrition

Nutritional evaluation of the three menus is shown in Table 2. Vitamin D, B_{12} and cholesterol, for instance, were not available in the basic vegetarian menu. The sufficiency ratio of amino acids was also found to be insufficient, with only 10% of the required lysine per day. Menu of both “insect and loach” and “insect, loach, and *Azolla*,” has no shortage of essential amino acids. Fat ratio (plant and fish) is evidently out of the recommended range. Other indices, n-6/n-3 fatty acid ratio, and cholesterol are not far from recommended values.

The PFC ratio is 19.3% protein, 16.4% fatty acid, and 64.3% carbohydrate, for the combination of 300 g rice, 120 g soybean, 200 g sweet potato, 300 g Komatsuna, 50 g insect (represented by grasshopper, of which nutritional value including minor component is known), 120 g loach, 100 g *Azolla*, and 3 g sodium salt. This ratio for fat is slightly lower than the recommendation. The ratios of protein and carbohydrate are within the recommended range. The PFC ratio is greatly improved from the basic vegetarian menu by the addition of insect, loach, and *Azolla*.

Both the “insect and loach”, and “insect, loach, and *Azolla*” menus meet human nutritional requirements. To efficiently use biomass energy produced by plants, we decided to use the menu consisting of the basic vegetarian menu plus insect, loach, and *Azolla* for our conceptual design of space agriculture.

10. Space agriculture based on the menu with *Azolla*

We estimated the farming area for our four major plant species will be 200 m^2 per person, considering that solar light intensity on Mars is about half of that on Earth [1,2]. Annual consumption of the four major crops per person is 110 kg rice, 44 kg soybean, 73 kg sweet potato, and 110 kg Komatsuna, postulating that

Table 2
Nutritional evaluation of model menu for space habitation

Nutrient	Unit	Recommendation (2005)/day person	Basic vegetarian	Basic menu + insect, loach	Basic menu + insect, loach, *Azolla*
Energy	kJ	8380	7777	8426	8455
Protein	g	55.0	69.7	96.1	97.2
Potassium	mg	1800	5410	5758	5851
Calcium	mg	800	905	2242	2277
Magnesium	mg	310	680	730	754
Phosphor	mg	1000	1793	2621	2644
Iron	mg	9.0	27.4	34.1	35.1
Zinc	mg	8.0	10.24	13.7	13.7
Copper	mg	0.8	2.5	2.6	2.7
Manganese	mg	4.0	9.7	10.16	11.14
Retinol equivalent	μg	700	785	803	803
Vitamin D	μg	5.0	0	5	5
Vitamin E	mg	9.0	11.7	12.4	12.4
Vitamin K	μg	70	652	653	653
Vitamin B_1	mg	1.0	2.7	2.8	2.8
Vitamin B_2	mg	1.3	0.93	2.2	2.2
Niacin	mg	12	26.1	30.9	30.9
Vitamin B_6	mg	1.3	2.9	3.0	3.0
Vitamin B_{12}	μg	2.4	0	10.2	10.2
Folic acid	μg	240	785	804	804
Pantothenic acid	mg	6.0	8.78	9.58	9.58
Vitamin C	mg	100	175	176	180
Cholesterol (upper limit)	mg	700	0	252	252
Dietary fiber	g	21	39.8	40.1	41.7
Sodium salt (upper limit)	g	9.0	5.9	8.50	8.5
n-3 Fatty acid (lower limit)	g	2.4	2.4	2.5	2.5
n-6 Fatty acid (upper limit)	g	11	13.1	13.2	13.2

multiple cropping is possible. Estimation of planting area required for mulberry trees to rear 18 kg of silkworm pupa is 64 m^2. Maximum production of loach per unit water surface area is 10 $\mathrm{kg/m}^2$, and it takes about 1 year to rear adult loach from the egg stage [10]. In order to produce 44 kg of loach per year, the minimum area for aquaculture is 4.4 m^2. Since loach will be cultured in rice paddies with *Azolla* on their surface, we can manage all these items within the original farming area. The water surface area will be sufficient for *Azolla* as well.

Azolla will be used not only for food material, but also for some other applications. For example, *Azolla* is able to remove heavy metals and antibiotics from waste water. As well, collection of trace elements by *Azolla* may prevent fish diseases caused by lack of those elements.

11. Concluding remarks

We confirmed that our proposed model diet of basic vegetarian foods with insects, loach and *Azolla* fills the nutritional requirements of humans. Based on this menu, we determined the specific requirements for the component system of space agriculture. Use of the agricultural resource on Mars could be optimized by the combination of crop production with chosen species, sericulture, and raising fish. Space agriculture is designed under the severe constraint of resources, with a materials recycle loop as closed as possible. We could apply this knowledge to the support and development of a sustainable civilization on Earth.

