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# **The application of fish wastewater to improve the plant growth, development and yield of lettuce (***Lactuca sativa* **L.)**

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#### **Abstract**

Demand for low-cost and affordable alternating sources of plant nutrient responses to boost the nutrient level of damaged arable farmlands has been a main concern for soil scientists, agronomists, and local farmers. The objective of this study is to investigate the effect of fish wastewater on the growth parameters, yield, and biomass productivity of lettuce (*Lactuca sativa* L.) as compared by using in aerated nutrient solution under deep water culture (DWC) technique. The experiment was carried out to investigate shoot and root fresh and dry weight, total leaf number, leaf chlorophyll content (SPAD), photosynthesis, leaf total chlorophyll (a+ b), leaf total carotenoid content, total leaf area, leaf NRA activity, total root length, root volume and average root diameter. Lettuce plants were examined by using an aerated deep-water culture (DWC) technique in a fully automated climate room for six weeks. The seedlings were transplanted onto 8 L continuously aerated pots containing mix of different ratios of fish effluent water with tap water with six different treatments (T1, T2, T3, T4, T5 and T6) and replicated three times. The fish wastewater effluents did not reduce the growth of lettuce plants. Shoot and root fresh and dry matter, total leaf number, leaf total chlorophyll (a+ b), leaf total carotenoid content, total leaf area, leaf NRA activity, total root length, root volume and average root diameter of lettuce plants were significantly increased with under T3 treatment (Tap water  $+ 1.5$  mM N  $+ 50$  ml Nutrient solution  $+ 8$  ml Fe  $+ 1000$  ml Fish effluent water). However, the lettuce plants grown under T4 treatment (Tap water  $+ 1.5$  mM N  $+ 250$  ml Fish effluent water) had the lowest shoot and root fresh matter, total leaf number, photosynthesis, total leaf area, leaf NRA activity, total root length, root volume and average root diameter. The compost derived from the fish wastewater plays an important role in supplying the nutrients for cultivating the lettuce plants. Also, in this study appreciable nutrients were significantly obtained in treatments treated with fish wastewater, as compared with the ground (tap) water. Thus, grown lettuce with aquaculture is a good source of nutrition for human consumption.

#### **Keywords**

Fertilizer, Fish effluent water, *Lactuca sativa* L, Nutrient solution. Wastewater

#### **Introduction**

Lettuce (*Lactuca sativa* L.) is one of the most popular leafy vegetables; grown around the world [\(FAO, 2018\)](#page-6-0), it is considered to be a healthy source of minerals, fiber, vitamins, and antioxidant compounds [\(Baslam et al. 2011;](#page-6-1) [Camejo et al. 2020\)](#page-6-2). Several epidemiological studies have shown that the consumption of leafy vegetables such as lettuce is important for reducing the risk of chronic diseases, such as diabetes, cancer, and cardiovascular disease [\(Wang et](#page-7-0)  [al. 2011\)](#page-7-0). These health benefits have been linked to a

range of micro- and macro-nutrients, vitamins, and biological compounds, including carotenoids, anthocyanins, and phenolic compounds. Lettuce is generally grown under controlled environments, including hydroponic systems, greenhouse, and plant factories, with the quality of the product dependent on several factors such as light quality, nutrient composition, water level, and salt stress [\(Fu et al. 2012;](#page-6-0) [Sofo et al. 2016;](#page-7-1) [Camejo et al. 2020\)](#page-6-2).

The world population is growing dramatically which is expected to increase from 7.7 billion in 2019 to 9.7

