

Asian Journal of Agricultural and Horticultural Research

8(3): 37-50, 2021; Article no.AJAHR.72616 ISSN: 2581-4478

## Effect of Organic Fertilizer on Tomato Growth and Production under Soil-less System

Kiyotaka Murakami<sup>1</sup>, André Freire Cruz<sup>2\*</sup>, Mateus De Freitas Ramos<sup>2</sup>, Osvaldo Kiyoshi Yamanishi<sup>3</sup> and Shuichi Date<sup>2</sup>

<sup>1</sup>Taiga Co. Japan, Shinagawa-ku, Tokyo, Japan. <sup>2</sup>Kyoto Prefectural University, Graduate School of Life and Environmental Sciences, Kyoto, Japan. <sup>3</sup>Universidade de Brasília, Faculdade de Agronomia e Veterinária, Brasília, DF, Brazil.

### Authors' contributions

This work was carried out in collaboration among all authors. Author KM Conceptualization. Data acquisition author s AFC and MDFR, Data analysis author AFC Design of methodology authors OKY and SD Writing and editing, authors KM, AFC and MDFR All authors read and approved the final manuscript.

### Article Information

DOI: 10.9734/AJAHR/2021/v8i330119 <u>Editor(s):</u> (1) Dr. Ahmed Medhat Mohamed Al-Naggar, Cairo University, Egypt. <u>Reviewers:</u> (1) Abdel-monnem Sadalaha Kahlel, Northern Technical University, Iraq. (2) Nasim Ahmad Yasin, University of the Punjab, Pakistan. Complete Peer review History: <u>https://www.sdiarticle4.com/review-history/72616</u>

Original Research Article

Received 24 June 2021 Accepted 28 August 2021 Published 31 August 2021

### ABSTRACT

Soil-less system constitutes an efficient approach for the cultivation of tomato; however, organic liquid amendments are very limited under such systems. The current experiment aimed to evaluate the effects of Kurojiru (K), an organic liquid fertilizer and fulvic acid (FA) on the growth and production of tomato, *Solanum lycopersicum* L. cv. 'Momotaro'. Recently, it is desirable to reduce the environmental impact and fertilizer cost by lowering the concentration of inorganic components in the culture medium. Therefore, we explored the effect of adding these organic fertilizers on the yield and quality of tomatoes by gradually reducing the phosphoric acid concentration in the culture medium (87, 58 and 29 mmol • pot<sup>-1</sup> • week<sup>-1</sup>). The whole experiment was conducted for 20 weeks (from seeding until harvesting). The plant biomass, tomato fruit weight and chlorophyll content were measured. The fresh weight (FW) of both root and shoot indicated a progress response according to phosphorus concentration in liquid media, in FA treatments. Total fresh weight was significantly higher in the treatment with K+FA than in the control. Additionally, the yield responded to the all treatments within the 58 mmol P. Especially, in this level of P the relative fruit weight was

higher only under K application. The chlorophyll content responded K+FA treatment under a low P content (29 mmol), and to all of those in 58 mmol. These results indicate that Kurojiru has some positive effects on tomato growth in soil-less systems.

Keywords: Chlorophyll; fulvic acid; phosphorus; soil-less.

### **1. INTRODUCTION**

The system of growing crops without soil has been shown to date back to the 18th century and is believed to have existed even prior to that period. Hydroponics is a set of technologies for growing plants in nutrient solutions (water and fertilizers) with or without the use of artificial medium (e.g., sand, gravel, vermiculite, rock wool, peat, coir, sawdust) to provide mechanical support (Benton Jones, 2005).

The cost of fertilizers for the nutrient solution is also a major issue. Plant nutritional status affects yield, fruit quality and susceptibility to pathogens. Hence, it is essential to have a good knowledge of the plant's mineral requirements in order to ensure a good yield and avoid nutrient wastage [1]. In addition, not just any fertilizer can be used in hydroponics. Conventional hydroponic systems cannot use organic fertilizer, which plant growth: compounds inhibits organic contained in hydroponic solutions have been regarded as phytotoxic [2] [3]. However, from the viewpoint of resource recycling, it is important to develop methods capable of using organic fertilizer sources in hydroponics [4] [5]. A big difference between organic and inorganic fertilizers is the form in which nitrogen appears, whose process goes from organic molecules to ammonium to nitrate, thanks to the activity of microorganisms in the system [6].

