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# Cultivation of lettuce (*Lactuca sativa* L.) and rainbow trout (*Oncorhynchus mykiss* W.) in the aquaponic recirculation system

## Култивиране на маруля (*Lactuca sativa* L.) и дъгова пъстърва (*Oncorhynchus mykiss* W.) в аквапонна рециркулационна система

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## ABSTRACT

Aquaponics is combined growing fish and plants in a recalculating system. Therefore, it is very important to achieve optimal conditions for their cultivation. The purpose of this study was to trace the influence of the water used by the cultivated fish on the biomass of the lettuce in the aquaponic system. In this connection, two types of hydroponic sections were built and integrated into an existing recirculation aquaculture system. The hydrochemical parameters were measured during the trial. The duration of experiment was 60 days. Forty specimens from the fish species rainbow trout (*Oncorhynchus mykiss*) with an average weight of 13.4 g in good health condition were growing of the each of tanks of the aquaponic system. Sixteen lettuce seedlings were planted on the hydroponic section filled with light weight expanded clay aggregate (LECA) and the other sixteen plants was measured. The productivity of lettuce plants is highly dependent on the type of plant growing medium and the fish biomass. A better removal capacity in ammonium, nitrate and ortho-phosphate were observed in the LECA section compared with the cleaning capacity in the raft section as a part of experimental aquaponics system.

Keywords: aquaponics, deep water culture, hydrochemical parameters, lettuce, rainbow trout

## АБСТРАКТ

Аквапониката е комбинирано отглеждане на риби и растения в рециркулационна система. Ето защо е много важно да се постигнат оптимални условия за тяхното отглеждане. Целта на това изследване е да се проследи влиянието на водата, използвана от култивирането на рибата върху биомасата на марулята в аквапонната система. В тази връзка бяха изградени и интегрирани два типа хидропонни секции в съществуващата рециркулационна система. Хидрохимичните параметри бяха измерени по време на опита. Продължителността на експеримента беше 60 дни. Четиридесет риби от дъгова пъстърва (*Oncorhynchus mykiss*) със средно тегло от 13.4 грама в добро здравословно състояние бяха отглеждани във всяка вана от аквапонната рециркулационна система. Шестнадесет марули бяха поставени в хидропонната секция пълна с лек експандиран глинен агрегат (LECA), а други шестнадесет растения бяха засадени на плаващия хидропонен участък. В края на изпитването беше измерено свежото тегло на марулята. Продуктивността на марулята е силно зависима от типа на хранителната среда и от биомасата на рибите. По-добър капацитет на отстраняване на амония, нитратите и фосфатите се наблюдава в LECA секцията в сравнение с капацитета на почистване в плаващата секцията, като част от експерименталната аквапонна система.

Ключови думи: аквапоника, дъгова пъстърва, дълбоководна култура, хидрохимични параметри, маруля

## INTRODUCTION

Aquaponics is the integration cultivation of hydrobionts and vegetables in one production system (Timmons et al., 2002; Rakocy et al, 2006; Karimanzira et al., 2016). Producing plants hydroponically and farming fish using aquaculture have their own special requirements in order to properly manage each system to produce maximum yields (Tyson et al., 2012). In aquaponics, the fish wastes are removed from the water through plant beds and not released to the environment, while at the same time the nutrients for the plants are supplied from a sustainable, cost-effective and non-chemical source (Gregory et al., 2018). In the country where land and water are limited, aquaponics can be more productive and economically feasible. Hydroponics and aquaculture combining allows the chemical nutrients needed for hydroponic plant growth to be replaced with fish wastes and to prevent environmental pollution (Bernstein, 2011).

