

Tilapia rearing with septic tank-high rate algal pond effluent: domestic wastewater polishing treatment and resource recovery

Cultivo de tilapia con efluente de tanque séptico-laguna de alta tasa: pulimento para tratamiento de aguas residuales domésticas y recuperación de recursos

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Abstract

In a pilot-scale experiment carried out in Brazil, genetically improved farmed tilapia juveniles were reared in septic tank-high rate algal pond (HRAP) effluent. The combination of three total ammonia nitrogen (TAN) surface loading rates (0.6; 1.2 and 2.4 kg TAN.ha⁻¹.d⁻¹) and three fish stocking densities (3; 6 and 12 fish.m⁻²) was evaluated during a 22-week research. The fish rearing tanks worked as wastewater treatment polishing units, adding (as the best results, achieved with the lowest fish stocking density and 1.2 kg TAN.ha⁻¹.d⁻¹) the following removal figures on top of those achieved at the HRAP: 78.3% total Kjeldahl nitrogen; 89.1% ammonia nitrogen; 63.9% total phosphorous; 57.2% chemical oxygen demand; 2.36 log units E. coli. Fish productivity was estimated at 2.67 ton.ha⁻¹ for fish culture over six month per year in a temperate climate region, using the domestic treated wastewater natural plankton population as the only food source.

Resumen

En un experimento a escala piloto desarrollado en Brasil, se cultivaron juveniles de tilapia genéticamente mejorada en efluente de tanque séptico seguido de laguna de alta tasa (LAT). Durante las 22 semanas de la investigación se evaluó la combinación de tres tasas de aplicación superficial de nitrógeno amoniacal total (NAT) (0,6; 1,2 y 2,4 kg NAT.ha⁻¹.d⁻¹) y tres densidades de cultivo (3; 6 y 12 peces.m⁻²). Los tanques de cultivo funcionaron como unidades de pulimento para tratamiento de las aguas residuales, adicionando (con los mejores resultados, alcanzados con la menor densidad de cultivo y 1,2 kg NAT.ha⁻¹.d⁻¹) las siguientes eficiencias de remoción adicionales a las obtenidas mediante el uso de la LAT: 78,3% para nitrógeno total Kjeldahl; 89,1% para nitrógeno amoniacal; 63,9% para fósforo total; 57,2% para demanda química de oxígeno; 2,36 unidades logarítmicas para E. coli. La productividad de los peces se estimó del orden de 2,67 ton.ha⁻¹ para cultivo durante seis meses por año para climas templados, utilizando la población de plancton natural del agua residual doméstica tratada como la única fuente de alimento.

Keywords: High rate algal ponds, Septic tank, Sewage-fed fish culture, Tilapia rearing, Water reuse

Palabras clave: Lagunas de alta tasa, Tanque séptico, Piscicultura con aguas residuales, Cultivo de tilapia, Reúso de agua

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Why was it carried out?

We made this research in order to confirmed the feasibility of genetically improved farmed tilapia juveniles rearing in tanks using septic tank-high rate algal pond effluent and to demonstrate the performance of the culture tanks as polishing units over warmer temperatures.

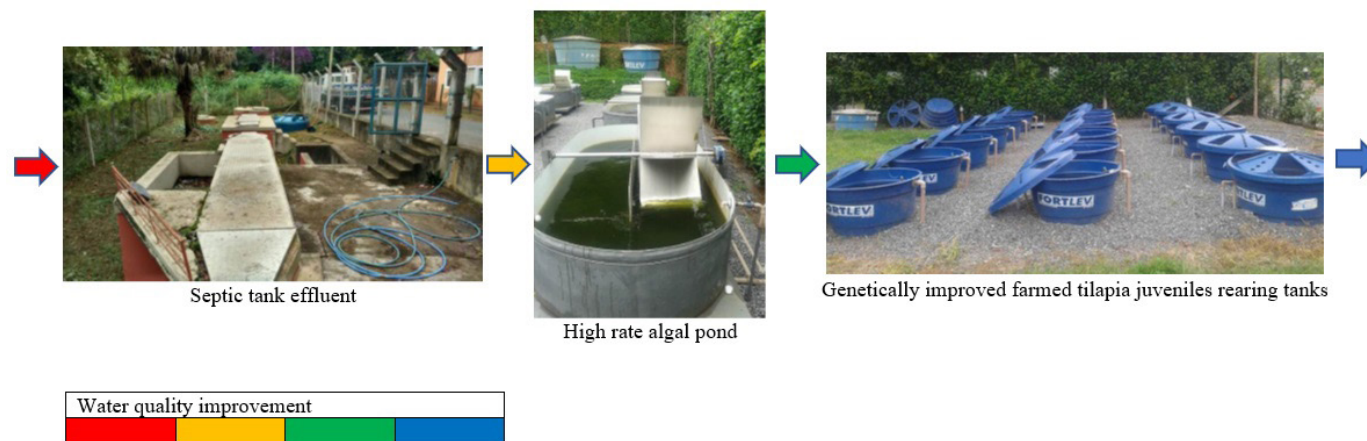
What were the most relevant results?