The direct use of organic fertilizer in hydroponics has proved to be deleterious to plant growth [2] [7]. Therefore, organic fertilizer has been microbial pre-processed before incorporation into hydroponic solutions [2] [8]. In particular, in the case of tomatoes, since reproductive and growth proceed simultaneously, it is important to clarify the effect of phosphoric acid fertilization when designing the optimum level of fertilizer based to reach high productivity [9]. A novel and practical hydroponic culture method that uses microorganisms to degrade organic fertilizer in the hydroponic solution has been developed, microorganisms mineralize organic whose nitrogen via ammonification and nitrification [4].

Therefore, the establishment of cultivation techniques to reduce the nitrate present in leafy greens in hydroponics is required. Additionally, the application of organic fertilizer instead of chemical fertilizer could reduce nitrate in honeybees in Japanese Hornwort cultivation [10]. However, not all organic fertilizers can be used for hydroponics. This is because hydroponic fertilizers require soluble elements, while common organic fertilizers leave many elements dissolved in the culture medium, and these cannot be absorbed by the plants. Liquid organic fertilizers derived from agricultural and industrial residues are available slowly as they are converted to soluble form by microorganisms. Previously, the waste-product bio-slurry as a nutrient source has been used in hydroponics [11]. For the reduction of chemical fertilizers and wider use of liquid organic fertilizers in hydroponics, it will be essential to establish a method of combining them with effective microorganisms [12].

The product Kurojiru has been applied to soils in Japan and succeeded to vegetable growth for most of the cases; it contains the aqueous phase of compost, i.e. the hydrophilic bacteria and some nutrients. This experiment is important because it is the most used organic fertilizer in Japan but there is no previous research, except for the application of such product as an alternative treatment for post-harvest storage of Papaya, where they could reduce the disease incidence [13]. Perhaps active bacteria contained in Kurojiru could function as biocontrol agent for this disease. Although it is a completely safe fertilizer, it is not realistic to export liquids from Japan, due to phytosanitary policies from other countries, but the manufacturers are considering ways to solve this problem. Also, the Fulvic acid can contribute on P solubilization in soils. Thus, it is possible to hypothesize the effects on soil-less system with decreased amount of P as using of effective microorganisms in such system could lead a more sustainable management. This study was conducted to evaluate the effect of different concentrations of P in the nutrient solution on the vield of tomatoes grown hydroponically using the Kurojiru and Fulvic acid [14-15].

### 2. MATERIALS AND METHODS

The experiment was conducted inside a greenhouse belonging to the Kyoto Prefectural University, in Kyoto, Japan. The climate is Cfa, according to the Köppen and Geiger classification.

Tomato (Solanum lycopersicum) seeds from the 'Momotaro' cultivar, a very common type grown under soil-less system, characterized by their high productivity, good quality and tolerance to diseases, was sown in polystyrene trays with commercial substrate Nippi Engei Baichi (Nihonhiryo Co., Japan). Thirty days after sowing, the seedlings were removed from the trays; the roots were detached from the substrate and washed. Then the seedlings were transplanted into plastic net pods, which were put into a big nutrient container with aerators in hydroponic Deep Water Culture System for acclimation to the greenhouse conditions. After another 3 weeks, plants were once again transplanted into individual pots; they were put into polystyrene rafts and in 28 L plastic boxes containing 20 L of nutrient solution and aerators for the start of the experiment. Two experiments were conducted, the first from March 15 to August 5 and the second from September 5 to February 28.

The experimental design was prepared with 48 tomato seedlings of the same size selected and distributed into 12 treatments. Each one consisted of three levels of phosphorus amendments (29, 58 and 87 mmol· pot<sup>-1</sup>· week <sup>1</sup>), the highest concentration is equivalent to one third of optimum level of P to reach a good productivity according to previous data. The treatments were distributed as following: 1) Kurojiru (K) - An organic liquid fertilizer (Yasaki Co. Japan), whose characteristics are described in Table 1; 2) Fulvic acid (FA), supplied by Yasaki Co, Japan, but not yet a commercial product; 3) a combination of the aforementioned Kurojiru and Fulvic acid substances (K+FA); and 4) a control group, where the only fertilizers were applied. The pots were amended with the fertilizers, K (5 ml) and FA (10 ml) once a week and their content were kept at 20 L of solution by completing with micronutrients. All essential nutrients (N, P, K, Ca, Mg, S, micronutrients) were applied to the plants, where the variation occurred only for P. From seedling until harvesting, the whole experiment was conducted for 20 weeks.