References

[1] M. Yamashita, M. Arai, C. Ishiii, Y. Ishikawa, R. Izumi, T. Oshima, H. Oshima, K. Omori, N. Katayama, A. Katayama, S. Kanazawa, T. Kariya, Y. Kawasaki, Y. Kitaya, E. Goto, T. Saito, T. Shimizu, A. Shiraishi, M. Takaoki, H. Takahashi, A. Tani, K. Toki, K. Tomita-Yokotani, A. Nakajima, M. Nagatomo, K. Nitta, H. Hashimoto, M. Hirafuji, Y. Fujii, H. Mizutani, K. Mihara, T. Miyagawa, C. Mukai, S. Mori, S. Yano, N. Yamazaki, H. Yokota, Conceptual study of space agriculture, Space Utilization Research 21 (2005) 323–326.

[2] M. Yamashita, Y. Ishikawa, M. Nagatomo, T. Oshima, H. Wada, Space Agriculture Task Force Space, Agriculture for manned

space exploration on Mars, Journal of Space Technology and Science 21 (2) (2005) 1–10.

[3] M. Yamashita, Y. Ishikawa, Y. Kitaya, E. Goto, M. Arai, H. Hashimoto, K. Tomita-Yokotani, M. Hirafuji, K. Omori, A. Shiraishi, A. Tani, K. Toki, H. Yokota, O. Fujita, An overview of challenges in modeling heat and mass transfer for living on Mars, Annals of the New York Academy of Sciences 1077 (2006) 232–243.

[4] S. Silverstone, Eating, in: From the Field to the Kitchen in Biosphere 2, The Biosphere Press, Oracle, 1993.

[5] T. Masuda, T. Ogasawara, E. Harashima, Y. Tako, K. Nitta, Evaluation and implementation of an advanced life support (ALS) menu for closed ecology experiment facilities (CEEF), Eco-Engineering 17 (2005) 55–60.

[6] T. Koike, A. Miyamoto, The dietary life of the isolated and closed environment, Japanese Journal of Aerospace and Environmental Medicine 42 (2005) 105–119.

[7] J. Mitsuhashi, People Who Eat Insect, Heibon-sha, Tokyo, 1997.

[8] N. Katayama, M. Yamashita, H. Wada, J. Mitsuhashi, Space Agriculture Task Force, Entomophagy as part of a space diet for habitation on Mars, Journal of Space Technology and Science 21 (2) (2005) 27–38.

[9] N. Katayama, Y. Ishikawa, M. Takaoki, M. Yamashita, S. Nakayama, K. Kiguchi, R. Kok, H. Wada, J. Mitsuhashi, Space Agriculture Task Force, Entomophagy: a key to space agriculture, Advances in Space Research online (2007).

[10] M. Kagehira, Loach, in: E. Takashima, M. Murai (Eds.), Aquaculture System 2, Freshwater Fishes, Koseishia-Koseikaku, Tokyo, 2005, pp. 211–225.

[11] E. Shepelev, N. Tkhyok, G.I. Meleshko, A.A. Antonian, T.B. Galkina, Physiological and ecological characteristics of *Azolla pinnata* and its possible use in biological life support systems, Kosmicheskaya Biologiya i Aviakosmicheskaya Meditsina 17 (5) (1983) 66–69.

[12] M. Chen, X.S. Liu, Z.C. Liu, The equipment of using *Azolla* for O_2-supplementation (correction of supplimentation) and its test, Space Medicine & Medical Engineering (Beijing) 13 (1) (2000) 14–18.

[13] IRRI, *Azolla* Utilization, IRRI, Philippines, 1987.

[14] FAO, The "Rice-Azolla-Fish" System, vol.4, RAPA Publication, FAO, Regional Office for Asia and Pacific, Bangkok, 1988.

[15] I. Watanabe, C.C. Liu, Improving nitrogen-fixing systems and integrating them into sustainable rice farming, Plant and Soil 141 (1992) 57–67.

[16] FAO, Bio and Organic Fertilizers: Prospects and Progress in Asia, vol. 10, RAPA Publication, FAO, Regional Office for Asia and Pacific, Bangkok, 1988.

[17] I. Watanabe, Genetic Enhancement and *Azolla* collection—problems in applying *Azolla anabaena* symbiosis, in: N.A. Hegazi, M. Fayez, M. Monib (Eds.), Nitrogen Fixation with Non-Legumes, The American University in Xairo Press, Xairo, 1994.

[18] Ministry of Health, Labour and Welfare, Japan Dietary Reference Intakes for Japanese, Dai-Ichi Shuppan, Tokyo, 2005.

[19] Science and Technology Agency, Resource Examination Committee, Fifth Japan Food Composition Tables, Tokyo, 2000.

[20] C. Drummer, P. Norsk, M. Heer, Water and sodium balance in space, American Journal of Kidney Diseases 38 (2001) 684–690.