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EFFECTS OF SILVER NANOPARTICLES ON GROWTH PARAMETERS OF RADISH (*Raphanus sativus* L. var. *radicula*). GROWN UNDER DEFICIT IRRIGATION

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Abstract
In agricultural production, fertilization has become mandatory for improving the soil and increasing the yield of the product. Especially in the production of horticultural crops, the need for precise and detailed fertilization management complicates the production. Intensive long-term use of conventional fertilizers causes serious environmental problems such as groundwater pollution, deterioration of soil quality and air pollution. This has led to the search for particularly effective and environmentally friendly fertilizers. Recently, nano-fertilizers are considered as a promising fertilization alternative. Nano-fertilizers benefit nutrition management due to their strong potential to increase nutrient utilization efficiency. The present study aimed to determine the effect of silver nanoparticles (AgNPs) on some growth parameters of radish grown under deficit irrigation conditions. For this purpose, in this study, four different irrigation water levels (I0; 100% full irrigation, I1; 20% deficit irrigation, I2; 40% deficit irrigation and I3; 60% deficit irrigation) and four different nanosilver doses (Ag0: 0 ppm, control; Ag1: 20 ppm; Ag2: 40 ppm and Ag3: 80 ppm) were applied. The study was carried out in a total of 48 pots in three replications according to the randomized plots experimental design under greenhouse conditions. The findings showed that root length, root diameter, root fresh weight and root dry weight significantly (p<0.01) decreased in deficit irrigation. While root parameters were significantly increased in Ag nanoparticle applications, the number of leaves were not varied statistically. The highest root height (33.21 mm) was determined in full irrigation application with Ag3 (80 ppm). As a result, it can be stated that radish plant growth in silver nanoparticles can be significantly improved under deficit irrigation conditions compared to control application non-silver nanoparticles.

Keywords: Drought stress, Nano-fertilizer, Radish, Silver nanoparticles.

1. INTRODUCTION

Agriculture is a multitrillion dollar industry worldwide related to the production, special horticultural, feed, food, and ornamental purposes. In agricultural production, nutrient fertilization is essential in soil fertility and improving crop productivity (Malhotra, 2016). It is estimated that the world population will reach approximately 11.0 billion by 2100 (Adam, 2021). Therefore, current food production should be increased by 1.5 times to meet the nutritional needs of the growing population (van Dijk et al., 2021). Limited resources (limited arable lands and water resources), ever-increasing demand for food and the rapidly-increasing human population has forced agricultural producer to increase fertilizer use (Zhang et al., 2015). However, conventional fertilizers use for long periods cause serious environmental problems such as air (toxic chemicals or

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gases like NH4, CO2, CH4, etc.) soil and water pollution, water eutrophication (Xiang et al., 2008; Congreves and Van, 2015). For example, because of without efficient fertilizer use, nitrogen is lost heavily through volatilization, leaching, and runoff. This heavy loss of fertilizer is revealed some of environmental problems (Xiang et al., 2008). New approaches are required that will protect natural resources and eliminate negative environmental effects (Babu et al., 2020). With new approaches that can increase farm production while protecting the environment, agricultural production can maintain its sustainability (Arora, 2018; Shang et al., 2019). This has led to the search for particularly effective and environmentally friendly applications (Digital agriculture solutions, Eco-efficient agriculture) (Kayad et al., 2021). Recently, nanofertilizers are considered as a promising fertilization alternative. Nanotechnology helps to improve agricultural production by increasing the efficiency of inputs and minimizing input losses. Nanofertilizers produced with nanotechnology improve nutrition management due to their strong potential to increase nutrient utilization efficiency (Shang et al., 2019).
Nanofertilizers regulate the availability of nutrients taken into plants, thanks to their release mechanism (Chen and Wei, 2018). Since nanofertilizers are in small quantities, transportation and application costs are less than conventional fertilizers (Fan, 2014). Because they are used in low doses, they prevent soil salinization in the short or long term caused by over-application with conventional fertilizers (Leon-Silva et al., 2018). Another advantage of using nanofertilizers is that they can be synthesized according to the nutritional requirements of the produced crop (Kah et al., 2018). Previous studies have shown that the effects of silver nanoparticles (AgNPs) against germination and growth differ according to plant species (Almutairi and Alharbi, 2015). Moreover, small concentrations of silver nanoparticles (AgNPs) can have a stimulating effect, while high concentrations of AgNPs can have a toxic effect on plant growth (Salama, 2012). Therefore, the present study aimed to determine the effect of different doses of AgNPs on some growth parameters of radishes grown under scarce irrigation conditions.

