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USING WASTEWATER FROM FISH FARMING FOR FERTIGATION OF LETTUCE CROP

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KEYWORDS

artesian well, irrigation, recirculation, sustainability, tilapia.

ABSTRACT

This study aimed to explore the efficacy of utilizing wastewater from fish farming tanks for fertigating lettuce crops as an alternative to inappropriate disposal methods. A randomized block experimental design was employed in a 3 x 8 factorial scheme, comprising three treatments: two varying percentages of wastewater (50% and 100%), alongside a control treatment employing only well water, with eight replications each. The trial was conducted in a field under protected cultivation, within a low tunnel-type structure. Biometric parameters of the lettuce plants were analyzed, including stem diameter, plant height, stem length, head diameter, root length, leaf fresh matter, leaf dry matter, stem fresh matter, stem dry matter, root fresh matter, root dry matter, leaf count, fresh shoot mass, and commercial productivity. Significant results were observed with the application of 100% wastewater treatment up to 20 days after transplanting (DAT), particularly for the biometric variables: plant head diameter (average 37.4 cm) and leaf count (average 13.7 units) at 36 DAT, of crisp lettuce Grand Rapids loose leaf seedlings. The reuse of wastewater during the experiment showed promising potential in replacing chemical fertilizers throughout the crop cycle. This practice facilitates the repurposing of water from fish farming, providing farmers with an alternative income source during the fish development phase, and mitigates improper wastewater disposal.

INTRODUCTION

Lettuce (*Lactuca sativa* L.), a member of the Asteraceae family, is one of the most popular and consumed vegetables globally. In Brazil, it holds a prominent position as the primary leafy vegetable marketed, cultivated extensively across the country, traditionally on small family farms. This cultivation has significant economic and social implications (Kapoulas et al., 2017).

Given Brazil's vast territorial expanse and specific climatic variations, studying and adapting cultivars for each region is essential. Currently, the main groups of lettuce cultivated in Brazil include curly, American, smooth, and others (e.g., red, mimosa, romaine), which constitute 70%, 15%, 10%, and 5% of the Brazilian market respectively (Guimarães et al., 2019).

Guedes (2017) posited that achieving high-quality vegetables can be realized through protected cultivation, a method that moderates environmental variables and offers physical protection to optimize and enhance production. This controlled environment mitigates pest and disease issues, thereby improving the productivity and quality of agricultural products. Additionally, protected cultivation permits the production of various crops under differing times and adverse weather conditions, resulting in greater profitability (Guimarães et al., 2022). Lettuce produced under protected cultivation stands out in the market for its

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superior quality, often fetching higher prices compared to conventionally grown lettuce (Silva et al., 2020).

With the mounting necessity to secure food availability and augment protein food production, aquaculture activity has intensified and expanded rapidly. Like all production processes, this expansion has precipitated several impacts, primarily concerning the inadequate disposal of untreated effluents, contributing significantly to water resource pollution (Hlordzi et al., 2020). Besides causing the degradation of natural water resources and numerous imbalances in aquatic life, aquaculture demands substantial water volumes (Cargnin & João, 2021).

Recent studies concerning water reuse in agricultural endeavours reveal a focus on food production to cater to rural populations' nutritional requirements (Lira et al., 2015; Alves et al., 2021). Moreover, this practice enhances rural livelihoods by boosting income through integrating two distinct productions. This dual-production approach is environmentally profitable as it precludes effluent discharge into natural environments or the need for wastewater treatment. Economically, it curtails costs associated with chemical fertilizers and water acquisition while also promoting enhanced production efficiency and value per unit of water used (Sátiro et al., 2022).

This study endeavours to evaluate the fertigation of lettuce crops using wastewater from fish farming. It aims to identify the optimal percentage of wastewater suitable for this cultivation, assess the crop's development without mineral fertilizers, and propose wastewater reuse as an alternative to disposing of untreated wastewater.

MATERIAL AND METHODS

The research was conducted within the premises of the experimental unit at the Universidade Federal Rural da Amazônia (UFRA) (Figure 1), facilitated by a collaboration between Campus Parauapebas and Embrapa Meio-Norte, situated in the rural coordinates 49º49'02” W 6º04'28” S. The region experiences a rainy tropical climate (AW), with substantial precipitation primarily from December to March. The average annual rainfall is approximately 1626 ± 84 mm year^{-1}, with a mean relative humidity of 78.2 ± 0.8%. The temperature within this locale ranges from a yearly average maximum of 31 to 34 ºC to a minimum of 22 to 23 ºC. According to the Köppen classification, the climate of Parauapebas, Pará, is categorized as Am type, signifying a humid tropical monsoon climate with excessive rainfall during certain months (January to March), and a dry season from June to August. The planting, growth, and harvesting of seedlings in this study were conducted from November 2020 to January 2021.