The fish rearing tanks worked as wastewater treatment polishing units that added, as the best results, the following removal percentages on top of those achieved at the high rate algal pond: 78.3% total Kjeldahl nitrogen; 89.1% ammonia nitrogen; 63.9% total phosphorous; 57.2% chemical oxygen demand; 2.36 log units E. coli.

What do these results provide?

The fish culture tanks offer an interesting alternative for production of an animal source of protein for different purposes and for improving the wastewater quality in terms of nutrients, organic matter and E. coli.

Graphical Abstract



Introduction

Septic tank is most commonly used for pre-treatment of domestic sewage in on-site applications. The basic function of the septic tank is to separate sludge, effluent and scum layer of the domestic water. It removes a portion of settleable solids by retention and organic matter by partial anaerobic digestion from the waste water (1). The effluents from septic tank still contain abundant ammonium and phosphate and might be favourable to microalgae cultivation compared to using raw wastewater (2). High rate algal ponds (HRAPs) are shallow, mixed systems consisting of a series of interconnecting baffled channels. Mixing by paddlewheel avoids thermal stratification and produces a homogenous chemical environment within the pond. This environment is conducive to high rates of algal photosynthesis and consequently more rapid treatment, reduced land area requirements and capital cost for construction compared with the deeper unmixed waste stabilisation ponds (3). HRAP are developed for the treatment of wastewater and resource recovery as harvestable algal/bacterial biomass for beneficial use as fertilizer, feed or biofuel (4).

The practice of resources recovering from using wastewater, e.g in wastewater-fed aquaculture, not only reduces the pollution load into water bodies but also makes a continuous system of food production (5, 6). Tilapia is one of the most important fish in freshwater aquaculture, and is reported to be the second most important group of farmed fin fish just after carps (7, 8).

In pilot experiments in Brazil, Sánchez *et al.* (9) suggested that fish rearing tanks with tilapia juveniles using septic tank-HRAP effluent with fish stocking density of 6 fish per m² and ammonia surface loading rate of 1.2 kg·ha⁻¹·d⁻¹ was feasible. At low and medium environmental temperatures the rearing tanks worked as wastewater polishing units adding the following average removal figures: 63% of total Kjeldahl nitrogen; 54% of ammonia nitrogen; 42% of total phosphorus; 37% of chemical oxygen demand; 1.1 log units of *Escherichia coli*. However, those authors concluded that feasibility of tilapia juveniles rearing and better performance of the culture tanks as polishing units still need to be confirmed over warmer temperatures than those tested then. This is essentially the objective of the present paper, which, in a way, follows up the work of Sánchez *et al.* (9).

Materials and methods

This study was carried out in Viçosa, State of Minas Gerais, Brazil (latitude: 20°45'14''S; longitude: 42°52'54''W; average altitude = 648 m), at an experimental site in the Federal University of Viçosa. The town presents a humid subtropical climate (according to the Köppen classification), with average, maximum and minimum temperatures of 19.8°C, 32.4°C and 7.2°C, respectively; annual average relative humidity and precipitation of, respectively, 81%, and 1221.4 mm - spread over rainy season (spring-summer) and dry season (autumn-winter).