2. MATERIALS AND METHODS

Plant material and growth conditions

The AgNPs were obtained from Nanografi Nano Technology commercial firm. The particle size ranged from 28-48 nm and it have 99.9% purity. The plant material used was radish (*Raphanus sativus* L. var. *radicula*). The seeds of radish were obtained from the AGR commercial firm, Konya/Turkey. For the study, soils with loamy texture were placed in 4-liter pots. Firstly, 8 seeds were planted in each pot, and then thinning was carried out to leave 4 plants. The study was carried out in the experimental area of Van Yüzüncü Yıl University, Faculty of Agriculture, under greenhouse conditions.

Experimental design

This study was carried out by applying four irrigation levels (I0: 100% full irrigation, I1: 20% deficit irrigation, I2: 40% deficit irrigation, I3: %60 deficit irrigation) and 4 silver nanoparticle doses (Ag0: 0 ppm, control, Ag1: 20 ppm, Ag2: 40 ppm, Ag3: 80 ppm) with three replicates in total of 48 pots as randomized plot design (Table 1). After emergence of true leaves, 100 ml of AgNPs at different concentrations (0, 20, 40 and 80 ppm) were given 2 times for each test plants. Moreover, Hoagland solution (50 ml) was applied to all pots in growth period. When the radish plant has 4-5 true leaves, deficit irrigation applications are started. Before the planned irrigations, water was

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applied to all pots up to the field capacity (pot capacity). The amount of irrigation water applied in each irrigation was calculated with the following equation.

$I = (W_{i-1} - W_i) \times IR$
where I is the irrigation water amount (ml), W_{i-1} and W_i are weights (kg) of the pot at three-four day i-1 and i, respectively. The plants were hand-irrigated with tap water (Kadayıfci et al., 2005; Ekinci et al., 2015).

Table 1. Irrigation and AgNPs application

AgNPs	Doses (ppm)
Ag0	0
Ag1	20
Ag2	40
Ag3	80
Irrigation	Deficit (%)
I0	0 (Full irrigation, control)
I1	20
I2	40
I3	60

Root Measurements
End of the experiment, root length and diameter were measured in mm with a caliper. The leaves of all plants were counted (number of leaves per plant) and subjected to statistical analysis. Root fresh weight were determined. Root dry weight was measured after it was kept in an oven at 65 °C for 48 hours.

Soil EC and pH
After the experiment, in the saturation extract pH of the soil was determined with a pH meter, and the electrical conductivity (EC) was determined using a conductivity meter (Rhoades, 1982).

Statical Analysis
All multivariate analyses were performed with IBM SPSS Statistics version 21.0. ANOVA was performed for data comparison, and the mean values of experimental treatments were compared by Duncan's multiple comparison test (Duncan, 1955).

3. RESULTS AND DISCUSSIONS
Changes in root length according to deficit irrigation water and AGNP applications are given in Figure 1. The effect of deficit irrigation water and AgNP applications on radish length was found to be significant ($p<0.01$). As expected, water stress caused to decreasing plant length significantly and AgNPs applications applied in increasing doses had shown variable results in plant development. Increasing doses of AgNPs led to an increase in plant length but interaction between applications is not significant. The highest root length value (33.21 mm) was detected in Ag3 and I0 application. The lowest root length value (19.59 mm) was Ag0 and I3 application. When the means are examined, it is seen that the highest result is in Ag2 and Ag3 AgNPs applications according to the without AgNPs (Ag0) (Figure 1).