The aquaponics includes three major groups of organisms: fish, plants and bacteria. Optimal plant growth and development in aquaponic system depends on the availability of light, water, conducive temperatures and mineral nutrients (Silva et al., 2015). Nutrition induces variable physiological and developmental responses in the plants. They need a large amount of nitrogen (N), which is the main compound of photosynthesis enzymes, pigments, and products. The increase in productivity observed for many plant species is often associated with the elevation in photosynthesis intensity (Jain et al., 1999). There are two nitrogen forms absorbed by plants - the nitrate ion  $(NO_3^-)$  and ammonium ion  $(NH_4^+)$ . Nitrate is assimilated by two reductive steps catalyzed by the enzymes NR and nitrite reductase (NiR). Nitrate reductase (NR) is one of the most significant enzymes in the assimilation of exogenous nitrate (NO<sub>3</sub><sup>-</sup>), regulates the protein synthesis, and play important role in amino acids biosynthesis (Campbell, 1999). The activity of this enzyme given the good assessment of the nitrogen level of the plant and is very often correlated with growth and yield (Barford and Lajtha, 1992). The microbes (nitrifying bacteria) are the third participant. These bacteria convert ammonia from the fish waste first into nitrites. and then into nitrates. Nitrates are the form of nitrogen that plants can uptake and use to grow. Thanks to them solid fish waste is turned into vermicompost and acts as food for the plants. The efforts of many researchers are connected to the optimization of aquaponics as an innovative technology. The main parameters, which defined the cleaning capacity of cultivated plants and the productivity of aquaponics systems, are the type of hydroponic compartment and plant's growing media.

A limited number of studies are connected with these topics (Lennard and Leonard, 2006; Roosta and Afsharipoor, 2012; Schmautz et al., 2016).

The purpose of this study was to trace the influence of the water used by the cultivated fish (*Oncorhynchus mykiss* W.) on the biomass of the lettuce in the aquaponic system. The impact of different media bed and raft hydroponic sections as a part of a model aquaponic recirculation system was traced.

#### MATERIALS AND METHODS

#### Aquaponics system

Two types of hydroponic sections (media bed and deep-water sections) were constructed and integrated into an existing recirculation aquaculture system situated at the Experimental aquaculture base in Trakia University, Stara Zagora, Bulgaria (Figure 1). For the cultivation of rainbow trout were used six tanks each with a volume of the fish tank 2 m<sup>3</sup>.

The total volume of the settling tank and biofilter was 5 m<sup>3</sup>. The water from the filters was pumped into fish tanks and aquaponics sections. The valve split the water between fish tanks and hydroponic sections. The water flow rate in hydroponic sections was maintained at 0.5 L/min. A water flow rate of 3 L/min was assured to fish tank. The light for each of the hydroponic compartments were assured from 2 plant growing lights (Osram Fluorescent Fluora Tubular Linear Lamp). The first hydroponic section was filled with light weight expanded clay aggregate (LECA) and the second hydroponic section used polystyrene sheet with 5 mm thickness which floated on the surface of water (Figure 1).

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Figure 1. View from a model aquaponics system with lightweight expanded clay aggregate (LECA) section and raft aquaponics section

Every day the bottoms of settling and fish tanks were siphoned and the sediments were removed. The water lost during the cleaning process and evaporation was compensated by adding of fresh water (up to 10% of recirculation aquaponics system' volume per day).

#### Experimental fish and plants

Forty specimens from the fish species rainbow trout (*Oncorhynchus mykiss*) with an average weight of 13.4 g in good health condition were adapted for one week to the condition of the each of tanks of the aquaponic system. The used stocking density was 0.67 kg/m<sup>3</sup>. The fish were fed manually three times per day. The daily feed ration was adjusted to 2% from trout's biomass. The mortality of experimental fish was registered daily.

For the trial 32 lettuce seedlings (15 days old *Lactuca* sativa variety "Jylta krasavica") were chosen and transported from greenhouse situated in Plovdiv to the Experimental aquaculture base at Trakia University.

The lettuce plants were transferred to rock wool (Grodan®) substrates and afterward, all plants were

placed in hydroponic pots. Sixteen lettuce seedlings were planted on the hydroponic section filled with light weight expanded clay aggregate (LECA) and the other sixteen plants were planted on the floating raft hydroponic section.