FIGURE 1. Drone image of the experimental area at UFRA

The experimental investigation commenced on September 16, 2020, with the delineation of the bed areas, comprising 4 blocks each measuring 9.00 m in length. Each block was further subdivided into six sub-blocks, cumulatively accounting for twenty-four beds with dimensions of 1.50 x 1.00 x 0.20 m and a spacing of 0.50 m, encompassing a total area of 55.00 m². The setup adhered to a randomized block design within a 3 x 8 factorial scheme, incorporating three treatments: two wastewater percentages (50% and 100%) and a control treatment utilizing only well water. Eight plants were evaluated in each repetition.

Initially, four plots of 10.00 m in length and 1.00 m in width were established. Following organic fertilization with 50 kg ha^{-1} of cured sheep manure, these plots were subdivided into subplots measuring 1.50 m in length, 1.00

m in width, and 0.20 m in height, with a spacing of 0.20 m between subplots. Soil properties such as pH, N, P, K, Ca, and Mg, among others, were maintained within the recommended ranges for lettuce cultivation. Each subplot hosted 20 plants, arranged with a spacing of 0.25 x 0.25 m, focusing on a usable area of two central rows consisting of six plants each, excluding borders.

Seedling formation employed black polyethene trays with 128 cells, utilizing açaí seed as a substrate. The composting process for this substrate commenced in August 2019, with an air-drying phase followed by grinding until particle sizes ranged between 1 and 5 mm in diameter. Exposure to microbial activity and oxygen facilitated decomposition, conducted in a rain-protected environment to stabilize temperature via manual watering and turning, forming an inert compound.

On November 14, 2020, sowing ensued with 6 to 10 seeds per cell in the seedling nursery module at the experimental area - Embrapa on the Campus of UFRA-Parauapebas, utilizing curly lettuce seeds of the Grand Rapids variety, recognized for year-round cultivation viability (Ziech et al., 2014). Thinning occurred 16 days after sowing (DAS), leaving a singular plant per cell. Transplantation was conducted on the afternoon of December 18, 2020, at 34 DAS, with seedlings exhibiting 3 to 4 true leaves, under protected cultivation to mitigate precipitation interference during the evaluation period.

Fertigation treatments were administered in two shifts, early morning and late afternoon, using a manual watering can, delivering a water depth of around 5 mm in each subplot until 20 days after transplanting (DAT). This was due to the winter season in the North Region, albeit with minimal thermal variation. Wastewater was sourced from a suspended pond with a recirculation system housing 5000 L of water and 172 tilapia (*Oreochromis* sp.), totalling an average biomass of around 9516 g. Wastewater parameters such as temperature, pH, nitrite, and ammonia were evaluated at the outset and after fifteen days, confirming suitability for the aquatic organisms used (Table 1). The experimental duration spanned 120 days from lettuce sowing to harvest.

TABLE 1. Physical and chemical parameters of water in fish farming tank during the experiment.

Period	Temperature (ºC)	pH	Nitrite (NO_2^-) ppm	Ammonia (NH_3) ppm
12/16/2020	28.00	7.00	0.00	0.50
12/31/2020	28.00	7.00	0.00	1.00

The entire field experiment lasted until 36 days after transplanting (DAT), with the crops receiving irrigation solely from well water following the initial 20-day development period. Upon analyzing the well water, its pH, nutrient contents, and conductivity were all found to be indicative of clean, drinkable water, free from impurities. Weeding was performed twice, and no control measures were taken for pests and diseases.

The variables studied included:

• Stem Diameter (SD, mm): Measured at the mid-point using a digital calliper, post removal of all leaves.

• Plant Height (PH, cm): Measured from the ground level to the plant apex using a graduated ruler.

• Stem Length (SL, cm): Measured from the base to the apex using a graduated ruler, post removal of all leaves.

• Plant Head Diameter (PHD, cm): Initially measured in the field across the bed length using a graduated ruler. Post-harvest, the more robustly rooted plants were cleaned with running water to remove coarse soil particles before the following measurements were taken:

• Root Length (RL, cm): Measured from the base to the terminal apex of the root using a graduated ruler.