billion in 2050 (United Nations 2019). About 5–7 million ha (0.6%) of world cropland are lost annually due to population growth, land degradation, and urbanization (WWAP 2012). Population growth with loss of cropland has resulted in a gradual decline of cultivated land worldwide from 0.44 to 0.25 ha per person over the last 50 years. Finding enough food for this population involves increasing crop production with practical fertilizer application methods. Applying chemical fertilizers to the crop production has been used to increase crop yield, development and quality for decades. Though, present agricultural trends focus on searching for alternatives to chemical fertilizers because of environmental contamination, huge procurement costs and couples with improper application leading to soil quality degradation [\(Almamori and Abdul-Ratha,](#page-6-3)  [2020\)](#page-6-3). Additionally, the world demands quality food production, getting more yield and maintaining soil biodiversity most sustainably. Therefore, for the future, it is necessary to develop and adopt strategies that provide optimal nutrition for plants and improve crop yields, whilst at the same time minimizing environmental pollution [\(Ronga et al. 2015\)](#page-7-2). The use of fish wastewater when applied to crops could be a better alternative method for efficient usage of limited water, reducing chemical fertilizer and pesticide use, enhancing crop yields, farm productivity and income. This is because of the accompanying nutrients in the fish wastewater, which could be beneficial for plant growth, yield and product quality. Previous studies have reported a significant increase in crop yields as well as an increased water use efficiency when crops are irrigated with fish effluents [\(Zajdband, 2011;](#page-7-3) [Mariscal-](#page-7-4)[Lagarda et al., 2012\)](#page-7-4). [Ramírez Sanchez et al. \(2011\)](#page-7-5) investigated the productivity of oregano in both aquaponics and hydroponics, and they reported higher fresh and dry yields in the case of aquaponics. Similarly, [Hussain and Al-Jaloud \(1995\)](#page-7-6) and [Limbu et al. \(2017\)](#page-7-7) reported a significant yield in barley and Chinese cabbage respectively the later, which yielded 80% more under fish effluent irrigation than the conventional production. Furthermore, [Castro et al. \(2006\)](#page-6-4) stated an increase in tomato yield from 64.5 to 95.8 t/ha when the plants were irrigated with aquaculture and lower yield were recorded when the plants were irrigated with well (ground water). To the best of our knowledge, limited studies have investigated replacing chemical fertilizers in nutrient solution experiments with fish effluents in lettuce production [\(Dediu et al.,](#page-6-5) 2012; [Abbey and](#page-6-6)  [Anderson, 2019;](#page-6-6) [Monsees et al., 2019;](#page-7-8) [Huang et al.,](#page-7-6)  [2021\)](#page-7-6). Therefore, the objective of this study is to investigate the effect of fish wastewater on the growth parameters, yield and biomass productivity of lettuce (*Lactuca sativa* L.) as compared by using in aerated nutrient solution under deep water culture (DWC) technique.

#### **Materials and Methods**

#### **Experimental Set-Up**

A hydroponic trial was set up using an aerated deep water culture (DWC) technique in a fully automated climate room in the Plant Physiology Laboratory of Erciyes University's Faculty of Agriculture, Department of Soil Science and Plant Nutrition, in Kayseri, Turkey. For the vegetation period, the average day/night temperatures were 25/22 °C, the relative humidity was 60-80%. The supplied photon flux in the growth chamber was almost 350 µmol  $m^{-2}$  S<sup>-1</sup> with an intensity of 16/8 h (light/dark) photoperiod. The seeds of lettuce genotype (Bachus LOL9666 variety) of the plant materials were sown in multipots in a mixture of peat (pH: 6.0-6.5) and perlite in a 2:1 ratio. Plants were transferred to 8 L plastic containers after roots were washed from growth media, each pot was filled with nutrient solution and aerated by an air pump. Due to transplanting small seedlings, the solutions were changed completely in the first two weeks and subsequently every 7<sup>th</sup> day.

The trial was set up in a completely randomized block design (CRBD) with three replications and six plants in each replication. To prepare the nutrient solution for the hydroponic experiment, analytical grade (99% pure) chemicals with distilled water were used according to the Hoagland (modified) formulation. In the solution,  $2000 \mu M$  nitrogen was supplied by using 75% calcium nitrate  $(Ca(NO<sub>3</sub>)<sub>2</sub>)$  and 25% ammonium sulfate  $(NH4)_2SO_4$ ) as the N sources. Moreover, the composition of the basic nutrient solution was as follows  $(\mu M)$ : CaSO<sub>4</sub> (1000), K<sub>2</sub>SO<sub>4</sub> (500), MgSO<sub>4</sub> (325),  $KH_2PO_4$  (250), NaCl (50),  $H_3BO_3$  (8.0), Fe-EDDHA (80), ZnSO<sup>4</sup> (0.4), CuSO<sup>4</sup> (0.4), MnSO<sup>4</sup> (0.4), MoNa2O<sup>4</sup> (0.4). All the nutrients were replaced to prior concentrations when the N concentration in the solution fell from 2.0 mM to below 1.0 mM. Daily nitrogen concentration was checked by nitrate test strips (Merck, Darmstadt, Germany) with the aid of a NitracheckTM reflectometer. Distilled water was added every 2 days to replenish the water lost to evaporation, and the solution was changed weekly.

