

THE INFLUENCE OF CROP DENSITY ON PHOSPHORUS DYNAMICS IN AN INTEGRATED STELLATE STURGEON - SPINACH RECIRCULATING PRODUCTION SYSTEM

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Abstract

In an integrated aquaponic system, the phosphorus present in the technological water can be recycled in the plant biomass, depending on a series of elements related to system design and production technology. The present research aims to identify the influence of crop culture density on phosphorus dynamics in an integrated stellate sturgeon – spinach recirculating production system, by using aquaponic substrate technique. A 44 days experimental trial was performed in order to test three spinach culture densities, in triplicate, as follows: B1H - 59 crops/m², B2H - 48 crops/m², B3H - 39 crops/m². A control variant (with no plants) was used, in order to estimate the spinach biomass capacity to remove phosphorus. Phosphorus removal rate had an upward trend at B1H, B2H and B3H, with an average value of 13.38 ± 5.45 g/m²/day at B1H, 11.57 ± 5.25 g/m²/day at B2H and 10.39 ± 4.4 g/m²/day at B3H. Statistically significant differences ($p < 0.05$) were recorded between B1H and the rest of experimental variants. Although the highest phosphorus removal rate was recorded at B1H, some spinach plants manifested signs of stress, fact that might affect the phosphorus removal rate capacity during a longer experimental period.

Key words: aquaponic, spinach, stellate sturgeon, phosphorus, removal rate.

INTRODUCTION

In order to maintain both environmental and economic sustainability, a proper waste management must be applied among aquaculture production systems, by identifying new techniques for water treatment. Several studies (Zhang et al., 2021; Robinson et al., 2019) confirmed that solving the phosphorus and nitrogen pollution occurred due to intensive aquaculture practices is the key to maintain the ecosystem health. Thus, both physico-chemical and biological methods can be applied in order to remediate the aquaculture effluents. However, according to several studies (Liang et al., 2020; Jinet et al., 2019; Benammar et al., 2015), the physico-chemical methods are less sustainable, from both economic and environmental perspective, compared to biological methods.

Phosphorus is commonly encountered in high concentration in aquaculture effluents and its concentration in water, over the maximum limit, can lead to algae blooms which produce harmful algal toxins.

Integrated multi-trophic aquaculture (IMTA) is considered as a solution for improving the sustainability of aquaculture production systems. The use of aquaponic techniques which consists in the integration of both fish and plants production, can be considered a solution in order to achieve the zero-discharge desideratum. However, according to several studies (Delaide et al., 2017; Groenveld et al., 2019; Jaeger et al., 2019), in most recirculating aquaponic systems, during some periods, up to 20% of wastewater is discharged to maintain water quality. Aquaponics ensure both water treatment by bio and phyto-remediation and improve the fish farm economical performances as a result of commercializing a secondary production - plant biomass. Phosphorus is considered, according to Yang and Kim (2020), a key nutrient that affects agricultural productivity. Also, Daniel et al. (1998) characterized phosphorus as the second most frequently limiting macronutrient for plant growth, making up about 0.2% of a plant's dry weight. Therefore, a balance between phosphorus inputs and outputs must be

assured within an IMTA system based on aquaponics techniques in order to assure both an optimum fish and plants production performance, correlated with low phosphorus discharges. However, some previous studies related to phosphorus dynamics in an IMTA system based on aquaponics techniques (Seawright et al., 1998) had reported that the total amount of phosphorus recovered in fish, plants and solids exceeded the quantity provided through the administrated diet.

Barben et al. (2010) revealed that phosphorus deficiency causes stunted plant growth, whereas phosphorus excess may lead to antagonistic interactions with micronutrients. For fish biomass, phosphorus is an important mineral in nucleic acids and cellular membranes, the main representative of the structural components of the skeletal tissues, and it is directly involved in energy processes (National Research Council, 1993).

The present research aims to identify the influence of crop culture density on phosphorus dynamics in an integrated stellate sturgeon – spinach recirculating production system, by using aquaponic substrate technique.

MATERIALS AND METHODS

Experimental design

A 44 days experimental trial was performed in order to test three spinach culture densities, in triplicate, as follows: B1H - 59 crops/m², B2H - 48 crops/m², B3H - 39 crops/m². A control variant (with no plants, only substrate) was used (Figure 1), in order to estimate the spinach biomass capacity to remove phosphorus.



