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# Growth Dynamics of Lettuce (Lactuca sativa) Coupled with the Nile tilapia (Oreochromis niloticus) in a Greenhouse Aquaponic System in Morocco

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#### **ABSTRACT**

Aquaponics offers a sustainable alternative to conventional agriculture, particularly in water-scarce regions such as Morocco. This study assesses the growth and biomass production of lettuce (Lactuca sativa var. Madrilène) cultivated in a greenhouse aquaponic system integrating Oreochromis niloticus and leafy plants. The system combined nutrient film technique (NFT) and deep-water culture (DWC) modules operating in a closed recirculating loop. Eighteen lettuce seedlings, germinated in rockwool, were grown for 30 days under aquaponic conditions. Plant performance was evaluated through height, leaf number, and fresh biomass. At harvest, the mean final height reached  $22.2 \pm 2.8$ cm, with an average fresh weight of 194.1  $\pm$  15.3g per plant, yielding a total biomass of 3.49kg. The system consumed 12.5L. day<sup>-1</sup> of water, of which 3.2L. day<sup>-1</sup> corresponded to plant transpiration. These results demonstrate that lettuce can achieve commercial yields within 30 days without chemical fertilizers, confirming the technical feasibility and agronomic potential of aquaponics for sustainable food production and urban agriculture under Moroccan climatic conditions.

# INTRODUCTION

By 2030, Morocco's urban population is projected to reach 26.7 million inhabitants (68%), and according to the **OECD** (2025), this proportion could increase to 81% by 2050. This rapid demographic shift intensifies the need for innovative agricultural systems capable of ensuring urban food security while preserving natural resources. In this context, controlled-environment agriculture, and particularly urban hydroponic and aquaponic systems, have emerged as sustainable solutions to address food production challenges in water-scarce regions.

Urban agriculture offers multiple environmental and social benefits, including the reduction of food transportation emissions, local availability of fresh produce, waste valorization, and public awareness of sustainable food systems (Mougeot, 2015; FAO, 2022). However, its development in Morocco remains limited, largely due to water constraints and the dependence of conventional agriculture on chemical fertilizers.

Aquaponics represents a promising approach to overcome these limitations. It integrates aquaculture and hydroponics within a closed recirculating system, where fish effluents serve as an organic nutrient source for plants (Somerville *et al.*, 2014; Love *et al.*, 2015). This circular mechanism allows the conversion of ammonia (NH<sub>3</sub>/NH<sub>4</sub><sup>+</sup>) from fish waste into nitrate (NO<sub>3</sub><sup>-</sup>) through bacterial nitrification, enabling efficient plant nutrient uptake while maintaining water quality for aquatic organisms (Lennard, 2017; Goddek *et al.*, 2019).

Several studies have reported that aquaponics can sustain high-yield leafy vegetable production, particularly lettuce (*Lactuca sativa*), while using up to 90% less water than conventional soil cultivation (**Rakocy et al., 2006; Graber & Junge, 2009**). In the Moroccan context, **Rharrhour et al.** (2022) demonstrated that aquaponic systems can mitigate the impact of drought and soil degradation by reducing water consumption and eliminating synthetic inputs. More recently, **Rharrhour et al,** (2024) confirmed that fish effluents alone can support commercial lettuce and basil yields in small-scale systems within 25 to 30 days of cultivation.

Greenhouse aquaponics offers additional advantages by optimizing light exposure, temperature regulation, and microbial balance, resulting in improved photosynthesis and nutrient absorption (**Delaide** *et al.*, **2016**; **Goddek** *et al.*, **2019**). Lettuce, in particular, has been widely used as a bioindicator species in aquaponic studies due to its short growth cycle, sensitivity to nutrient variations, and high market demand (**Tyson** *et al.*, **2011**; **Buzby & Lin**, **2014**).

Therefore, the present study aims to evaluate the growth performance and fresh biomass yield of lettuce (*Lactuca sativa* var. Madrilène) cultivated in a greenhouse aquaponic system integrating the Nile tilapia (*Oreochromis niloticus*) under Moroccan climatic conditions. The objective is to determine the technical feasibility and agronomic potential of aquaponics as a sustainable horticultural production model for arid and semi-arid regions.