The fishpond was covered with a black net to reduce evapotranspiration losses. A fish a stage of fingerlings (mean weight  $(8-10 \text{ g})$ ) was stocked into the pond in the laboratory condition. The pond was aerated with 1 hp ring blower that was connected with one airlift units to ensure efficient water aeration. The fish was fed 3-4 times to satiation daily with commercial pellets specific to each growth stage. The fish effluent water (3300 ml) was taken from the fishpond and transferred to 8-liter plastic containers. Then the tap water (4700 ml) was added into 8-liter plastic containers. Both of them were mixed and completed to 8 liters. Different levels (250 ml-500 ml-1000 ml) of fish water were taken from these containers and used in the experiment. The treatments used at the experiment were shown in Table 1.

#### **Plant Growth Measurements**

Plant growth was measured by using three uniform plants from each replication. Shoot and root were fractioned into the leaf, stem and roots for the fresh weight determination. And then, samples were stored separately in paper bags and dried in a ventilated oven at 74 °C for 72 hours. Root to shoot ratio was calculated by dividing the root dry weight by the sum of leaf and stem dry weights.



#### Table 1. Treatments applied to lettuce grown hydroponically at the experiment

## **Total Leaf Number and Leaf Physiological Measurements**

Each fully developed leaf was counted and recorded as a total leaf number  $(LN$  plant<sup>-1</sup>). The total leaf area  $(cm<sup>2</sup>)$  of the plants was measured with a leaf area measuring device LI-COR (LI-COR Model 3100, LI-COR. Inc., Lincoln, NE, USA). The measurements were carried out at the temperature of  $25/22$  °C (average day/night temperatures), the relative humidity of 60- 80%. The supplied photon flux in the growth chamber was almost 350 µmol m<sup>-2</sup> S<sup>-1</sup> with an intensity of 16/8 h (light/dark) photoperiod. Prior to harvest, nondestructive measurements of the leaf-level  $CO<sub>2</sub>$  gas exchange ( $\mu$ mol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>) were done in a controlled growth chamber by using a portable photosynthesis system (LI-6400XT; LI-COR Inc., Lincoln, NE, USA). Gas exchange in the leaves was performed on the youngest fully expanded leaves, using four replicate leaves per treatment in the third and fifth weeks of the growth period. The Minolta SPAD-502 chlorophyll meter was used to measure SPAD index. During the growth period, fully expanded leaves of whole plants for each treatment were twice measured for SPAD data.

## **Leaf Total Chlorophyll (a+ b) and Carotenoid Content Measurements**

A day before harvesting, extraction of the photosynthetic pigments from 100 mg (0.1 g) of fresh leaf samples from each replication of the two treatments was taken for measuring the leaf total chlorophyll and carotenoid contents using UV-VIS Spectroscopy. The leaf samples used for chlorophyll and carotenoid determinations were of the same physiological age as those used for the leaf net photosynthesis measurements. The samples were put into 15 ml capped containers where 10 ml of ethylene alcohol of 95% concentration was added. They were then kept in darkness at room temperature overnight, to allow the extraction of the leaf pigments. Measurements were done using the spectrometer (UV/VS T80+ of PG Instruments Limited, UK) at wavelengths of 470 nm, 648.6 nm, and 664.2 nm. Total chlorophyll (Total-Chlo) and total carotenoids (TC) were then estimated from the spectrometric readings using the formulae o[f Lichtenthaler \(1987\).](#page-7-9)

Total-Chlo (mg/g plant sample) =  $[(5.24 \text{ WL664.2} -$ 22.24 WL648.6 x 8.1]/ weight of plant sample (g)

TC (mg/g plant sample) =  $[(4.785 \text{ W}L470 + 3.657$ WL664.2) -12.76 WL648.6) x 8.1]/ weight of plant sample (g)

Note: WL470, WL648.6 and WL664.2 refers to spectrometric readings at wavelength 470 nm, 648.6 nm and 664.2 nm respectively.