Three days before harvesting the chlorophyll content (SPAD meter) was measured at the second leaf. At harvesting time, the fruit weight and total plant biomass (root and shoot fresh weight) were measured too. A leaf sample from third one was sampled, drought 60 °C for 3 days, grained and stored under closed conditions until the measurement of P (PO<sub>4</sub><sup>-</sup>) content. The fruits from the second panicle were also sampled for further fruit quality analysis (Soluble solids – Brix, Titratable acidity). The phosphorus concentration was measured in the shoot by ICP spectrometry of acid wet digestion using Nitric acid and Hydrogen peroxide.

Fruits from the 1<sup>st</sup> to 4<sup>th</sup> clusters were harvested at ripen time, followed by weighting individually and assigned to fruit quality tests (henceforth referred to as "individual fruit weight"). Fruits from the 5<sup>th</sup> cluster onwards were harvested still unripe at the end of the experiment, and weighed as a collective for each treatment (henceforth referred to as "remaining fruit weight").

The whole experiment was conducted twice and only one representative experiment is shown. The statistical analysis was done with the SPSS program, where the standard error, Tukey tests and ANOVA were used to compare the results. Before the ANOVA the Shapiro-Wilk test was applied to the samples and the data of Fresh weight (root and shoot), Soluble Solids and Titratable acidity was transformed in order to reach the normality.

### 3. RESULTS AND DISCUSSION

According to the ANOVA table the treatments had significant effects for the Root fresh weight, Average fruit weight, and Titratable acidity. As stated in such table, the P concentration influenced on all the parameters, except for Remaining fruit weight, and soluble solids. Furthermore, the effects of interaction between P and treatments were detected for Average Fruit Weight (Table 2).

For the total fresh weight (Shoot and Root) or plant biomass, all treatments effectively lead to higher biomass in the treatments of 58 mmol and 87 mmol phosphorus levels, but showed no effect at the 29 mmol level (Fig 1).

The tomato plant biomass indicated some responses of the treatments as compared to the control at the 58mmol of P, but the accumulated progress of this parameter according to P level was observed only in FA treatments. This

suggests that the response of plant growth to Kurojiru can be achieved only with 58mmol of P applied to the solution. Too much and/or low P might not induce the effects of this organic fertilizer. It contains N, P, and K together with some hydrophilic bacteria, which are in charge of nutrient cycling and disease control [13]. The effects of Kurojiru were not as high as expected; thus, its effects as a P supplier are strict limited, and the action of these bacteria is necessary to solubilize P. The use of organic fertility is a big challenge in soil-less systems, and a comparison between organic and conventional inorganic fertilizers treatments in `Rex` lettuce indicated that Bombardier, Espartan and Caos fertilizers were more effective [6]. The organic fertilizers contain some N in the form of complex compounds, such as proteins, that need to be broken down before plant absorption, which is usually performed by microorganisms.

Analisys	Element	Unit	
	total N	%	0.04
	total $P(P_2O_5)$	%	0.04
	total K(K <sub>2</sub> O)	%	0.07
	Са	%	0.02
	Mg	%	0.01
	S	%	0.04
	В	%	<0.01
al	Zn	%	<0.01
nic	Cu	%	<0.01
Cher	Mn	%	<0.01
	Fe	%	0.02
	Ni	mg/kg	12.0
	Cd	mg/kg	7.0
	Cr	mg/kg	1.0
	Pb	mg/kg	<0.1
	рН		8.71
Microb.	Humidity	%	99.91
	Organig C	%	<0.01
	Helmint eggs	Eggs/4q TS	<1
	Coliforms	MPN/100mll	<1.8
	Humidity(65°C)	%0	70.41
	Bacillus subtilis	CFU/g	1.4x 10'
	Acinetobacter modestus	CFU/g	2.2 x10"
	Methylobacterium adhaesivum	CFU/g	1.8 x 10

Table 1.	Elemental	composition	of Kurojiru	product
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TS – Total Solids; NMP – Most probable number; CFU – Colonies per unit

# Table 2. Two-way ANOVA showing the effects of phosphorus concentration and treatments (Kurojiru and Fulvic acid) on tomato plants under soil-less system