# **MATERIALS AND METHODS**

# 1. Experimental Site and System Design

The experiment was conducted from July to August 2025 in the glass greenhouse of the Faculty of Sciences, Mohammed V University, located in Rabat, Morocco (Fig. 1). The study was performed under natural light conditions, with an average ambient temperature of  $24 \pm 3$  °C and relative humidity of 65-75%.

The system corresponded to a coupled recirculating aquaponic configuration. It consisted of an 800L high-density polyethylene (HDPE) fish tank filled to 500L, connected to two 20L filtration barrels (mechanical and biological). The mechanical filter contained Sera® wool for particle removal, while the biological filter was filled with Sera® Siporax substrate to promote nitrifying bacterial colonization. The filtered effluent was collected in a sump tank and recirculated by a submersible pump (3.8W, 400L.h<sup>-1</sup>) to the hydroponic unit. Water then flowed by gravity back to the fish tank, completing the loop.

The aquaculture unit, stocked with *Oreochromis niloticus* juveniles, functioned primarily as the biological nutrient source for the plants. No synthetic fertilizers, pH stabilizers, or external nutrient solutions were added throughout the experiment.

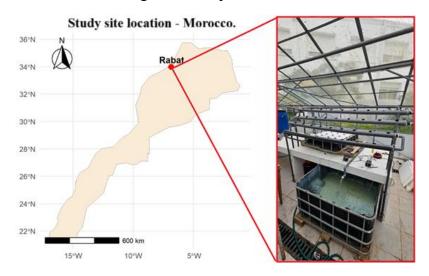


Fig. 1. Study site location and experimental aquaponic system

# 2. Hydroponic Unit and Plant Material

The hydroponic module integrated two complementary systems: a nutrient film technique (NFT) and a deep-water culture (DWC) unit (Fig. 2). The NFT consisted of four PVC tubes (175cm long, 50mm diameter), each perforated with eight planting holes spaced at 20cm intervals. The effluent from the sump was distributed evenly across the tubes, forming a thin nutrient film that flowed continuously toward the DWC compartment.



**Fig. 1.** Views of the hydroponic unit of the experimental aquaponic system: (**A**) Nutrient film technique (NFT) module, (**B**) Deep water culture (DWC)

The DWC unit was composed of a rectangular plastic basin (150L capacity) supporting three floating rafts, each with 20 openings for plant pots. This configuration allowed root systems to develop in direct contact with the nutrient solution, ensuring efficient nutrient uptake.

The plant species selected was lettuce (*Lactuca sativa* var. Madrilène), chosen for its short growth cycle and high compatibility with aquaponic systems. Seeds were germinated in rockwool cubes under greenhouse conditions. After seven days of germination, 18 uniform seedlings were transplanted into the NFT channels, where they remained for 10 days before being transferred to the DWC unit for the final 20 days of growth. The total cultivation period under aquaponic conditions was 30 days.

# 3. Water and Nutrient Monitoring

Water quality was monitored using a multiparameter probe (HI 9829, HANNA Instruments), which measured temperature (°C), pH, dissolved oxygen (DO, mg. L<sup>-1</sup>), and electrical conductivity (μS.cm<sup>-1</sup>) *in situ*. Nutrient concentrations were analyzed with a spectrophotometer following the manufacturer's protocols for ammonium (NH<sub>4</sub>+), ammonia (NH<sub>3</sub>), nitrite (NO<sub>2</sub>-), nitrate (NO<sub>3</sub>-), phosphorus (P), potassium (K), molybdenum (Mo), and copper (Cu).

The frequency of measurements varied according to the experimental phase: daily during the biological start-up period and two to three times per week after system stabilization. These data were used to assess the biogeochemical evolution of the system and its stability over time.

# 4. Plant Growth Measurements

The agronomic monitoring of lettuce (*Lactuca sativa*, Madrilène variety) focused on the evaluation of plant growth and total biomass production. The parameters recorded included the initial weight ( $W_i$ , g), initial height ( $H_i$ , cm), plant height during growth (cm), and number of leaves. At harvest (after 30 days of cultivation), the final height ( $H_f$ , cm) and final fresh weight ( $W_f$ , g) were measured.