## **Root Morphological Measurements**

The plant root morphological parameters such as total root length  $(m)$ , total root volume  $(cm<sup>3</sup>)$  and average root diameter (mm) were measured by using a special image analysis software program WinRHIZO (Win/Mac RHIZO Pro V. 2002c Regent Instruments Inc., Québec, QC G1V 1V4, Canada) in combination with recording device of Epson Expression 11000XL scanner (Long Beach, CA, USA).

## **Leaf Nitrate Reductase (NRA) Activity Measurement**

Nitrate reductase (NRA) activity in the leaf was determined following the method proposed by Harley (1993). At harvesting fresh plant samples were taken and chopped into pieces; two grams of the latter were placed in each of two falcon tubes and labeled time-0 (T0) and time-60 (T60). The tubes were covered with aluminum foil to be screened from light. Ten ml of assay buffer solution [100 mM phosphate buffer, pH 7.5; 30 mM KNO<sub>3</sub>; 5%  $(v/v)$  propanol] was added to each tube (T0 and T60). The T0 container was immediately placed into boiling water for five minutes, removed and allowed to cool to room temperature. While the T60 was kept for 60 minutes at room temperature; after which it was also placed into boiling water for five minutes and allowed to cool to room temperature. To detect nitrite in the assay tubes, the optical density (OD) of each standard tube was determined at 540 nm wavelength in the spectrometer.

#### **Statistical Analysis**

All measured physiological and morphological parameters were analyzed using SAS Statistical Software (SAS 9.0, SAS Institute Inc., Cary, NC, USA). A two-factorial analysis of variance was performed to study the effects of genotypes and salt and their interactions on the variables analyzed. Levels of significance are represented by  $* p < 0.05$ ,  $** p < 0.01$ , \*\*\*  $p < 0.001$ , and ns means not significant. Differences between the treatments were compared using Duncan's Multiple Test  $(p < 0.05)$ .

#### **Results and Discussion**

#### **Biomass Production and Partitioning**

The results of lettuce harvest after 35 days of cultivation are shown in Table 2. The shoot and root fresh matter, and total leaf number were significantly (*p* < 0.001) affected by the treatments. It was obvious that treatment T3 performed remarkably better than other treatments. The significantly highest shoot (265.5 g plant<sup>-1</sup>) and root fresh matter (48.7 g plant<sup>-1</sup>), and total leaf number (60 LN plant<sup>-1</sup>) were produced in the treatment T3. Though, significantly lowest shoot (51.1  $g$  plant<sup>-1</sup>) and root fresh matter (10.9 g plant<sup>-1</sup>), and total

leaf number  $(47 \text{ LN plant}^{-1})$  were produced in the treatment T4 (Table 2). This result shows that in the treatment T3 of lettuce is more nutritious than others which helps lettuce to grow and develop better. While the treatment T4 is less nutritious so it grows and develops slowly. The average fresh weight of lettuce in this study was greater than fresh lettuce weights from other aquaponic studies conducted (Table 2). The average fresh weight of lettuce (Bachus LOL9666 variety) from this study (152 g) was more than the lettuce genotype of 'Salanova' from both aquaponic (89 g) and hydroponic production (91.18 g; [Søberg, 2016\)](#page-7-10). According to the University of California Davis, the average fresh weight of a loose-leaf lettuce in field production is 415.79 g [\(Takele, 1996\)](#page-7-11). This is significantly more than the average fresh weight of any cultivar from this study or other aquaponic and hydroponic lettuce production studies reviewed. It appears that across studies, the average weight of lettuce produced in an aquaponic or hydroponic production system will be less than half of typical field production.