• Leaf Fresh Matter (LFM): Post leaf count, they were weighed, and the weight was recorded in grams (g) using a precision scale accurate to 0.1 g, with the stem excluded.

• Leaf Dry Matter (LDM, g).

• Stem Fresh Matter (SFM, g): After separating from the aerial part at the collar region and removing the leaves, the composite weight of the samples per treatment repetition was measured using a precision scale (0.1 g).

• Stem Dry Matter (SDM, g).

• Root Fresh Matter (RFM, g): Lastly, the roots from each treatment repetition were measured as a composite sample.

• Root Dry Matter (RDM, g).

• Number of Leaves (NL, dimensionless): Counted from leaves larger than 3 cm, devoid of visual damage that could compromise commercialization.

• Fresh Shoot Mass (FSM, g): Obtained by weighing only the aerial part (stem + leaves), with leaves larger than 3 cm and without visual defects, discarding the roots at the collar region. For drying the samples, paper bags of known volume (2 kg) were utilized, wherein they were separated and identified according to the treatment and repetition. The drying process employed a forced-air circulation oven at 55 ºC, and the samples remained therein until a constant weight was achieved over 72 hours (Barbosa, 2017). An analytical scale with four digits and a precision of 0.001g was utilized for the quantitative determination of dry samples.

• Commercial Productivity (CP, kg m-2) (Souza et al., 2022); Calculated using [eq. (1)]:

$$CP = (LS \times SP) \times (FSM) \quad (1)$$

Where:

LS represents Line Spacing (m);

SP represents Spacing Between Plants (m), and

FSM represents Fresh Shoot Mass (g).

The obtained results were subjected to an analysis of variance, and upon verifying significance at a 5% probability, Tukey's test was employed for means comparison, facilitated using SISVAR software (Ferreira, 2019).

RESULTS AND DISCUSSION

Upon completing the analyses, results for each variable were described separately as follows:

Stem diameter (SD; mm)

Analyzing the lettuce growth variables serves as a foundational reference in the study of plant species production. Such analyses often require information easily accessible without sophisticated equipment (Demartelaere et al., 2020). An example of this is the stem diameter, which is derived from the phenological development of plants.

The findings indicate that treating with 100% wastewater (R) yielded the largest stem diameter, measuring up to 1.8 cm. Interestingly, this result was statistically comparable to that of plants treated solely with well water (A). Furthermore, the A+R treatment also yielded similar results (Table 2).

TABLE 2. Stem diameter (SD), plant height (PH), stem length (SL), plant head diameter (PHD), and root length (RL) of lettuce plants (*Lactuca sativa* L.) irrigated with different concentrations of fish farming wastewater.

Treatment	SD (mm)	PH (cm)	SL (cm)	PHD (cm)	RL (cm)
A	1.69 ab	21.30 a	8.40 a	34.20 b	12.20 a
A + R	1.62 b	20.00 a	7.90 a	35.30 b	13.30 a
R	1.80 a	20.90 a	9.30 a	37.40 a	12.60 a
CV (%)	7.39	7.90	15.65	4.14	8.01

A - Irrigation with 100% water; A + R - irrigation with 50% water and 50% fish farming wastewater; R - irrigation with 100% fish farming wastewater. Means followed by the same letter within a column do not differ from each other by Tukey's test at 5% probability.

Plant height (PH; cm)

No significant difference ($p>0.05$) was observed among treatments for plant height (Table 2). However, the average attained in this study surpassed that reported by Maia et al. (2008), who evaluated the use of residual water in biofertilizer formulation for the curly cultivar "Lucy Brow." In their study, the highest recorded plant height was 17.49 cm, whereas, in ours, a height of 21.3 cm was achieved with well water alone (A). This elevation in plant height may be attributed to the type of coverage, which encouraged plants to grow taller in pursuit of optimal solar radiation exposure.

Guimarães et al. (2022), exploring the impact of different greenhouse coverings on lettuce growth, discovered that certain varieties exhibited enhanced growth under transparent plastic coverage. Conversely, blue hue coverage led to an incidence of etiolated plants. These findings resonate with our study, underscoring the influence of coverage on lettuce development.

Stem length (SL; cm)

No significant difference was observed for stem length (Table 2), a phenomenon potentially linked to the partial shading induced by plastic film, which may have favoured etiolation. As noted by Radin et al. (2004), cultivation in a greenhouse can reduce solar radiation exposure by up to 30%. Though stem lengths of up to 6 cm are recommended, they can extend up to 9 cm (Resende et al., 2017). Notably, in this study, a stem length of 9.3 cm was achieved under treatment (R).