Based on this data, several growth indicators were calculated as follows:

- Increase in plant height ( $\Delta H$ , cm):  $\Delta H = H_f H_i$ , where  $H_f$  is the final plant height measured at harvest, and  $H_i$  is the initial height measured prior to transplantation.
- Increase in total fresh biomass ( $\Delta W_{total}$ , g):  $\Delta W_{total} = W_f W_i$ , where  $H_f$  is the total final fresh biomass (g) measured at harvest, and  $W_i$  is the initial total biomass (g) of the plants, usually negligible, measured at transplantation.
- Increase in fresh biomass per plant ( $\Delta W_plant$ , g):  $\Delta W_{plant} = \frac{W_f W_i}{n}$ , where  $W_f$  is the total final fresh biomass (g) at harvest,  $W_i$  is the initial total biomass (g), and n represents the total number of plants.

These indicators were used to characterize growth dynamics and biomass accumulation throughout the aquaponic cultivation cycle, thereby linking plant performance to the physicochemical stability of the system and the productivity of the aquaculture compartment.

# 5. Statistical Analysis

All analyses were performed using RStudio (R version 4.4.2, R Core Team, 2024). Descriptive statistics were used to summarize growth and water-quality data, including means, standard deviations, and ranges. Growth curves and boxplots were produced to visualize temporal variation and data dispersion.

Relationships among variables were explored using Kendall's correlation coefficient  $(\tau)$ , suitable for non-parametric datasets.

To characterize growth dynamics, linear mixed-effects models (LMMs) were fitted to account for repeated measurements and individual variability. These models also supported simple predictive analyses of plant growth trends over time.

Finally, the principal component analysis (PCA) was implemented to identify key gradients and associations between water-quality parameters and plant growth indicators, providing an integrative overview of system performance.

### RESULTS AND DISCUSSION

# 1. Growth Dynamics of Lettuce

The growth of lettuce (*Lactuca sativa* var. Madrilène) was monitored over 40 days based on daily height measurements from 18 individual plants. This approach provided a clear view of temporal growth patterns and individual variability within the aquaponic system.

Descriptive statistics (Table 1) indicated a steady increase in mean and median plant height throughout the experiment, reflecting homogeneous development among plants. The standard deviation remained moderate during the early phase, rising slightly toward the end of the cycle, which suggests increasing variability as plants reached maturity.

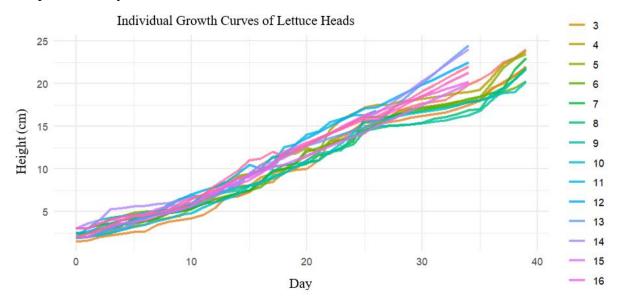
Table 1. D	Descriptive	statistics of	lettuce	growth
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Day	Mean (cm) ± SD	Min (cm)	Max (cm)	n
0	$2.32 \pm 0.48$	1.5	3	18
9	$5.37 \pm 0.66$	4	6.5	18
19	$11.0 \pm 1.14$	9.9	12.9	18
29	$16.6 \pm 0.89$	15.2	18.0	18
39	$22.2 \pm 1.34$	20.2	24	18

The individual growth curves (Fig. 3) revealed a consistent upward trend across all plants, demonstrating continuous vegetative development. During the first 10 days, plant heights ranged from 2 to 4cm, with slow growth corresponding to the adaptation and rooting phase. Between days 10 and 30, growth accelerated sharply, showing an almost linear increase among individuals, which reflected stable nutrient availability and optimal environmental conditions.

After day 30, the growth rate began to decrease, marking the onset of a physiological plateau as plants reached commercial size. Final heights ranged between 18 and 24cm, with a

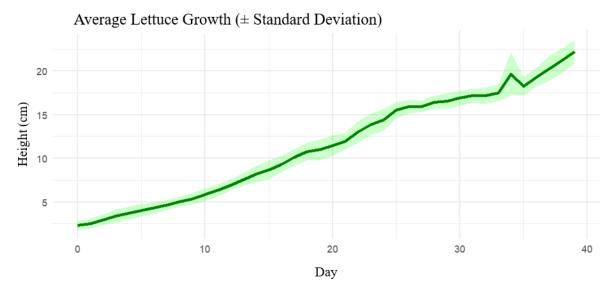
mean of  $\sim$ 21  $\pm$  3cm. The slight variability observed among individuals was likely due to microenvironmental, genetic, or competitive factors (e.g., light or nutrient access), which are common in experimental systems.