[Al-Jaloud and Hussein \(1995\)](#page-6-7) stated that increase in the yield of wheat and French bean with the application of fish pond, while [Nadafi et al. \(2005\)](#page-7-8) wood also reported increased growth rate and improved quality of garden purslan, sweet basil, and radish and cucumber crops with fish pond water application. Also, higher yield of fresh bean pod and fresh Kale leaf increases via fish pond water application as a source of fertilizer [\(Wood et al. 2001\)](#page-7-12). These results showed that irrigating with fish pond water; results in greater yield. This also revealed that nutritional values are present in the fish pond water which is the obvious factors responsible for the improved yield increased.

[Delaide et al., \(2016\)](#page-6-5) reported that the supplementation of fish water with mineral fertilizer increased the fresh weight of lettuce by nearly 40%. In contrast, [Suhl et al. \(2016, 2018\)](#page-7-13) documented no significant differences in tomato yield between conventional hydroponics and supplemented aquaponics.

|                | Shoot Fresh Weight         | Root Fresh Weight          | <b>Total Leaf Number</b>    |
|----------------|----------------------------|----------------------------|-----------------------------|
| Treatments (T) | $(g$ plant <sup>-1</sup> ) | $(g$ plant <sup>-1</sup> ) | $(LN$ plant <sup>-1</sup> ) |
| T1             | 215.6c                     | 24.1 c                     | 58 c                        |
| T <sub>2</sub> | 220.5 <sub>b</sub>         | 17.3d                      | 59 b                        |
| T <sub>3</sub> | 265.5a                     | 48.7 a                     | 60 a                        |
| T <sub>4</sub> | 51.1 f                     | 10.9f                      | 49e                         |
| T <sub>5</sub> | 55.1e                      | 13.9 <sub>e</sub>          | 47f                         |
| T <sub>6</sub> | 106.6d                     | 35.4 <sub>b</sub>          | 52 d                        |
| F-Test:        |                            |                            |                             |
| Treatments     | ***                        | ***                        | ***                         |

Table 2. Shoot and root fresh weight and total leaf number of lettuce grown under different six treatments

<sup>1</sup>Values denoted by different letters are significantly different between treatments within columns at  $p < 0.05$ . ns, nonsignificant. \*  $p < 0.05$ , \*\*  $p < 0.01$  and \*\*\*  $p < 0.001$ .

Table 3 shows the shoot and root dry weight and root: shoot ratio of lettuce plants grown in different treatments hydroponically. The shoot and root dry matter, and root:shoot ratio were significantly ( $p <$ 0.001) affected by different treatments. The results show that shoot  $(17.6 \text{ g plant}^{-1})$  and root dry weight  $(2.5 \text{ g})$ plant-1 ) were significantly higher at the treatment of T3. The lowest shoot dry matter of plants is at treatment T5, and the average value is  $4.8$  g plant<sup>-1</sup>. The highest root:shoot ratio of lettuce plants was observed at the treatment T5 recording an average value of 0.23 g  $g^{-1}$ . When comparing the shoot dry matter among the treatments, the results show that using the fish effluent water (1000 ml) combined with nutrient solution (50 ml) has helped to increase the shoot dry matter faster compared with using fish effluent water only. This is an indication that there is an obvious relationship between the shoot and root dry weights with respect to volume of fish pond water applied. Similar results were observed by [Akindele et al. \(2021\)](#page-6-8) at sweet pepper. They stated that the highest number of leaves, stem girth, biomass and sweet pepper yield, root weight and leaf area index (LAI) were recorded in treatment, 100% of Potential Evapotranspiration (PET) for aquaculture water (T2) as compared to 50% of PET for aquaculture water (T1) and

100% of PET for ground water (T0). [Li et al \(2021\)](#page-7-14) stated that the rice and fish yield increased in integrated system compared to monoculture. Similar results were obtained by [García-Santiago et al \(2021\).](#page-6-9) They stated that leaf dry weight, total plant biomass dry weight, fruit number and total yield were higher in the organic fertilization treatment (including fish-derived protein hydrolysate as an N-source), surpassing the conventional treatment by 35%, 9%, 21%, and 4% for these parameters, respectively, though the difference was only significant for leaf dry weight in grape tomatoes.