	Treatments		Phosphorus		Treat. x Phosp.	
	F-Value	p-Value	F-Value	p-Value	F-Value	p-Value
Shoot Fresh Weight	0.503	0.684	10.430	0.000	0.822	0.563
Root Fresh Weight	2.489	0.083	4.589	0.020	0.588	0.737
Remaining Fruit Weight	0.372	0.774	1.844	0.178	0.683	0.665
Relative Fruit Weight	1.810	0.170	5.810	0.008	0.806	0.574
(Remaining)						
Total Fruit Weight	0.680	0.572	6.817	0.004	0.452	0.837
Average Fruit Weight	7.892	0.001	13.138	0.000	2.498	0.048
(Total)						
Chlorophyll Content	1.354	0.279	3.690	0.039	1.167	0.354
Soluble Solids	2.187	0.114	1.133	0.337	0.934	0.488
Titratable Acidity	5.263	0.006	6.557	0.005	0.756	0.611
Phosphorus Content	0.129	0.942	90.820	0.000	0.692	0.658

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Fig. 1. Influence of phosphorus level, Kurojiru (K) and Fuvic Acid (FA) treatments on biomass production (Shoot Fresh Weight – SFW; and Root Fresh Weight-RFW) of soil-less tomato plant. Bars represent standard errors. \* - The differences of the treatment as compared to the control one is significant in each P concentration by the Tukey test

Individual fruit weights from the 1<sup>st</sup> to the 4<sup>th</sup> cluster were impacted by the 58 and 87 mmol phosphorus levels for all treatments, except for Kurojiru at the 87 mmol level. There was no difference at the 29 mmol level (Fig 2). The remaining fruit weight from the 5<sup>th</sup> cluster onwards were significantly higher with treatments of the 87 mmol phosphorus level, for the fulvic acid and K+FA treatments (Fig 3). There are possibilities of interaction between fruit weight and P amount applied to the solution.

Fruit production, as indicated by the weight in clusters 1-4 and from the 5<sup>th</sup> on indicated some response of P in all treatments, and at each level of this element the positive effects of Fulvic Acid and K+FA were found in 58 and 87 mmol of P. However, for the Kurojiru individually, the maximum was reached at 58 mmol of P. Perhaps, in terms of fruit production, the effects of P can be enhanced by the little supplying of nutrients from Kurojiru and FA, although a dissimilar result was found for plant biomass. Since the kurojiru contains the liquid phase of the compost, the small amount of nutrients, especially N, P and K could partially contribute for tomato growth under soil-less system. Moreover, the bacterial metabolites could assist the plants in terms of growth and resistance to diseases. When organic fertilizers are applied, the plants allocate more nutrients to fruit production in instead of growth [16]. The use of liquid fertilizers from agricultural and industrial wastes has been well applied in soil-less systems for Green Cos Lettuce, where fertilizers from sugarcane leaves and distillery slop provided the best response, sometimes comparable to inorganic fertilizers [16]. Usually, organic fertilizers are insoluble in water, and their nutrients are slowly released to plants through soluble compounds converted hv microorganisms. Sometimes the organic fertilizers contain plant-growth-promoting rhizobacteria (PGPR), which function well in both soil and hydroponic systems [17].

Chlorophyll content was higher in all treatments at the 29 mmol and 58 mmol levels, but not at the 87 mmol phosphorus level (Fig 4). The soluble solids indicated a non-change in their values among the treatments, except for the one at 89mmol P, where those with Kurojiru and control were higher than the others (Fig 5). The titratable acidity indicated an increasing in relation to P level, except for the Kurojiru treatment. Additionally, the Fulvic acid and Kurojiru lead to higher values with the 29 mmol

P. At the highest level of P the Fulvic acid treatment lead to higher Titratable acidity as compared to the other ones (Fig 6).

As indicator of plant health, the chlorophyll content showed an effect of treatments only at low and middle levels, which generally contradicts the effects of plant biomass and fruit production. However, similar patterns were found at the middle levels of P. The synchronism among these effects might occur at the middle levels of P; thus, according to these results the best P level that can guarantee the effects of the treatments, especially the Kurojiru, could be the 58 mmol. Usually the chlorophyll content is related to the N in plants [18] [19], and thus the response to this element in the solution would be more limited than the P. In addition, the N concentration and Electrical conductivity in organic fertilizers must be considered when analyzing the chlorophyll content [20].