**Fig. 3.** Individual growth curves of eighteen lettuce (*Lactuca sativa* var. Madrilène) plants cultivated in the greenhouse aquaponic system over a 40-day period

Each line represents one plant's height progression, showing consistent and continuous growth across all individuals

The mean growth curve ( $\pm$  SD) confirmed this overall dynamic (Fig. 4). Lettuce height increased gradually during the first days, followed by a rapid exponential phase between days 5 and 25. Growth continued thereafter but slowed after day 30, indicating the onset of a physiological plateau. The final height reached approximately  $21 \pm 3$ cm, reflecting uniform and healthy development across all plants.



**Fig. 4.** Average growth curve of lettuce (*Lactuca sativa* var. Madrilène) cultivated in the greenhouse aquaponic system over a 40-day period. The solid green line represents the mean height, while the shaded area indicates the standard deviation among 18 plants

# 2. Modeling of Mean Height Growth

To account for the longitudinal structure of the data and the dependence between repeated measurements on the same individuals, a linear mixed-effects model (LMM) was fitted. The model assessed the effect of time (Day) on lettuce height while considering interindividual variability among plants as a random effect.

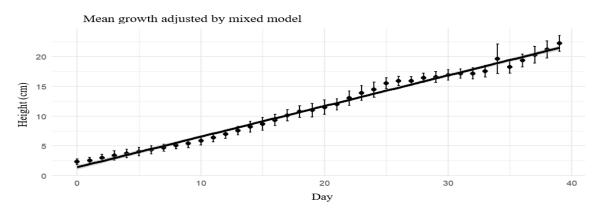
Model outputs (Table 2) indicate an estimated initial height of 1.30cm at day 0. The effect of time was highly significant, with an average daily increase of 0.53cm (t= 148.1, P< 0.001), demonstrating a consistent and sustained growth trend throughout the experiment. The inter-individual variability was low (SD  $\approx$  0.65cm), reflecting uniform growth across the 18 plants. The residual standard deviation (SD  $\approx$  0.91cm) remained moderate, confirming the model's strong ability to describe the observed growth dynamics.

**Table 2.** Results of the linear mixed-effects model (LMM) describing the effect of time (Day) on lettuce height growth

Fixed effects	Estimate (β)	Std. Error	t-value	<i>P</i> -value
Intercept	1.304	0.169	7.71	<0.001 ***
Day	0.528	0.004	148.11	<0.001 ***

Note: The model indicates a significant positive effect of time on plant height (P< 0.001), with an average daily growth rate of 0.53cm.day<sup>-1</sup>.

As shown in Fig. (5), the linear mixed-effects model reveals a strong linear relationship between time and lettuce height. The estimated slope indicates an average daily increase of 0.53 cm, reflecting a steady and consistent growth rate throughout the experimental period.



**Fig. 5.** Mean lettuce height (*Lactuca sativa* var. Madrilène) fitted by a linear mixed-effects model over the 40-day experimental period

The solid black line represents the model's predicted values, while the dots indicate observed daily means with standard error bars. The strong alignment between predicted and observed values confirms the model's accuracy and the consistent growth pattern of plants within the aquaponic system.

The observed daily means closely followed the predicted trend, confirming the excellent fit of the model. The narrow error bars illustrate low inter-individual variability and highlight the uniform development of the 18 lettuce plants. Overall, the model confirms that growth in the aquaponic system was stable and homogeneous across the 40-day monitoring period.

#### 3. Growth Indices

The results (Table 3) show a clear increase in vegetative growth throughout the experiment. The mean plant height rose from 2.32 to 22.23cm, representing an average gain of 19.9cm. In parallel, the total fresh biomass reached 3,494.2g, corresponding to an average yield of 194.1g per plant. These findings confirm uniform growth and satisfactory development of lettuce plants under the conditions of the aquaponic system.

**Table 3.** Growth indices of lettuce (Lactuca sativa var. Madrilène) cultivated in the greenhouse aquaponic system

Index	Value	Unit
Mean initial height	2,32	cm
Mean final height	22,23	cm
Mean height gain ( $\Delta H$ )	19,9	Cm
Total fresh biomass produced ( $\Delta W$ )	3 494,2	G
Average yield per plant	194,1	g/plante

# 4. Complementary Test: Kendall Correlation

Correlation analyses were conducted to examine the relationships between lettuce growth and key physico-chemical parameters of the aquaponic system (NH<sub>4</sub><sup>+</sup>, NH<sub>3</sub>, NO<sub>2</sub><sup>-</sup>, NO<sub>3</sub><sup>-</sup>, temperature, EC, and TDS). These tests aimed to identify significant associations between nitrogen cycle dynamics and plant development.