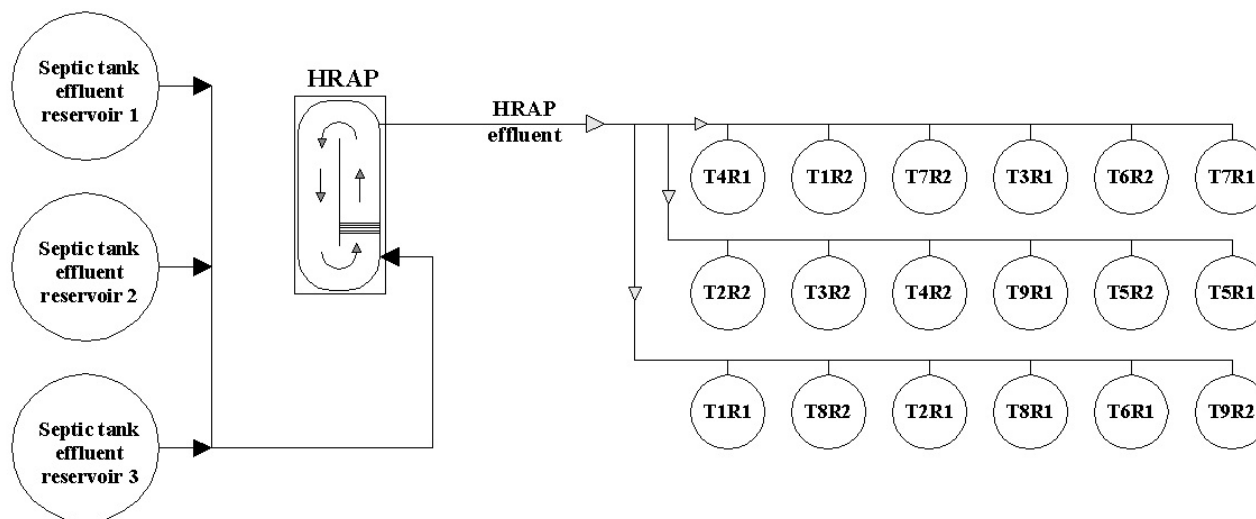
Domestic sewage was collected fortnightly, using a submersible pump, from a septic tank of a local small wastewater treatment plant. This effluent was transported to the experimental site, where it was transferred to 3000 and 5000 L reservoirs, which fed a single loop raceway fiberglass HRAP with the following characteristics: length = 2.86 m, width = 1.28 m, surface area = 3.3 m², free board = 0.2 m, pond depth = 0.3 m,

volume = 1 m³. The HRAP water was circulated continuously by a 6 blade stainless steel paddlewheels driven by a 1 hp electric motor, whose rotation was controlled by a frequency inverter (WEG, series CFW-10) to provide a mean horizontal water velocity between 0.10 and 0.15 m.s⁻¹. The HRAP were operated at an average hydraulic retention time (HRT) of 7 days.

The HRAP effluent fed 18 fish rearing plastic tanks with 210 L as useful volume. Three groups of six tanks randomly distributed received, each, a continuous flow rate so that three different ammonia surface loading rates (SLR) were tested: SLR1 = 0.6, SLR2 = 1.2 and SLR3 = 2.4 kg TAN.ha⁻¹.d⁻¹. The lowest loading rate figure was chosen based on values published in the literature of sewage-fed fish ponds (10), and in order to evaluate the effect of higher levels two and four times the lowest value were applied. The wastewater flow applied to the rearing tanks resulted in the following average HRT: SLR1 = 55 d, SLR2 = 27.5 d, and SLR3 = 13.8 d.

Juveniles of genetically improved farmed tilapia (GIFT) were reared using three stocking densities (D1 = 2, D2 = 4 and D3 = 8 fishes per tank, corresponding respectively to 3, 6 and 12 fishes per m²). D1 was based on suggestions from Edwards *et al.* (11) and Bastos *et al.* (10); D2 and D3, twice and four times D1, were used taking into account the high phytoplankton concentration of HRAP effluents. Thus, based on the combination of these two factors levels (ammonia surface loading rates and fish stocking densities), nine treatments with two repetitions were evaluated in a fully crossed factorial design: T1:SRL1-D1, T2:SRL1-D2, T3:SRL1-D3, T4:SRL2-D1, T5:SRL2-D2, T6:SRL2-D3, T7:SRL3-D1, T8:SRL3-D2, and T9:SRL3-D3 Figure 1 presents, schematically, the experimental set up.