Figure 1: Control variant consist in aquaponic modules with no plants, only substrate - light expanded clay aggregate (LECA)

The detailed description of integrated aquaponic system is presented in a previous study (Petrea et al., 2014). Therefore, the recirculating aquaculture system consists in 4 rearing units and a series of water treatment modules, as follows: sump, mechanical drum filter, UV, oxygenation unit and biological trickling filter. Aquaponic modules, in triplicate, were placed above each of the rearing units and water recirculation was continuously assured during the experimental period by using a recirculation submersible pump, placed in the rearing units, as described in previous study (Petrea et al., 2014). Before starting the experiment, the activation of biological trickling filtration unit was made as described by Petrea et al. (2014).

A number of 184 stellate sturgeons with an average weight of 170 g/exemplar was equally distributed within 4 rearing units and fed with Clasic Extra 1P-41% brute protein and 0.9% phosphorus, by applying a feeding ratio of 1.75% of total biomass weight (BW). Therefore, a daily average input of 6.9 ± 0.52 g phosphorus/day, by fish feed, was assured during the entire experimental period.

Each rearing unit was connected to 3 aquaponic modules filled with LECA, where Matador variety spinach (*Spinacia oleracea*) seedlings were placed according to culture densities previously mentioned (Figure 2).

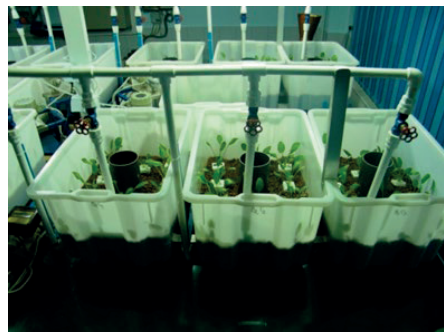


Figure 2. Spinach biomass distributed in each aquaponic module

A constant luminous power of 5800 lm, measured with TESTO 545 light meter, was assured during the entire experimental period.

The hydraulic loading rate (HLR) and hydraulic retention time (HRT) were calculated using the following equations (eq. 1 and eq. 2):

$$HLR = \frac{Q}{S} \quad (1)$$

$$HRT = \frac{S \times h \times p}{Q} \quad (2)$$

where, Q is water flow rate [m^3/s]; S is total surface area of hydroponic module [m]; h is water depth [m] and p is porosity of hydroponic module.

Thus, a constant value of 16 m/day for HLR and 0.48 min for HRT were maintained during the entire experimental period.

Sampling and analysing methods

Technological water analysis was performed by using Spectroquant Nova 400 spectrophotometer, with Merk compatible kits. Samples of water were collected once a week from both the outlet of biological filter (inlet of hydroponic units) and the outlet of each hydroponic unit.

The phosphorus removal rates for each experimental variant were presented as average of the triplicate aquaponic units. The following equation was used in order to determine phosphorus removal rates (eq. 3):

$$PR = \left[\frac{Q}{V} \times (C_{in} - C_{out}) - \frac{\Delta C_{out}}{\Delta t} \right] \times d \quad (3)$$

where PR is phosphorus removal rate ($g/m^2/day$), Q is the flow rate (m^3/day), V is the system volume (m^3), C is the concentration of phosphorus (g/m^3), d is the water depth (m) and t is the time (days).

The fish faeces were collected by using a EHEIM water vacuum cleaner provided with a mesh compartment for solids retention, both at the beginning and at the end, but also during the experimental period (2 intermediary determinations). In order to estimate the phosphorus retention in muscle tissues during the experimental trial, samples were collected both at the beginning and at the end of the experimental period. Phosphorus concentration in spinach was determined at the end of the experimental trial, for each of the experimental variant triplicate (B1H1, B1H2, B1H3, B2H1, B2H2, B2H3, B3H1, B3H2, B3H3). The results are presented as 5 samplings average. For determining the phosphorus concentration in fish muscle tissues, spinach and fish faeces, the SR ISO 2294:2009 reference method was used.

Statistical methods

The software IBM SPSS Statistics 20 for Windows was used for the statistical analysis presented in present paper.

The T test ($\alpha = 0.05$) was applied in order to identify the statistical differences between treatments, after the Kolmogorov-Smirnov normality test was performed. The ANOVA test (post-hoc Duncan test) was performed in order to compare variants.

RESULTS AND DISCUSSIONS

Technological water P_2O_5 concentration

The evolution of water P_2O_5 concentration at the inlet and outlet of both mechanical drum filter and biological trickling filter revealed a decrease trend until the end of the second week of the trial (Figure 3). However, a small increase of P_2O_5 concentration can be observed during the last week of the experimental period. This can be due to the accumulation of fish wastes within the integrated system since, according to some studies (Prüter et al., 2020), fish wastes can contain up to 1.7% phosphorus.