The Kendall correlation analysis revealed several significant relationships between plant height and the system's nitrogenous and physico-chemical parameters (Fig. 6). Plant height was strongly and positively correlated with ammonia (NH<sub>3</sub>) and ammonium (NH<sub>4</sub>+) concentrations ( $\tau = 0.78$ , P < 0.001), highlighting the central role of reduced nitrogen in plant nutrition and its immediate availability for root uptake.

Conversely, negative correlations were observed with nitrite (NO<sub>2</sub><sup>-</sup>,  $\tau = -0.54$ , P < 0.001) and nitrate (NO<sub>3</sub><sup>-</sup>,  $\tau = -0.58$ , P < 0.001), suggesting that higher levels of oxidized nitrogen species may coincide with a relative slowdown in growth. Temperature showed a moderate positive correlation ( $\tau = 0.37$ , P < 0.001), reflecting its favorable influence on vegetative development, possibly through indirect effects on nitrification efficiency. The interdependencies among chemical parameters confirm the expected pattern of the nitrogen cycle. A strong positive correlation between nitrate (NO<sub>3</sub><sup>-</sup>) and nitrite (NO<sub>2</sub><sup>-</sup>) ( $\tau = 0.75$ , P < 0.001) reflects the intermediate step of nitrification, while the negative relationship between ammonium (NH<sub>4</sub><sup>+</sup>) and nitrate (NO<sub>3</sub><sup>-</sup>) ( $\tau = -0.33$ , P = 0.006) indicates the progressive oxidation of ammonium into nitrates. Temperature showed a negative correlation with NO<sub>2</sub><sup>-</sup> ( $\tau = -0.60$ ,

P< 0.001), suggesting faster nitrite oxidation at higher temperatures. Additionally, electrical conductivity (EC) and total dissolved solids (TDS) were perfectly correlated with ammonia/ammonium (NH<sub>3</sub>/NH<sub>4</sub><sup>+</sup>) ( $\tau$  = 1.00, P< 0.001), confirming that the ionic load of the medium was primarily governed by reduced nitrogen species. Overall, these results demonstrate that lettuce growth is closely linked to the availability of reduced nitrogen, and that the dynamic sequence of the nitrogen cycle (NH<sub>4</sub><sup>+</sup>  $\rightarrow$  NO<sub>2</sub><sup>-</sup>  $\rightarrow$  NO<sub>3</sub><sup>-</sup>) is clearly reflected in the observed correlations.

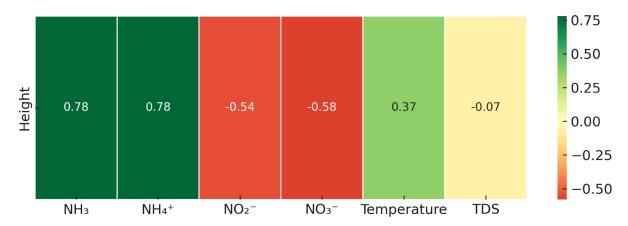


Fig. 6. Kendall correlation coefficients ( $\tau$ ) between lettuce height (*Lactuca sativa var. Madrilène*) and key physico-chemical parameters of the aquaponic system

Positive correlations (in green) indicate parameters that favor plant growth, such as ammonia (NH<sub>3</sub>), ammonium (NH<sub>4</sub><sup>+</sup>), and temperature, while negative correlations (in red) reflect inhibitory or indirect effects linked to oxidized nitrogen forms (NO<sub>2</sub><sup>-</sup>, NO<sub>3</sub><sup>-</sup>). The weak relationship with TDS suggests limited direct influence on vegetative growth.

In summary, the correlation patterns highlight the functional coupling between biogeochemical processes and plant growth within the aquaponic system. The strong dependence of lettuce development on reduced nitrogen forms (NH<sub>3</sub>/NH<sub>4</sub><sup>+</sup>) and the clear trace of nitrification dynamics reflect the system's biological equilibrium and nutrient recycling efficiency. These findings confirm that maintaining a balanced nitrogen transformation pathway is essential to sustain optimal plant productivity while ensuring water quality stability in integrated aquaponic environments.