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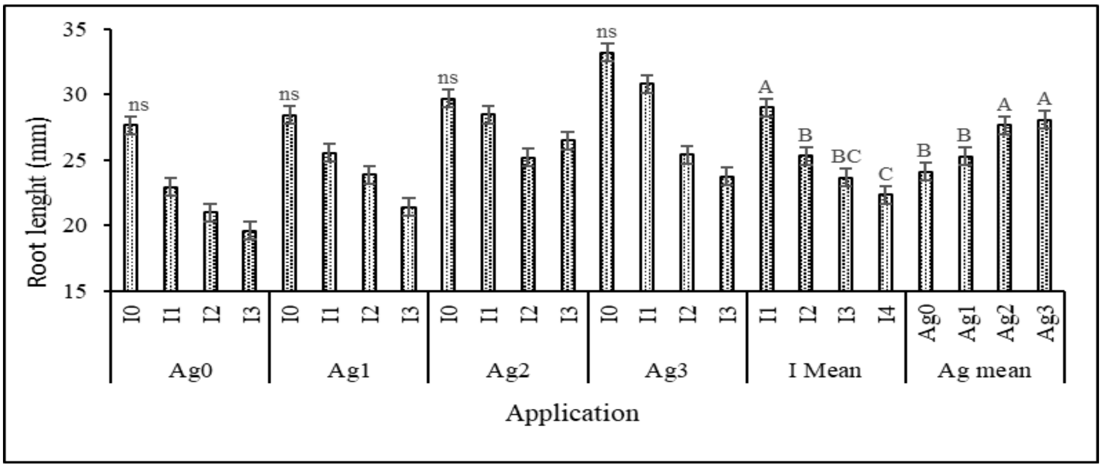

Figure 1. Effect of irrigation and AgNPs applications to root length (mm).
I mean and Ag mean application were found significant (P<0.01).
ns: Interaction between Irrigation and AgNPs application was not significant.

Results regarding root diameter of radish (Figure 2) showed that it is decreased significantly ($p<0.01$) under water stress in application I3 than control (I0). AgNPs applications had significant effects on the root diameter in compared to without AgNPs (Ag0). The highest root diameter value (19.62 mm) was detected in Ag0 and I1 application. The lowest root diameter value (10.81 mm) was Ag0 and I3 application.

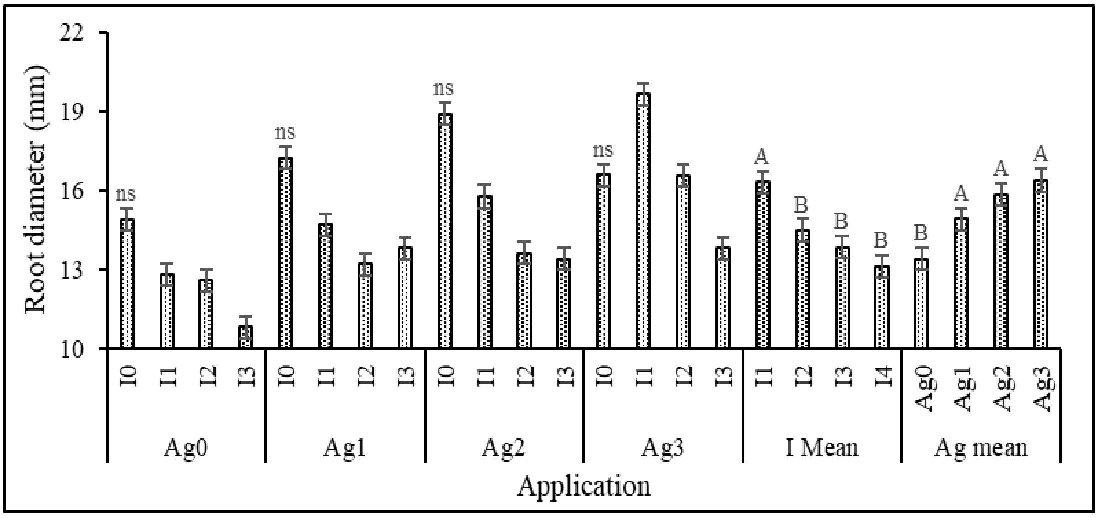

Figure 2. Effect of irrigation and AgNPs applications to root diameter (mm).
I mean and Ag mean application were found significant (P<0.01).
ns: Interaction between Irrigation and AgNPs application was not significant.