Figure 1. Sewage reservoirs, HRAP and fish rearing tanks.



Source: Authors own creation

The wastewater rearing tanks were stocked with GIFT tilapia juveniles with an average initial weight of 7.55 ± 0.58 g. The fishes were previously reared in a recirculating aquaculture system and fed with commercial fish feed with 34% dietary protein.

Fish were weighed at the beginning and at the end of the experiment in order to calculate the average total weight gain for each treatment, as well as the daily weight gain (total weight gain/duration of the experiment). Fish tanks were monitored twice

a day in order to monitor fish mortality, and to remove and weigh dead animals. Both weight gain and mortality data met the assumptions of normality and homogeneity, thus the parametric test of analysis of variance was applied to look at differences between treatments using the software R 3.1.0.

During the experiment, 2 L-samples were taken from the HRAP and from each fish tank to measure the following physical and chemical variables according to the recommendations of APHA *et al.* (12) (the methods used are within brackets): COD - chemical oxygen demand (5220D), TP - total phosphorous (4500-P C), AN - ammonia nitrogen 4500 – NH₃D), TKN - total Kjeldahl nitrogen (4500-N D), DO - dissolved oxygen - membrane electrode method (4500-O G), pH - electrometric method (pH: 4500-H+ B), temperature (2550 B); TS - total solids (2540 D), FS – fixed and VS- volatile solids (2540 E), SS- suspended solids (2540 B). pH, DO and temperature were monitored using a Hach portable meter, model HQ40d; the chromogenic-fluorogenic method (Colilert®) was used to measure *E. coli* levels (enzyme substrate coliform test: 9323). Chlorophyll-*a* was extracted with 80% ethanol and measured by spectrophotometry (12).

COD, TP, AN, TKN, DO, Chlorophyll-*a*, pH, temperature and *E. coli* were monitored fortnightly in the septic tank and HRAP effluents, as well as in the 18 fish tanks (composite samples of each SLR treatment during the day of collection and analysis). Temperature, pH and DO were measured twice a day in the HRAP and in every fish culture tank. During the third month of the experiment, TS, FS, VS and SS were measured once in the fish tanks (again, composite samples of each SLR treatment during the day of collection and analysis). In addition, following events of total mortality, measurements of TS, FS, VS, SS and COD were carried out in tanks without fish and the respective replicate with fish; simultaneously, mosquito larvae and pupae were counted in triplicate in 2 L samples of these same tanks.

The productivity of GIFT tilapia cultured in the HRAP effluent fed tanks was calculated using Equation 1.

$$\text{Prod.} = \left(\frac{\text{IW} + (\text{WG} * t)}{1000} \right) * (\text{SD} * (1 - \text{M}/100) * \text{cf}) \quad \text{Eq. (1)}$$

Prod.: productivity (kg.ha⁻¹.yr⁻¹)

IW: average fish initial weight (g)

WG: weight gain per time unit (g.d⁻¹)

t: rearing period of time (d)

SD: stocking density (fish.m⁻²)

M: average mortality (%)

cf: conversion factor from m² to hectares = 10,000

Results and discussion

Table 1 shows the average and standard deviations values of TKN, AN, TP, COD, *E. coli* and chlorophyll-*a* found in the septic tank and HRAP effluents, and in the fish rearing tanks. The table also presents the individual and accumulated removal efficiencies calculated for the HRAP compared the septic tank effluent and for the different HLR tanks compared to the HRAP effluent.

Table 1. Average and standard deviation values of TKN, AN, TP, COD, *E. coli* and Chlorophyll-*a* in the septic tank and HRAP effluents, and in the rearing tanks; removal efficiencies individual and accumulated (within brackets).