#### **DISCUSSION**

The establishment of the aquaponic system led to a progressive stabilization of the nitrogen cycle, as reflected by the low concentrations of ammonium  $(1.13 \pm 1.15 \text{mg/ L})$  and free ammonia  $(1.07 \pm 1.09 \text{mg/ L})$ , along with the accumulation of nitrates  $(75.5 \pm 39.8 \text{mg/ L})$  and the decline of nitrite levels after transient peaks around 1.97mg/ L. This pattern indicates the successful installation of an efficient nitrifying community, consistent with reports from tropical aquaponic systems (**Rakocy** *et al.*, **2006**; **Goddek** *et al.*, **2019**). The physico-chemical conditions remained globally favorable for both compartments.

Plant growth followed a regular and sustained pattern, increasing from an average height of 2.3 to 22.2cm in 39 days ( $\approx$  20cm gain). The total fresh biomass reached 3.49kg, corresponding to an average yield of 194g/ plant, values comparable to hydroponic systems

under controlled climates (**Somerville** *et al.*, **2014**). These results confirm the good adaptation of lettuce to aquaponic conditions, supported by balanced nutrient availability and stable water quality. The linear mixed-effects model revealed a significant daily height increase of 0.53cm. day<sup>-1</sup>, confirming steady vegetative growth with low inter-individual variability.

Kendall correlations reinforced the link between plant development and nitrogen dynamics. Lettuce height showed strong positive associations with reduced nitrogen forms (NH<sub>3</sub>, NH<sub>4</sub><sup>+</sup>), while oxidized forms (NO<sub>2</sub><sup>-</sup>, NO<sub>3</sub><sup>-</sup>) were negatively correlated, indicating that excessive nitrification may coincide with a relative slowdown in growth. These relationships demonstrate that reduced nitrogen plays a central role in root nutrition and immediate nutrient uptake. Moreover, the correlations among nitrogen compounds reflect the functional structure of the nitrification process: a strong positive association between NO<sub>3</sub><sup>-</sup> and NO<sub>2</sub><sup>-</sup> ( $\tau$ = 0.75, P< 0.001) represents the intermediate stage of nitrification, while a negative correlation between NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup> ( $\tau$ = -0.33, P= 0.006) indicates the progressive oxidation of ammonium. Temperature was inversely related to NO<sub>2</sub><sup>-</sup> ( $\tau$  = -0.60, P< 0.001), suggesting faster nitrite oxidation at higher temperatures, while EC and TDS were strongly correlated with NH<sub>4</sub><sup>+</sup>/NH<sub>3</sub>, confirming that the ionic load of the medium was dominated by reduced nitrogen species.

Together, these results highlight the functional coupling between nutrient recycling and plant performance. The efficient microbial conversion of nitrogen compounds sustained both plant and fish growth while maintaining water quality, consistent with findings of **Graber and Junge (2009)** and **Delaide** *et al.* (2016). The growth of tilapia, a tenfold increase in weight (4.5 g  $\rightarrow$  47.4g in 50 days)—and the healthy development of lettuce confirm that the system achieved biological equilibrium and optimized nutrient exchange between compartments. This synergy illustrates the potential of aquaponics as a sustainable production model, capable of delivering high yields while minimizing resource use and environmental impact, especially under semi-arid urban Moroccan conditions.

#### **CONCLUSION**

This study demonstrates that a greenhouse aquaponic system integrating tilapia (Oreochromis niloticus) and lettuce (Lactuca sativa var. Madrilène) can sustain efficient nutrient recycling and stable biological performance under Moroccan climatic conditions. The results confirmed a strong coupling between nitrogen dynamics and plant growth, with reduced nitrogen forms (NH<sub>3</sub>/NH<sub>4</sub><sup>+</sup>) playing a key role in nutrient availability. Lettuce exhibited vigorous and homogeneous growth, achieving commercial size and yields comparable to those of controlled hydroponic systems. In addition, the system ensures an optimal water quality for fish, reflecting an effective balance between aquaculture and hydroponic components. Overall, these findings highlight the technical feasibility and biological efficiency of small-scale aquaponics as a sustainable food production approach for urban and peri-urban environments in arid regions.

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