In terms of leaf number, effect of AgNPs and irrigation applications were not significant. However, in general, as the dose of limited irrigation increased, the number of plants decreased. Moreover, in average AgNPs applications, the leaf number was higher than the control (Ag0) (Figure 3). Interaction between the applications were not found significant. The highest leaf number (5.20)

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was found in Ag1 and I0 application. The lowest leaf number (4.45) was determined in Ag0 and I3. All application showed similar leaf number value statically.

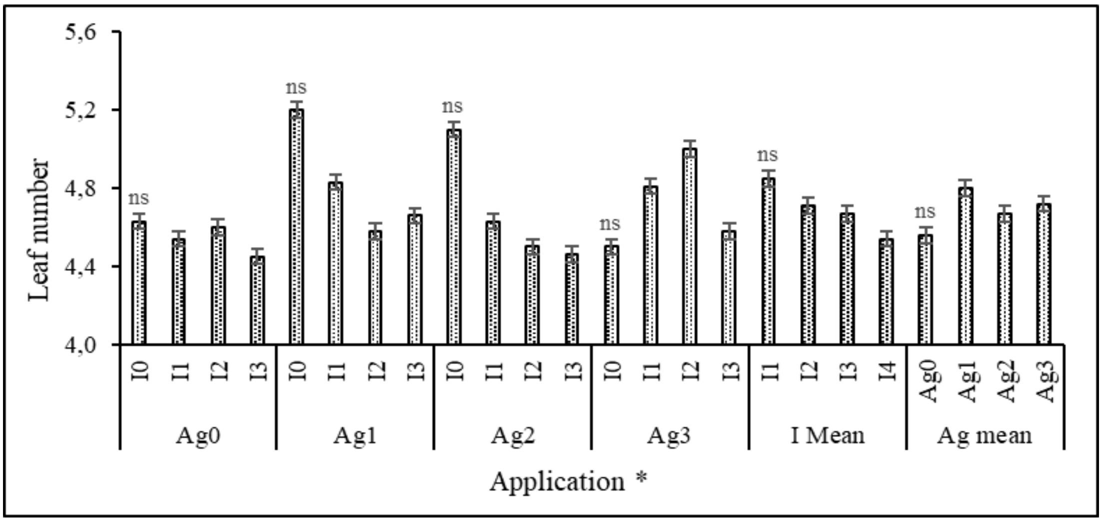

***Figure 3. Effect of irrigation and AgNPs applications to leaf number*.**
*ns: Applications was not significant.

In terms of root fresh weight, effect of AgNPs and irrigation application were found significant. It was decreased progressively subject to the deficit water stress. On the other hand, the AgNPs applications increased the root fresh weight. The highest root fresh weight value (3.85 g) was determined in Ag3 and I0 applications and lowest value (2.45 g) was determined in Ag0 and I3 applications (Figure 4). There is no significant interaction between the applications.