The P content in leaves showed a progress curve according to increase in P concentration in solution, but a slight higher value was observed in the other treatments compared to the control only with 58mmol P (Fig 7). This higher P content in leaves by the treatments only at the middle level might be due to the microorganisms contained in the Kurojiru also need P for their survival, and they may compete with the plant for the solution nutrients and at the same time receive photo-assimilates from the plant. Thus, at the minimum level, the P uptake is not different among the treatments, and at the highest one, the plants would not need the microorganisms in the Kurojiru for this element. We believe that the middle level would be the ideal one to have effects arising from organic fertilizer amendment. However, this pattern was not detected for the fruit quality evaluation, where the best results for the treatments were found at the low level, so perhaps the efficiency of P in plants for fruit production might be better at the low levels, especially those with Kurojiru and Fulvic acid individually. Sometimes the presence of Nitrogen fixers and phosphate solubilizing agents in the fertilizers causes a remarkable effect on the soilless system, especially with a deficiency of these elements [18]. An organic fertilizer made of sugarcane residues applied to a soil-less system contains many phosphate-dissolving microorganisms, which are crucial in mineralizing Ρ in both soils and water [21]. These microorganisms are very important for plant growth too, since they help nutrient cycling in the water [22].

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Fig 2. Influence of Phosphorus level, Kurojiru (K) and Fuvic Acid (FA) treatments on the Total Fruit Weight (1st – 4th panicle g/plant) and the Relative Fruit Weight (g/fruit) of soil-less tomato plant. Bars represent standard errors. \* - The differences of the treatment as compared to the control one is significant in each P concentration by the Tukey test

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Fig. 3. Influence of phosphorus level, Kurojiru (K) and Fuvic Acid (FA) treatments on the Remaining Fruit Weight (After 5th cluster – g/plant) and the Relative Fruit Weight (g/fruit) of soil-less tomato plant. Bars represent standard errors. \* - The differences of the treatment as compared to the control one is significant in each P concentration by the Tukey test



Fig. 4. Influence of phosphorus level, Kurojiru (K) and Fuvic Acid (FA) treatments on the Chlorophyll Content of tomato cultivated under soil-less system. Bars represent standard errors. \* - The differences of the treatment as compared to the control one is significant in each P concentration by the Tukey test

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Fig. 5. Influence of phosphorus level, Kurojiru (K) and Fuvic Acid (FA) treatments on the Soluble Solids concentration of tomato fruits cultivated under soil-less system. Bars represent standard errors. \* - The differences of the treatment as compared to the control one is significant in each P concentration by the Tukey test



Fig. 6. Influence of phosphorus level, Kurojiru (K) and Fuvic Acid (FA) treatments on the Titrable Acidity of tomato fruits cultivated under soil-less system. Bars represent standard errors. \* - The differences of the treatment as compared to the control one is significant in each P concentration by the Tukey test



Fig. 7. Influence of phosphorus level, Kurojiru (K) and Fuvic Acid (FA) treatments on Phosphorus Content of tomato plant cultivated under soilless system. Bars represent standard errors. \* - The differences of the treatment as compared to the control one is significant in each P concentration by the Tukey test.annex

Although some effects of kurojiru were detected in the middle level of P, their mechanisms could not be explained in this research, maybe due to the presence of chemical fertilizers in the solution that could suppress the bacterial activities. Further researches are necessary to elucidate the kurojiru activities under soil-less systems, especially targeting on their bacterial population that could assist on N and P cycling to make the nutrient available to the plants. This contribution would be additional effect to the nutrient supplying by the liquid fertilizer.

### 4. CONCLUSIONS

- The effect of K and FA in three P levels was evaluated in tomato plants under soil-less system.
- The plant biomass was affected by K+FA treatments as compared to the control ones.
- The tomato yield suffered effect of both K and FA within the 58 mmol P.
- The Kurojiru has some positive effects on tomato growth in soil-less systems under the middle level of P.

### DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

### ACKNOWLEDGEMENTS

This research was supported by Taiga Co. (Japan) and Quimiway Co. (Brazil). Thanks to students and researchers from Kyoto Prefectural University, Laboratory of Pomology and University of the Philippines – Los Banos for their assistance during the experiment. The authors are also grateful to Prof. Dr. Go Takeba and Dr. Masafumi Ueda, Kyoto Prefectural University, for allowing to use their facilities to plant mineral nutrition and chlorophyll analysis, respectively.

### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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Peer-review history: The peer review history for this paper can be accessed here: https://www.sdiarticle4.com/review-history/72616