#### **Leaf Chlorophyll Content (SPAD), Photosynthesis, Leaf Total Chlorophyll (a+ b) and Carotenoid Content, Total Leaf Area and Leaf NRA Activity**

The results indicate that leaf chlorophyll content (SPAD), photosynthesis, leaf total chlorophyll (a+ b), leaf total carotenoid content, total leaf area and leaf NRA activity at the end of the growing cycle were significantly  $(p < 0.001)$  affected by treatments (Figure 1). Concerning SPAD, lettuce plants grown under the treatment T4 had the highest leaf chlorophyll content, reaching an average of 34.2 SPAD as shown in Figure 1A. This was closely followed by lettuce plants grown under T3 treatment, with an average number of 31.5 SPAD. The treatment T5, however, recorded the significantly lowest with 20.8 SPAD. In plants, chlorophyll is a green pigment, which is vital as far as photosynthesis is concern; it helps in transforming light energy to chemical energy during the activities of photosynthesis. Also, the amount of chlorophyll present in a leaf is very paramount to depicts the growth of plants [\(Bannari et al. 2007\)](#page-6-10). Therefore, in crop production chlorophyll is necessary for photosynthetic activities.



<sup>1</sup>Values denoted by different letters are significantly different between treatments within columns at  $p < 0.05$ . ns, nonsignificant. \*  $p < 0.05$ , \*\*  $p < 0.01$  and \*\*\*  $p < 0.001$ .

Regarding photosynthesis, the results showed that treatment T1 performed best compared to the rest of the treatments. It recorded an average of 8.16  $\mu$ mol CO<sub>2</sub> m<sup>2</sup> s -1 photosynthesis as shown in Fig. 1B. This was followed by treatment T2 with the value of  $5.82 \text{ CO}_2 \text{ m}^2$ s<sup>-1</sup> and closely by treatment T3 with the value of 5.61  $CO<sub>2</sub>$  m<sup>2</sup> s<sup>-1</sup>. Data in Fig. 1C and 1D shows that lettuce plants grown mixed treatment with 1000 ml fish effluents and 50 ml nutrient solution (T3) obtained significantly higher values in leaf total chlorophyll (a+ b) and leaf total carotenoid content at  $p < 0.001$ , with a value of 1.269 µmol g<sup>-1</sup> and 0.214 µmol g<sup>-1</sup>, followed by the mixed treatment with 500 ml fish effluents and 100 ml nutrient solution (T2) treatment at 0.752 µmol  $g^{-1}$  and 0.133 µmol  $g^{-1}$  for leaf total chlorophyll (a+ b) and leaf total carotenoid content respectively. Results from treatment T4 had significantly lower leaf total chlorophyll (a+ b) and leaf total carotenoid content with a value of 0.451 µmol  $g^{-1}$  and 0.083 µmol  $g^{-1}$ . Similar results were observed by [Akindele et al. \(2021\)](#page-6-8) at sweet pepper. They stated that the highest number of leaves, stem girth, biomass and sweet pepper yield, root weight and leaf area index (LAI) were recorded in treatment, 100% of Potential Evapotranspiration (PET) for aquaculture water (T2) as compared to 50% of PET for aquaculture water (T1) and 100% of PET for ground water (T0). Few studies have attempted to clarify the

effect of fish effluents in crop production but mostly under the rice-fish culture [\(Jamu and Piedrahita, 2002;](#page-7-15) [Koide et al., 2015\)](#page-7-16). A few researchers have examined the influence of organic fertilization on marjoram crops and stated paramount positive effects [\(Gharib et al.,](#page-6-11)  [2008;](#page-6-11) [Naguib, 2011\)](#page-7-17).

In terms of total leaf area, the treatment T3 gave the highest value of  $4331.1 \text{ cm}^2 \text{ plant}^1$ , while followed by the treatment T1 with the value of  $3347.2 \text{ cm}^2 \text{ plant}^{-1}$ , respectively. Evidently, the treatment T4 gave the lowest value  $(1363.7 \text{ cm}^2 \text{ plant}^{-1})$  for the total leaf area production. Thus, affirmed that grown plants with fish pond water (1000 ml) does directly increased leaf area development in lettuce production. This is as a result of enhancement of photosynthesis due to higher total leaf area [\(Ogbonnaya et al. 1998\)](#page-7-17).