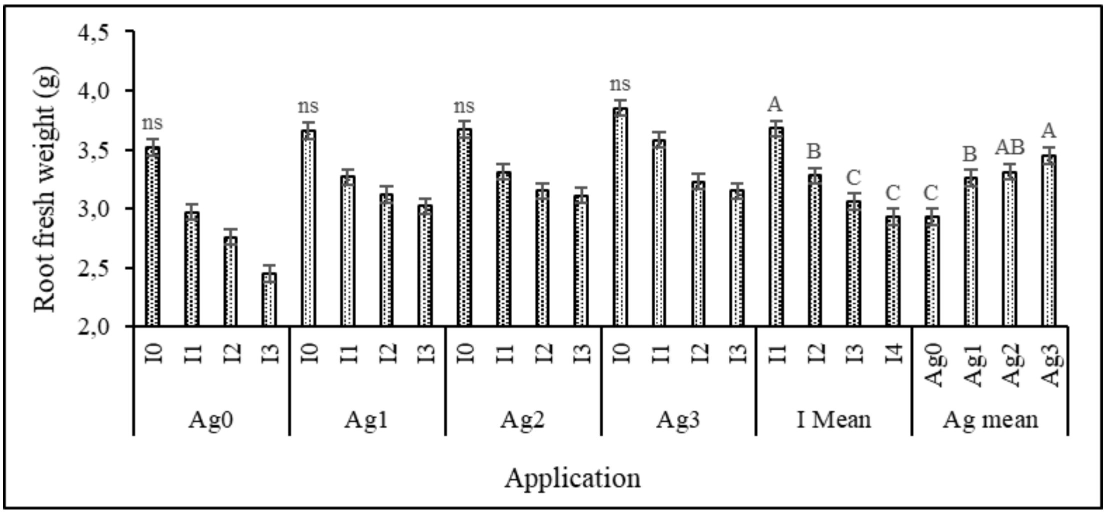

Figure. 4. Effect of irrigation and AgNPs applications to roof fresh weight (g).
I mean and Ag mean applications were found significant (P<0.01).
ns: Interaction between Irrigation and AgNPs application was not significant.

Effect of AgNPs and deficit irrigation applications on root dry weight were found significant (Figure 5). Root dry weight decreased progressively subject to the deficit water stress. It was

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affected positively from AgNPs applications. The highest value (0.48 g) was determined in Ag3 and I0 applications. The lowest value (0.34 g) was determined in Ag0 and I3 applications. In average, Ag3 application was higher than control AgNPs applications (Ag0). But Ag2 and Ag3 applications are statistically in the same group. There is no significant interaction between applications.

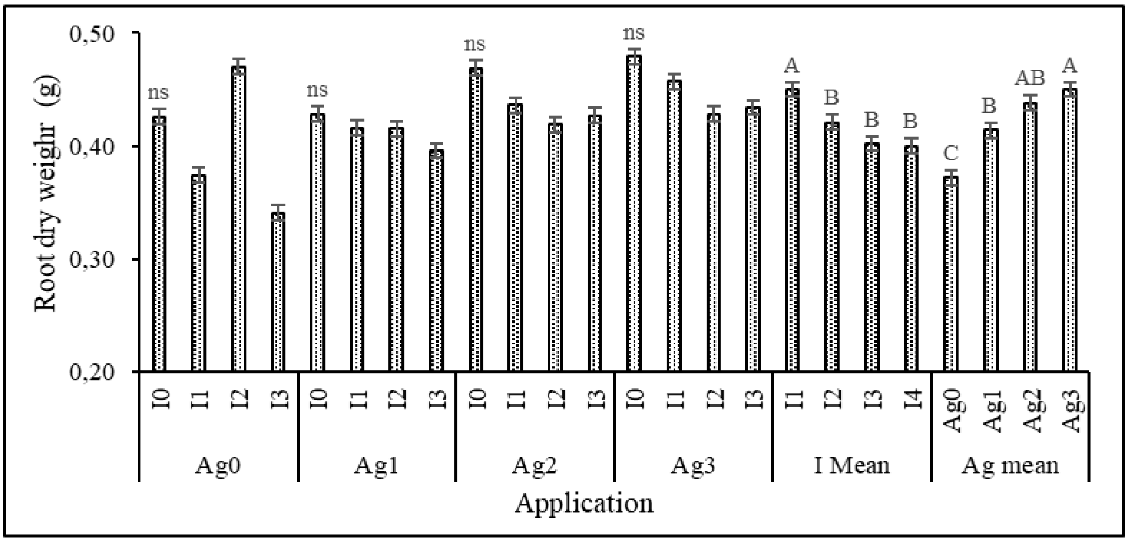

Figure 5. Effect of irrigation and AgNPs applications to roof dry weight (g).
I mean and Ag mean applications were found significant (P<0.01).
ns: Interaction between Irrigation and AgNPs application was not significant.