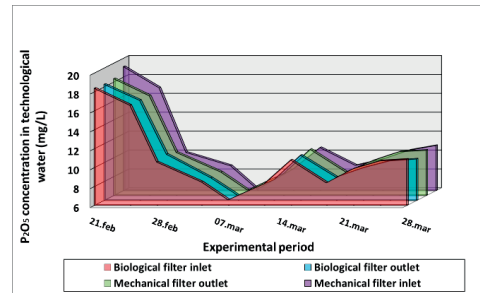


Figure 3. The dynamics of water concentration of P_2O_5 at the inlet and outlet of both mechanical drum and biological trickling filters

According to the integrated system description, the biological trickling filter outlet sampling point can be considered as inlet for all aquaponics modules. The water concentration of P_2O_5 registered in the aquaponics modules sampling points is presented, as triplicate average, in Figure 4.

Therefore, it can be observed that the entire experimental period average concentration of water P_2O_5 was 10.8 ± 3.85 mg/L at the aquaponic units' inlet, while at the outlet the following average concentrations were recorded: 9.52 ± 4.2 mg/L at B1H, 9.49 ± 3.95

mg/L at B2H, 9.72 ± 3.94 mg/L at B3H and 10.81 ± 3.86 mg/L at B4H (control variant), respectively.

The entire experimental period average concentration of water P_2O_5 recorded at the mechanical filter outlet is 10.86 ± 3.86 mg/L, while at biological filter inlet and outlet concentrations of 10.89 ± 3.57 mg/L, 10.75 ± 3.58 mg/L, respectively, were registered.

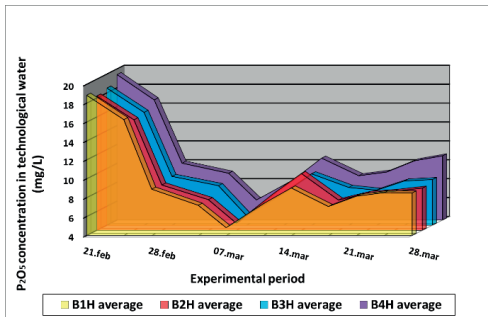


Figure 4. The dynamics of water concentration of P_2O_5 at the aquaponic modules outlet (presented as triplicate average)

The P_2O_5 concentration in fish wastes and fish muscle tissue

The entire experimental period average concentration of P_2O_5 recorded in fish wastes is 5.47 ± 1.01 g% dry weight (DW). The evolution is related to fish necessity for this nutrient, during their growth period and indicates a decrease trend during the first two weeks of the trial, follow by an upward trend until the end of the trial (Figure 5).

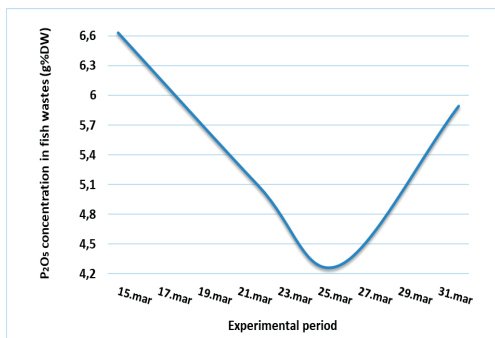


Figure 5. The dynamics of P_2O_5 concentration in fish wastes

However, it is highlighted that, according to some authors (Olson, 1992; Westerman et al., 1993; Naylor et al., 1999), the fish wastes

chemical composition may differ significantly, mostly depending on fish metabolism and its body capacity to absorb nutrients. Also, Naylor et al. (1999) stated that a comparison between fish wastes chemical concentration, reported in different studies, is difficult to be performed due to differences between wastes separation time and sampling method.

There is the need of synchronizing both fish and plants biomass in terms of P_2O_5 requirements dynamics, in order to maintain the bio-phytoremediation capacity of the integrated system.

An increase of average P_2O_5 concentration in fish muscle tissue was observed at the end (194.62 ± 12.15 mg/100 g fresh weight - FW), compared to the beginning of the experimental trial (187.92 ± 11.49 mg/100 g FW). Lazzari et al. (2008) pointed out that fish can absorb phosphorus from the water, but due to the low waterborne concentration of this compound, dietary supplementation is necessary. Also, Jahan et al. (2002) revealed that phosphorus retention is also directly correlated with fish growth rate.