Plant head diameter (PHD; cm)

The analysis revealed that a concentration of 100% residual water (R) had a significant effect on the head diameter among the treatments ($p<0.05$), as depicted in Table 2. Specifically, twenty-day usage of this treatment led to better development, with the plant head diameter averaging 37.4 cm—a phase indicative of heightened nutrient absorption in the culture (Trani, 2014). This underscores the potent influence of macronutrients such as nitrogen (N) in solution during the critical period of the crop cycle, favouring vegetative growth, mass accumulation, and increased leaf area (Oliveira et al., 2020).

In line with these findings, Maia et al. (2008) observed positive outcomes concerning this variable while evaluating the impact of biofertilizers prepared with nursery water. Similarly, Fiorini et al. (2016) demonstrated significant results when assessing cultivars across two seasons under organic management in Baixada Fluminense, highlighting the seasonal influence on the development of plant head diameter (PHD).

Supporting this observation, Torres et al. (2023) found in their study on lettuce and nutrients that mineral fertilizer application was more efficient than organomineral fertilizer in enhancing the production of curly lettuce within a protected and irrigated environment.

Root length (RL; cm)

No statistical difference was observed for root length among treatments (Table 2). Barbosa (2017) examined the influence of filtering elements in an aquaponics system and recorded values akin to those in this study when the filter was not used. It was noted during the experiment that a compact soil layer formed, which was alleviated through surface scarification between the plants.

Leaf fresh matter (LFM; g)

The R treatment yielded superior results, with an average of 83.2 g $plant^{-1}$, compared to the A+R treatment, with no visible nutritional deficiency in any treatment (Table 3). On the other hand, Cuba (2015), while evaluating the interaction between reused water and nutrient solution, attained a higher average of 118.57 g $plant^{-1}$. Even though this value surpassed that observed in this study for the treatment with reused sewage water alone, it was accompanied by apparent nutritional deficiency and elevated pH levels.

TABLE 3. Leaf fresh matter (LFM), leaf dry matter (LDM), stem fresh matter (SFM), stem dry matter (SDM), root fresh matter (RFM), root dry matter (RDM) of lettuce plants (*Lactuca sativa* L.) irrigated with different fish farming wastewater concentrations.

Treatment	LFM (g)	LDM (g)	SFM (g)	SDM (g)	RFM (g)	RDM (g)
A	71.60 ab	3.10 ab	15.40 a	0.48 a	3.89 a	0.23 a
A + R	69.80 b	2.95 b	14.30 a	0.45 a	4.09 a	0.26 a
R	83.20 a	3.65 a	18.40 a	0.54 a	4.29 a	0.28 a
CV (%)	15.98	14.41	24.07	20.64	10.26	15.72

A - irrigation with 100% water; A + R - irrigation with 50% water and 50% fish farming wastewater; R - irrigation with 100% fish farming wastewater. Means followed by the same letter within a column do not differ from each other by Tukey's test at 5% probability.

Leaf dry matter (LDM; g)

The findings for leaf dry matter indicate that the treatment with 100% wastewater (R) surpassed the A+R treatment (Table 3). Barbosa (2017), assessing the use of filter elements along with effluent from fish farming in aquaponics across two lettuce varieties and periods, reported values of 3.22 ± 1.33 g for the smooth variety without a notable difference between treatments. However, for the curly variety in the subsequent period, a lesser value of 0.37 ± 0.06 g was obtained, underscoring that the accumulation of solids in the roots hinders plant development.

Stem fresh matter (SFM; g)

Consistent with the LDM variable, where no significant difference between treatments was found, a similar outcome is noted here.

Stem dry matter (SDM; g)

The findings for this variable mirrored those of the SFM, showing no divergence between treatments.

Root fresh matter (RFM; g)

The mean values for this variable revealed no significant difference between treatments (Table 3). Barbosa (2017), evaluating filtering elements in lettuce of the smooth variety, achieved better values of 17.51 ± 7.20 g compared to this study. However, with the curly variety, a lesser value of 2.87 ± 1.29 g was observed when using a filter, yet unlike in this study, no phytotechnical compromise in crop development was noted.

Root dry matter (RDM), (g)

The observations for this variable align with those of the RFM, with no significant variation between treatments.