Under water-deficit conditions, plant growth is retarded (Sensoy et al., 2007; Coban et al., 2018). In cases where water is limited, applications that will increase the water uptake of the plant (Cakmakci et al., 2017), increase the water holding capacity of the soil (Cakmakci et al., 2021) or alleviate the stress conditions are recommended. Previous studies have been reported that the tolerance of abiotic stresses with nanomaterials that affect plant growth both positively and negatively under abiotic stress conditions (Khot et al., 2012; Zulfiqar et al., 2019). For example, AgNPs decreased root length on corn, whereas watermelon and zucchini seedling growth were positively affected by different doses of AgNPs (Almutairi and Alharbi, 2015). Small concentrations of AgNPs had a positive effect on the growth of the plantlets, while high concentrations induced a negative effect. The growth promoter dose rate was determined as 20-60 ppm in corn and it is known that this dose rate differs according to the plants (Salama, 2012). In our study, it was concluded that radish plants up to 80 ppm were positively affected by AgNPs applications. Another study was reported that AgNPs did not affect to leaf parameter similar to our study results (Gruyer et al., 2013). There are also studies where AgNPs applications affect root weight in tomato under salt stress. Different doses of AgNPs (20 nM; 0.05, 0.5, 1.5, 2 and 2.5 mg L^{-1}) were tested on germination and growth of tomato (Solanum lycopersicum L.) under salinity and reported that the dry weights and root length were improved under salinity (Almutairi, and Alharbi, 2015). There are also many studies on the toxic effect of AgNPs applications (Zuverza-Mena et al., 2016; Zhou et al., 2021). It has been determined that AgNPs applications cause lower toxicity compared to AgNO3 (Cañas et al., 2008).
The electrical conductivity varied between 280.33 µS/cm and 392.33 µS/cm. It has been observed that the electrical conductivity decreases slightly with AgNPs applications. On the other hand, the pH of the soil increased with the increase of AgNPs applications (Figure 6). The uptake of nano-

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fertilizers depends on various factors such as the distribution, accumulation, size and surface area of the particles. Soil structure, organic matter content, and soil pH are also of great importance in the application of nano fertilizers (particles) (El-Ramady et al., 2018; Ma et al., 2018). In addition, since nano particles can be taken up by both plant roots and leaves, the method of application also affects the activity of the nano particles.

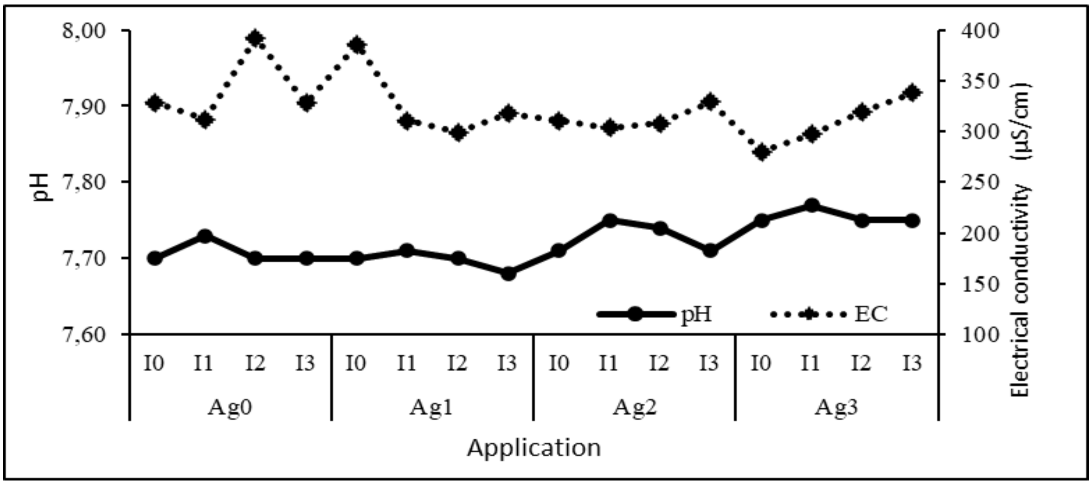

Figure 6. Soil pH and electrical conductivity values after the experiment

4. CONCLUSIONS

Nanomaterials can encourage plant growth and improve plant production based on dose. This study demonstrated the effect of silver nanoparticles on the cultivar radish (*R. sativus* L.). The presence of AgNPs at different concentrations affects root development positively of radish but did not affect to leaf parameters under deficit irrigation conditions. The maximum effect for root development was found at 80 ppm and the water stress factor affected negatively to root growth. This study showed that silver nanofertilization in radish can produce positive effects. However, in order to obtain definitive results, it should be investigated in a wide concentration range and in field conditions.

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