Treatment or rearing unit	TKN (mg.L ⁻¹)	Removal efficiency (%)	AN (mg.L ⁻¹)	Removal efficiency (%)
Septic tank	114.80 ± 31.41	---	121.70 ± 36.91	---
HRAP	29.90 ± 9.66	73.90	30.40 ± 12.58	75.00
SLR1	6.50 ± 1.51	78.30 (94.30)	3.31 ± 2.71	89.10 (97.30)
SLR2	8.80 ± 2.97	70.60 (92.30)	5.80 ± 5.79	80.90 (95.20)
SLR3	11.90 ± 4.79	60.20 (89.60)	8.10 ± 8.55	73.30 (93.30)
Treatment or rearing unit	TP (mg.L ⁻¹)	Removal efficiency (%)	<i>E. coli</i> (MPN.100mL ⁻¹)	Removal efficiency (Log units)
Septic tank	12.50 ± 1.91	---	9.60 x10 ⁵ ± 1.24 x10 ⁶	---
HRAP	10.80 ± 2.07	13.60	3.70 x10 ³ ± 3.62 x 10 ³	2.41
SLR1	3.90 ± 1.43	63.90 (68.80)	1.60 x 10 ¹ ± 1.33 x 10 ¹	2.36 (4.78)
SLR2	5.00 ± 2.22	53.70 (60.00)	3.40 x10 ¹ ± 3.59 x 10 ¹	2.04 (4.45)
SLR3	6.80 ± 2.72	37.00 (45.60)	2.90 x10 ² ± 5.17 x 10 ²	1.11 (3.52)
Treatment or rearing unit	COD (mg.L ⁻¹)	Removal efficiency (%)	Chlorophyll- <i>a</i> (µg.L ⁻¹)	Concentration Increase (%)
Septic tank	361.00 ± 235.01	---	---	---
HRAP	180.90 ± 83.24	49.90	619.60 ± 266.36	---
SLR1	77.50 ± 28.95	57.20 (78.50)	156.90 ± 91.38	-74.70
SLR2	92.10 ± 34.64	49.10 (74.50)	259.60 ± 184.19	- 58.10
SLR3	101.00 ± 35.22	44.20 (72.00)	306.70 ± 193.23	- 50.50

Source: Authors own creation

The concentrations of Chlorophyll-*a* varied widely in the HRAP effluent, from 316 to 1,003.90 µg.L⁻¹, as well as in the SLR: 11.7 to 293.70 µg.L⁻¹ in SLR1; 22.90 to 469.90 µg.L⁻¹ in SLR2, and 56.10 to 534.00 µg.L⁻¹ in SLR3.

Overall, the removal efficiencies recorded in the HRAP (Table 1) were in accordance with figures usually reported in the literature, e.g. Young *et al.* (13). More specifically, the AN removal in the HRAP was similar to that reported by Assemany *et al.* (14) in the same place in Brazil and in a HRAP with similar geometric and hydraulic characteristics. Clearly, and as previously reported by Sánchez *et al.* (9), the fish rearing tanks worked as polishing units, with the highest removal efficiencies being recorded with surface loading rate of $0.60 \text{ kg TAN}\cdot\text{ha}^{-1}\cdot\text{d}^{-1}$, probably due to the high hydraulic retention time.

Table 2 shows the average values of total suspended solids (TSS), fixed suspended solids (FSS), volatile suspended solids (VSS) and COD in the effluents of tanks with and without fish (after events of total fish mortality).

Table 2. Average values of TSS, FSS and VSS and COD in tanks without (wo) and with (w) fish

Treatments	TSS (mg.L ⁻¹)	FSS (mg.L ⁻¹)	VSS (mg.L ⁻¹)	COD (mg.L ⁻¹)
T2 (SLR1-D2) wo	0.14	0.01	0.13	190.35
T2 (SLR1-D2) w	0.09	0.01	0.08	145.95
T3 (SLR1-D3) wo	0.15	0.01	0.14	266.47
T3 (SLR1-D3) w	0.07	0.01	0.06	120.58
T9 (SLR3-D3) wo	0.11	0.01	0.10	175.13
T9 (SLR3-D3) w	0.07	0.01	0.06	135.80

Source: Authors own creation

In addition to the lower values of TSS, FSS, VSS and COD, no mosquitoes' larvae and pupa were found in tanks with fish, whereas both larvae and pupa of *Culex quinquefasciatus* were found in tanks without fish in numbers close to 230 larvae and 20-30 pupa.

Chlorophyll-*a* levels were reduced by 74.70, 58.10 and 50.50%, respectively in the SRL1, SRL2 and SRL3 tanks, probably due to phytoplankton consumption by the fish. Such an assumption is confirmed with the values of TSS, FSS, VSS and COD in the effluents of tanks without and with fish (Table 2).