The P_2O_5 concentration in spinach leaves

At the end of the trial, the following average P_2O_5 concentrations in spinach leaves were recorded, in triplicate: B1H (B1H1 - 51.09 ± 2.78 mg/100 g fresh weight (FW); B1H2 - 52.39 ± 1.19 mg/100 g FW; B1H3 - 52.57 ± 1.49 mg/100 g FW), B2H (B2H1 - 60.83 ± 1.38 mg/100 g FW; B2H2 - 59.95 ± 1.68 mg/100 g FW; B2H3 - 58.36 ± 1.42 mg/100 g FW) and B3H (B3H1 - 72.44 ± 1.54 mg/100 g FW; B3H2 - 74.73 ± 0.77 mg/100 g FW; B3H3 - 71.6 ± 1.35 mg/100 g FW) (Figure 6).

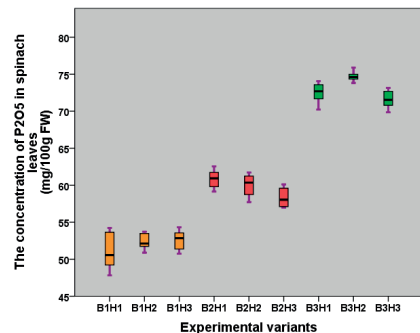


Figure 6. The P_2O_5 concentration in spinach leaves, for each experimental variant, in triplicate

Thus, statistically significant differences ($p < 0.05$) are recorded between the experimental variants. The registered concentrations are superior to those recorded in spinach leaves (29-37 mg/100 g FW) by other authors (Kaya et al., 2001) [252].

The results (Figure 6) emphasizes that the culture density can significantly influence the P_2O_5 concentration of spinach leaves. In order to assure a high phytoremediation capacity of the integrated system, spinach biomass welfare must be maintained at an optimum level. However, signs of stress among spinach biomass were observed at B1H experimental variant, most probably due to high culture density applied (Figure 7).



Figure 7. Visible signs of stress recorded among spinach plants cultured in B1H experimental variant

The P_2O_5 removal rate

The P_2O_5 removal rate had registered a general upward trend in case of B1H, B2H and B3H experimental variants, while a relatively constant trend is revealed at control variant (B4H) (Figure 8).

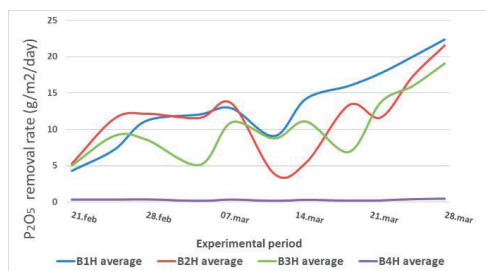


Figure 8. The P_2O_5 removal rate as triplicate average

It can be observed that B2H registered the highest values in terms of P_2O_5 removal rate in the first part of the experimental period (figure

8). However, after the first half of the trial, a considerable decrease of removal rate value is observed (figure 8), most probably due satiety effect in terms of phosphorus. The highest average value for P_2O_5 removal rate is recorded at B1H (13.38 ± 5.45 g/m²/day), followed by B2H (11.57 ± 5.25 g/m²/day), B3H (10.39 ± 4.4 g/m²/day) and B4H – control variant (0.29 ± 0.09 g/m²/day). The distribution of P_2O_5 removal rate values, for each of the experimental variant triplicate, is presented in figure 9.

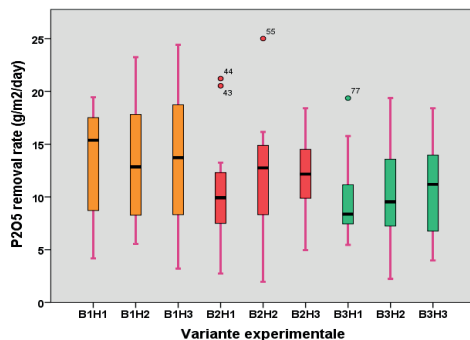


Figure 9. The distribution of P_2O_5 removal rate values, for each of the experimental variant triplicate

Statistically significant differences ($p < 0.05$) were recorded between the first three experimental variants (B1H, B2H, B3H) and control variant, as well as between B1H and the rest of the experimental variants. No statistically differences ($p > 0.05$) were recorded between the triplicate of each experimental variant.

CONCLUSIONS

The present study revealed that substrate aquaponics techniques is efficient in terms of effluent phosphorus removal from an integrated stellate sturgeon - spinach production system.

Best phytoremediation results were recorded at the experimental variant B1H, where the highest spinach culture density was applied (59 crops/m²). However, within this variant some spinach plants presented signs of stress, fact that can affect the phytoremediation capacity during a longer experimental period.

Future research must be made in terms of fish feeding rates, fish-plants stocking densities optimisation and, also, the optimization between fish rearing stage and plants stocking density, in order to balance the integrated

system. It is recommended to consider a CPS (conveyor production system) in order to balance the integrated system in terms of phosphorus availability.

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