Results of leaf NR activity indicates that plants grown under treatment T3 recorded the highest leaf NR activity, though plants grown under treatment T4 recorded the lowest leaf NR activity. Nitrogen is a decisive nutrient for plant growth [\(Hawkesford et al.,](#page-6-12)  [2012\)](#page-6-12). Therefore, the focus for mixing the nutrient solution in the present study was to align N in the different treatments, which was only possible with regard to the total N.







Figure 1. Leaf chlorophyll content (SPAD) (A), photosynthesis (B), leaf total chlorophyll (a+ b) (C), leaf total carotenoid content (D), total leaf area (E) and leaf NRA activity (F) of lettuce grown under different six treatments. <sup>1</sup>Values denoted by different letters are significantly different between treatments within columns at  $p < 0.05$ . ns, non-significant. \* *p* < 0.05, \*\* *p* < 0.01 and \*\*\* *p* < 0.001.

#### **Total Root Length, Total Root Volume and Average Root Diameter**

Total root length, root volume and average root diameter were significantly ( $p < 0.001$ ) affected by different treatments (Table 4). Results show that plants grown at the treatment T3 recorded relatively higher values for total root length  $(476.8 \text{ m plant}^{-1})$ , root volume  $(45.4 \text{ cm}^3 \text{ plant}^{-1})$  and average root diameter (16.12 mm) compared to the other treatments. This was closely followed by the treatment T6. This result shows

that in the treatment T3 of lettuce is more nutritious than other treatments which helps lettuce to grow and more root development better. Similar results were observed by [Akindele et al. \(2021\)](#page-6-8) at sweet pepper. They stated that the highest number of leaves, stem girth, biomass and sweet pepper yield, root weight and leaf area index (LAI) were recorded in treatment, 100% of Potential Evapotranspiration (PET) for aquaculture water (T2) as compared to 50% of PET for aquaculture water (T1) and 100% of PET for ground water (T0).





<sup>1</sup>Values denoted by different letters are significantly different between treatments within columns at  $p < 0.05$ . ns, nonsignificant. \*  $p < 0.05$ , \*\*  $p < 0.01$  and \*\*\*  $p < 0.001$ .

#### **Conclusion**

This study was conducted to investigate the effect of fish wastewater on the growth parameters, yield and biomass productivity of lettuce (*Lactuca sativa* L.) as

compared by using in aerated nutrient solution under deep water culture (DWC) technique. Results showed that the lettuce plants grown under T3 treatment (Tap water  $+ 1.5$  mM N  $+ 50$  ml Nutrient solution  $+ 8$  ml Fe + 1000 ml Fish effluent water) produced the highest

shoot and root fresh and dry matter, total leaf number, leaf total chlorophyll (a+ b), leaf total carotenoid content, total leaf area, leaf NRA activity, total root length, root volume and average root diameter, while the lettuce plants grown under T4 treatment (Tap water + 1.5 mM  $N + 250$  ml Fish effluent water) had the lowest shoot and root fresh matter, total leaf number, photosynthesis, total leaf area, leaf NRA activity, total root length, root volume and average root diameter. The compost derived from the fish wastewater plays an important role in supplying the nutrients for cultivating the lettuce plants. Also, in this study appreciable nutrients were significantly obtained in treatments treated with fish wastewater, as compared with the ground (tap) water. Thus, grown lettuce with aquaculture is a good source of nutrition for human consumption.

#### **Compliance with Ethical Standards Conflict of interest**

The authors declared that for this research article, they have no actual, potential, or perceived conflict of interest.

## **Author contribution**

The contribution of the authors to the present study is equal. All the authors read and approved the final manuscript. All the authors verify that the Text, Figures, and Tables are original and that they have not been published before.

#### **Ethical approval**

Not applicable. **Funding**

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**Data availability**

#### Not applicable. **Consent for publication**

Not applicable.

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