Physical, chemical and biological characteristics of space flown tomato (*Lycopersicum esculentum*) seeds

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Abstract. Several research showed that space flown treated seeds had a different characteristic with that of ground treated seed, which eventually produced a different characteristic of growth and productivity. Research was conducted to study the physical, chemical and biological properties, such as the rate of germination and the growth of tomato (*Lycopersicum esculentum*) space flown seeds compared with that of control one. Observations of physical properties using a SEM showed that there were pores on the surface of some tomato space flown seeds. Observations using a stereo and inverted microscope showed that the coat layer of space flown seeds was thinner than control seeds. The total mineral content in the control seeds (22.88%) was averagely higher than space flown seeds (18.66%), but the average carbohydrate content in control seed was lower (15.2 ± 2.79 %) than the space flown seeds (9.02 ± 1.87 %). The level of auxin (IAA) of control seeds (142 ± 6.88 ppm) was averagely lower than the space flown seeds (414 ± 78.84 ppm), whereas the level of cytokinins (zeatin) for the control seeds (381 ± 68.86 ppm) was higher than the space flown seeds (68 ± 9.53 ppm), and the level of gibberellin (GA3) for the control seeds (335 ± 10.7 ppm) was higher than the space flown seeds (184 ± 7.4 ppm). The results of this study showed that the physical and chemical properties of tomato space flown seeds were generally different compare with that to control seeds, so that it might also be resulted in different germination and growth characteristic. The germination test showed that space flown seeds had lower germination rate compare to control. The growth pattern indicated that planted space flown seeds generally grew better than control. However, those data were more homogenous in control seeds compare to that in space flown tomato seeds.

1. Introduction

Tomato (*Lycopersicum esculentum*) is a member of Solanaceae which has high nutrient content in addition to its high economic value. The average of tomato production in Indonesia in 2000 was 80 kg/ha. This number is considered low compare to the potential of the production, 150 kg/ha [1]. Productivity can be improved by selecting improved crops. One of the methods used in producing improved crops is radiation exposure [2]. Radiation can cause genetic variation in the seed. This variation can be used as an approach to produce improved crops [3]. Experiments in China showed that space flown seeds have produced new kinds of rice, wheat and vegetables which are more productive, larger, and more nutritious than their earthly treatment [4]. Recent publication also noted that space flown seed program in China also produced cucumbers and tomatoes which are oversize with remarkably high sugar content as a result of cosmic radiation and microgravity. However, similar
research in the US showed non comparable results. Other research in Alfalfa showed that the rate of germination of the space-flight seed and the growth of the seedlings of Alfalfa was higher compare to control on Earth [5].

The purpose of this study was to compare the rate of germination, growth, and productivity of space-flight tomato seeds to the control seeds on Earth, in addition to the evaluation on physical and chemical properties of the seeds.

2. Methods
The tomato seeds (Lycopersicum esculentum cv. Arthaloka) used in this experiment had been sent to the International Space Station (ISS) by Japanese Exploration Agency (JAXA) in 2010 and stored in Japanese Experiment Module (JEM) which also known as Kibo (hope) for three months before it was sent back to Indonesia.

2.1. Physical observation.
The Scanning Electron Microscope (SEM) imaging was done in Pusat Penelitian dan Pengembangan Geologi Kelautan (PPGL) Bandung. The sample seeds were coated by palladium-gold (PdAu) in Fine Coat Sputter JFC-1100. The image was observed with SEM JEOL JSM 6063LS and the image was captured by Back Electronic Scatter (BES) 10 kV. The seeds were also observed by stereo microscope after they were soaked in water.

2.2. Chemical content of the seeds
2.2.1. Mineral content measurement. The mineral content was measured by Energy Dispersive x-Ray Spectroscopy (EDS) in PPGL Bandung.

2.2.2. Carbohydrate content. Carbohydrate content of the seeds was measured using Luff Schroll method [6]. The known volume was converted to mass based on Luff Scroll table, the carbohydrate content was then calculated using the equation (1) below.