Number of leaves (NL; dimensionless)

For the number of leaves variable, treatment R yielded the highest average of 13.7 leaves, as depicted in Table 4. This was followed by treatment A+R with an average of 11.8 leaves. The treatment using only well water showed a comparable result with an average of 11.7 leaves, indicating no significant difference.

TABLE 4. Number of leaves (NL), fresh shoot mass (FSM), commercial productivity (CP) of lettuce plants (*Lactuca sativa* L.) irrigated with different fish farming wastewater concentrations.

Treatment	NL (dimensionless)	FSM (g)	CP (kg m^{-2})
A	11.70 b	87.30 ab	1.36 ab
A + R	11.80 b	84.10 b	1.26 b
R	13.70 a	101.60 a	1.71 a
CV (%)	5.68	17.05	16.94

A - irrigation with 100% water; A + R - irrigation with 50% water and 50% fish farming wastewater; R - irrigation with 100% fish farming wastewater. Means followed by the same letter within a column do not differ from each other by Tukey's test at 5% probability.

According to Brzezinski et al. (2017), the number of leaves is a crucial factor in determining the viability of a plant for consumption in natura, as the leaves constitute the marketable part of the plant. Although a greenhouse serves as a physical protective barrier, it creates a microclimate that significantly influences the growth and development of the vegetable (Rebouças et al., 2015).

Fresh shoot mass (FSM; g)

Fresh shoot mass exhibited a higher average (101.6 g $plant^{-1}$) for the treatment with 100% wastewater residue, distinguishing it from the A+R treatment (Table 4). Rezende et al. (2017), in their assessment of lettuce production, fertigated with nitrogen, silicon, and potassium using multiple linear regression analysis, identified an enhancement in the commercial FSM. Specifically, for every 100.00 mg N $plant^{-1}$ added to the soil, there was an increment of 16.20 g $plant^{-1}$. To draw a parallel, it would require 627 mg N for the 480 plants used in the experiment, summing up to more than 300 kg N. Considering urea as the most cost-effective source with 45% N, around 135 kg would be needed, translating to a cost exceeding $200 (two hundred US dollars) in Parauapebas for this nutrient alone, an expense that was avoided in this study.

The average weight in treatment R was 101.6 g, showing no significant difference when compared to other treatments. Cuba (2015), while evaluating the use of wastewater from the water treatment station, inferred that there is a necessity for nutrient supplementation to make up for the low nutrient content. However, in this study, the wastewater treatment alone achieved satisfactory yield without manifesting a deficiency throughout the lettuce development period.

Commercial productivity (CP; kg m^{-2})

The treatment with 100% wastewater (R) exhibited the highest performance for average commercial productivity, yielding an average of 1.71 kg m^{-2}, which is notably higher than the 1.26 kg m^{-2} observed in the A+R treatment (Table 4). In a comparable study, Barbosa (2017) evaluated a biofloc system with and without filters in an aquaponic system across two varieties of smooth and curly lettuce. These authors found significantly lower values (0.9 and 0.3 kg.m^{-2}) compared to the present study.

Souza et al. (2022) emphasized that full sunlight incidence leads to heavier plants, i.e., higher commercial productivity (in kg m^{-2}) compared to shaded environments. However, the same authors noted that shade screens tend to result in larger-sized lettuce plants, characterized by greater height and diameter.

Silva Peixer et al. (2019) highlighted that although annual investment in hydroponic lettuce production is substantial, their feasibility study yielded satisfactory results. The authors reported an annual revenue of approximately US $17,500, highlighting substantial promise for investment in rural areas for this type of production.

CONCLUSIONS

Using fish farming wastewater at 100% during the initial twenty days positively influences the development of lettuce across all analyzed variables, particularly the plant head diameter (37.4 cm) and the number of leaves (13.7 units). This practice can replace the use of chemical fertilizers during the growth cycle. Therefore, the proposed production system presents an additional profit avenue for entrepreneurs during the fish development period. This sustainable production practice prevents the improper disposal of water enriched with nitrogenous nutrients and orthophosphates, which are known for compromising the physical, chemical, and biological quality of river and groundwater bodies.

Employing fish farming water, under the modeled conditions, led to an over 26% increase in commercial productivity compared to solely using well water for irrigation. However, the study is constrained by a small sample size, and the evaluation period within a single season may have influenced the results. Thus, further research is recommended to continue exploring food production using wastewater. This endeavour aims to minimize environmental impacts by preventing the release of inappropriate water into springs, reusing essential nutrients, enhancing producers' income, and fostering employment opportunities in the agricultural sector.

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