## Measured parameters

The duration of experiment was 60 days. The biomass of the experimental trouts was calculated at the start and end of experiment.

The percentage weight gain (PWG) in experimental fish was calculated according following equation:

where:

W1 - initial weight of trout;

W2 - final weight of trout at the end of trial.

The survival of fish (%) during the trial was also registered.

The cleaning capacity of different hydroponic sections was investigated by measurement of hydrochemical parameters in sump and after raft and LECA bed hydroponic compartments. The oxygen content (mg/L), pH and electrical conductivity ( $\mu$ S/cm<sup>3</sup>) were measured daily with a portable meter (HQ30D) accordingly with LDO, pH (liquid) and conductivity electrodes. Dynamics of nitrogen (ammonium and nitrate) (mg/L) and phosphorus (ortho-phosphate-phosphorus) (mg/L) compounds were measured spectrophotometrically with the DR 2800 (Hach Lange) every 10 days with appropriate tests for them (Hach Lange, 2007).

At the end and middle of the trial the fresh weight of lettuce plants was measured on technical balance with 0.01 g accuracy. The length of roots in experimental plant cultivated at a two production technology was also measured (cm).

#### Statistical analysis of data

The data received from the trial were statistically analysed with ANOVA single factor (MS Office, 2010).

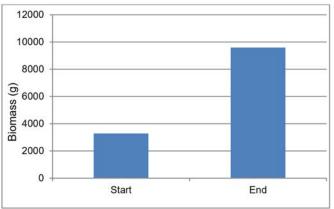
#### **RESULTS AND DISCUSSION**

The most commonly used aquaponics systems are a media-filled raised bed (McMurtry et al., 1997; Lennard and Leonard, 2006; Tyson et al., 2008), NFT or nutrient-film technique (Adler et al., 2000; Lennard and Leonard, 2006; Nelson, 2007), or a floating raft system (Rakocy, 1997; Lennard and Leonard, 2006; Nelson, 2007) for the plant growing area integrated with a recirculation aquaculture tank system (Timmons et al., 2002) for the fish production area. For this reason, this research is based on these two hydroponic sections - LECA and raft.

According to Tyson et al. (2012) sizing aquaponic systems so the plant production area is large enough to re-use this excess water through plant uptake and evapotranspiration increases system sustainability by reducing waste effluent discharges to the environment while supplying nitrogen and other nutrients to the plants. This aquaponic system has a sufficient volume for growing plants and large biomass fish.

#### **Experimental fish**

Survival rate was 97.5% in experimental fish during the trial. At the end of the experimental period the biomass in trout increased 2.91 times compared to its initial value. It was found that percentage weight gain (PWG) of trout was 206.12% at the end of the experiment (Figure 2).



**Figure 2.** Biomass of experimental rainbow trout (*Oncorhynchus mykiss*) cultivated in aquaponics system

#### **Experimental plants**

The average weight of lettuce plants cultivated in LECA compartment was higher with 4.7% at the end of the current trial in comparison to the received values for this parameter in the experimental plants in the raft section (Figure 3).

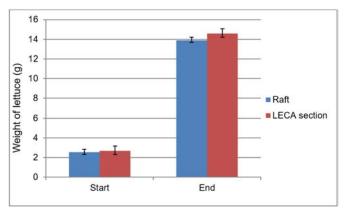


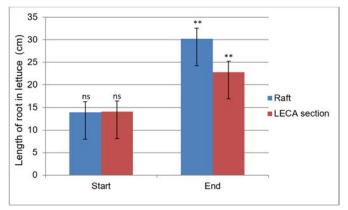
Figure 3. The average weight of lettuce (g) plants cultivated in Raft and LECA section

The length of the lettuce roots in both hydroponic sections increased significantly (Figure 4). At the end of the trial the average length of the roots in lettuce cultivated in the raft section was higher with 24.49% compared with the average length of the roots in the experimental plants cultivated in the LECA section and the difference was significant (Figure 5).