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\text{Carbohydrate content} = \frac{\text{mass of glucose} \times \text{dilution factor} \times 0.95}{\text{mass of sample}}
\]

2.2.3. Auxin, Cytokinin and Gibberellin contents. 1 gram of seed sample was extracted in 25 ml of 80% methanol. The methanol was evaporated in room temperature. The pH of the extract was then lowered to 2.5 before the addition of 5 ml ethyl acetate. The water and ethyl acetate phase were separated. The water phase was evaporated. The dried sample was added by 1 ml methanol before it was analysed [7,8]. The sample was analysed in high performance liquid chromatography (HPLC). The eluent was methanol and water (7:3). The flow rate was 0.8 ml/minute. The standard used for auxin was IAA, zeatin for cytokinin and GA3 for gibberellin.

2.3. Germination rate in soil
The germination test was conducted by germinating the seeds in the pots (5 cm x 10 cm). The soil surrounding the seeds was kept moist until the seeds were fully germinated.

2.4. Plant growth and productivity
The seedlings with 3-4 leaves were transferred to polybags. The seedlings were watered regularly up to 14 weeks. For maintenance, the plants were fertilized and treated with fungicide. Every week the height of the plants was measured.

3. Results and discussion
The image from SEM observation showed some pores (see arrow in figure 1) were formed on the surface of space-flight seed, while that of control seed had no pore. This observation was supported by the
research on Alfalfa seeds which showed pores formed after cosmic radiation during the space flight [5]. Tomato seeds exposed to gamma ray radiation would have morphological damage, such as pore formation on the seed coat surface [9]. The gamma ray would interact with the water molecule in the cells and formed free radical. The energy released during the free radical formation would go around the seed surface and induce cell damage. Another research on space-flight tomato seeds showed similar result [10]. The pore produced due to radiation would cause water and air enters the seeds faster during germination and could speed up the germination process.

![Figure 1. (A). Control seed had no pore. (B). Space-flight seed had a pore (arrow) in the seed coat.](image)

Visual observation on the seed surface of space-flight seeds showed thinner seed coat compare to control (figure 2). Other research on rice seeds showed that cosmic radiation could change the morphology of the seeds [11]. The changes varied based on the dose and duration of gamma ray exposure.

![Figure 2. Control seed (A) has thinner seed coat (s) compare to space flight seed (B).](image)

The total mineral content of the space flight seed was also generally lower (18.66%) than control seed (22.88%). This result was similar to Alfalfa seeds flown to the space, which were then resulted in softer surface than control seed [5] even though the cause has yet to be found. This result might also be supported by the calcium content of the space flight seed, which had lower calcium content (6.27%) compare to control seed (7.44%). The carbohydrate content of the space flight seed (9.02±1.87%) was lower than the control seed (15.2±2.79%). The result was similar to the result of the research on the hypocotyl of the Arabidopsis, which showed that space treated hypocotyl contained less cellulose and xyloglucan [12]. The cytokinin and gibberellin content of the space flight seeds were lower, but the auxin content was higher than control (table 1). Low cytokinin concentration could prevent mitosis [13]. Lower gibberellin content would slow down the germination process in the space flight seeds, while high concentration of auxin in the seeds would promote plant growth [4].

| Hormone         | Control (ppm)     | Space flight seed (ppm) |
|-----------------|-------------------|-------------------------|
| Auxin           | 142.055± 6.88     | 414.445 ± 78.84         |
| Cytokinin       | 381.605± 68.86513 | 68.885 ± 9.53887        |
| Gibberellin     | 335.01 ± 10.7     | 184.465 ± 7.4           |

The germination test showed that space seeds had lower rate of germination (73%) compare to control (83%). The pore on the seed coat could probably increase the water imbibition into the space flight seeds, but the low level of gibberellin might slow the process of germination. This result was supported by another research on Arabidopsis seeds [15], which stated that radiation disturbed the radicle and root formation in the seedlings.
The observation of the growth rate was presented in figure 3. After the fifth week, there was no significant difference between the space flight plants compare to the control. The higher growth rate during the sixth to the tenth week in space flight plant was probably caused by the higher auxin concentration in the seeds, which then recovered to the same growth rate after plant could produce its own auxin.

4. Conclusion
The physical characteristic of the space flight seeds support water imbibition but chemical content in the seed slowed the germination rate. The plant growth from the space flight seed was initially better than control, but at the end there was not significantly different.

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