As a whole, the results suggest the contribution of fish culture to further improving of the treated wastewater quality, as previously reported by Reed *et al.* (15) and Sánchez *et al.* (9) for avoiding mosquitoes breeding.

The average and standard deviation values of DO concentrations were: $7.74 \pm 2.31 \text{ mg/L}$; $9.23 \pm 3.82 \text{ mg}\cdot\text{L}^{-1}$; $10.34 \pm 4.85 \text{ mg}\cdot\text{L}^{-1}$ and $10.79 \pm 5.02 \text{ mg}\cdot\text{L}^{-1}$ in the HRAP effluent, SRL1, SRL2 and SRL3 fish tanks respectively, revealing, therefore, intense photosynthetic processes. The average and standard deviation values of pH were: 6.85 ± 0.99 ; 8.27 ± 1.47 ; 8.66 ± 1.46 and 8.73 ± 1.38 in the HRAP effluent, SRL1, SRL2 and SRL3 fish tanks respectively. The average and standard deviation values of temperature were: $23.85 \pm 3.20 \text{ }^\circ\text{C}$; $25.42 \pm 3.44 \text{ }^\circ\text{C}$; $25.68 \pm 3.50 \text{ }^\circ\text{C}$ and $25.71 \pm 3.56 \text{ }^\circ\text{C}$ in the HRAP effluent, SRL1, SRL2 and SRL3 fish tanks respectively

The average and the standard deviations values of fish weight gain -in grams- were: 43.66 ± 25.65 ; 16.14^* ; 3.54^* ; 117.51^* ; 20.04 ± 11.17 ; 4.82 ± 0.03 ; 19.83^* ; 11.47 ± 6.15 ; 15.89^* in the T1, T2, T3, T4, T5, T6, T7, T8 and T9 treatments respectively. (*) Standard deviation values were not reported due to total mortality of fish in one of the replicates. Significant differences were found between treatments.

Productivity of GIFT tilapia was estimated at $2.67 \text{ ton}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$; calculated on the following basis: (i) SLR = $1.2 \text{ kg TAN}\cdot\text{ha}^{-1}\cdot\text{d}^{-1}$; (ii) fish weight gain = $0.84 \text{ g}\cdot\text{d}^{-1}$; (iii) average initial fish weight = 5 g; (iv) fish culture over six month per year in a temperate climate; (v) mortality rate = 43.00%, as a result of the average mortality of 50.00% recorded in treatments with $\text{SD} = 3 \text{ fish per m}^2$, and 35.40% mortality with $\text{SLR} = 1.20 \text{ kg TAN}\cdot\text{ha}^{-1}\cdot\text{d}^{-1}$.

The best weight gain values recorded here were higher than those reported for tilapia rearing in waste stabilization ponds or in sewage fed tanks by other authors, e.g.: $0.31 \text{ g}\cdot\text{d}^{-1}$ in cages inside secondary facultative and maturation ponds (16); $0.41 \text{ g}\cdot\text{d}^{-1}$ in tanks fed with waste stabilization pond effluent (17) and $0.26 \text{ g}\cdot\text{d}^{-1}$ in tanks receiving maturation ponds effluent (18).

The productivity of GIFT tilapia estimated for over six month per year in temperate climate regions as $2.67 \text{ ton}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$ is higher than the productivity for tilapia reported by Sin and Chiu (19) = $1.08 \text{ ton}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$ in maturation ponds; Silva *et al.* (20) = $1.71 \text{ ton}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$ in maturation ponds and Abdul-Rahaman *et al.* (21) = $0.27 \text{ ton}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$ in submersed cages in facultative and aerated ponds.

Conclusions

The fish rearing tanks worked as wastewater polishing units, improving the effluent water quality in terms of nitrogen, phosphorus, organic matter and *E. coli*, demonstrating better performance for warmer climate conditions. Tilapia juveniles rearing using tanks fed with HRAP effluent showed to be a feasible practice. Using the wastewater natural plankton population as the only food source supported an estimated fish productivity up to $2.67 \text{ ton}\cdot\text{ha}^{-1}\cdot\text{year}^{-1}$ in temperate climate regions. Through the microbiological analysis of farmed fish, it will be possible to evaluate, in future research, their potential as a source of safe food for human consumption.

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