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**Figure 4.** Length of the root in lettuce cultivated in Raf and LECA section



**Figure 5.** The average length of root in lettuce cultivated in Raft and LECA section (cm), \*\*significantly different (P<0.01), ns – not significantly different

## Hydrochemical parameters

In aquaponic recirculation system a hydroponic section serves as a biofilter (Endut et al., 2009), decreasing amount metabolites excreted from fish and toxicological compounds which are released into water from feces and uneaten feed. According Lennard and Leonard (2006) lettuce grown in gravel as a hydroponic media was efficient at removing nutrients from fish culture water.

For good growth of hydroponically grown lettuce and for the highest uptake of N, P and K, the optimum range of pH was found to be 5–6 (Knight and Mitchell, 1983; Karimaei et al., 2004). The optimum value for fish does not match with the optimum pH for plant growth (Rakocy and Hargreaves, 1993).

In this study the level of pH before aquaponics section was much closer to neutral pH compared to both experimental hydroponic sections, but the difference was not statistically proven (Table 1).

According to Chun and Takakura (1994) the critical dissolved oxygen concentration for root respiration of lettuce plants are 2.5 mg/L. During this experiment the dissolved oxygen in the systems was over 7 mg/L. The quantity of oxygen in the raft and LECA sections was a little higher accordingly with 2.34% and 0.4% compared with its value in sump before the hydroponic sections but the differences were not significance (P>0.05).

The quantity of ammonium nitrogen, nitrate nitrogen and ortho-phosphate-phosphorus significantly decreased after the experimental hydroponic sections. The quantity of ammonium nitrogen was with 84.6% higher after raft section compared to its quantity in water after LECA section.

The concentration of nitrate was higher with 4.44% in the section used deep water technology compared with its value in the LECA section.

The lower concentration of ortho-phosphate phosphorus was observed also in the LECA section in comparison to the concentration of this compound measured in the raft section (with 17.3% higher value) (Table1).

Parameters	Before aquaponics section $\bar{x}$ + Sx	After RAFT aquaponics section $\bar{x}$ + Sx	After LECA aquaponics section $\bar{x}$ + Sx
pH	7.2±0.21	7.9±0.38	7.8±0.32
Oxygen (mg/L)	7.09±0.34	7.26±0.25	7.12±0.12
Conductivity (µS/cm)	272±1.03	274±1.16	273±1.09
Ammonium nitrogen (mg/L)	0.087±0.02*	0.078±0.03	0.012±0.01*
Nitrate nitrogen (mg/L)	6.72±0.33*	5.63±0.28*	5.38±0.21
Ortho-phosphate-phosphorus (mg/L)	0.26±0.28*	0.168±0.06*	0.139±0.02*

Table 1. Hydrochemical parameters in aquaponics system

\* Significantly different at P<0.05.

These results are confirmed by Lenard and Leonard (2006), according to which the gravel bed and floating hydroponic section are suitable for integration with recirculation aquaculture system.

Also, they found that oxygen concentration and nitrate removal was higher in the raft system but phosphate removal was higher in the gravel bed section. In this trial results showed a better removal capacity of ammonium, nitrate and ortho-phosphate in the LECA section compared to the cleaning capacity in the raft section. Chaves et al. (2000) established improvement of water quality through an integration of hydroponically grown lettuce where nitrate concentration was reduced by 20-27%.

## CONCLUSIONS

The two subsystems, LECA bed and floating hydroponic are suitable for integration with the recirculation aquaculture system. A better removal capacity in ammonium, nitrate and ortho-phosphate were observed in the LECA section compared to the cleaning capacity in the raft section as a part of experimental aquaponics system. The LECA bed technology showed better plant biomass production, but in raft technology, root length was higher.

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