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Monitoring of the Physico-Chemical Parameters of Water in a Small Scale Aquaponic System

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ABSTRACT

Climate change, water scarcity, soil degradation and food security threaten the Moroccan agricultural production. Aquaponics is an approach that can offer an alternative sustainable solution for covering local food demand, offering inhabitants arid regions access to both animal and vegetable proteins. However, this technique was never performed in Morocco. This study aimed to contribute to the development of economically feasible aquaponics systems in Morocco. For this, 46 individuals of the Nile Tilapia (*Oreochromis niloticus*) were raised and likewise three varieties of leafy vegetables (*Lactuca sativa* and Basil *Ocimum basilicum*) were grown in order to assess the impact of nutrients contained in aquaculture effluent on plant growth. Temperature, pH, electro-conductivity (E.C), and total dissolved solids (TDS) were daily measured using a multiparameter instrument "Hanna HI9814", while dissolved oxygen (DO), ammonia (NH_3), nitrite (NO_2^-), nitrate (NO_3^-), phosphorus (PO_4^{3-}), and potassium (K^+) were measured twice a week utilizing specific spectrophotometer HI801 of Hanna instruments. The Pearson coefficient, two-way ANOVA, principal component analysis (PCA), and cluster analysis were applied to test the correlation between different water quality parameters. Good adaptation of fish and plants was observed, with zero mortality of fish and commercial plant yields after only 30 days of introduction to the system. Results have shown that fish excrements are enough to grow lettuce and basil in aquaponics. Strong and significant correlation under 95% of confidence was obtained between DO, phosphorus and pH, while temperature recorded a significant correlation with both DO and nitrates. With regard to this experience, it was concluded that the installation of aquaponic systems is highly feasible and that the water circulating in aquaponics has an acceptable nutrient potential in order to ensure good plant growth.

INTRODUCTION

Aquaponics has been declared as a sustainable local food production technology contributing to global food security. It can be an alternative solution for covering local food demand, offering inhabitants of poor and/ or arid regions access to both animal and vegetable proteins, especially where water supply is relatively limited **(Rharrhour *et al.,* 2022)**. Aquaponics technology is based on the coupling of RAS (Recirculation aquaculture systems) with hydroponics (soil-less crop culture), in which nutrient rich aquaculture effluent is used to grow plants in a hydroponic system **(Diver, 2006; Klinger & Rosamond, 2012; Yep &**

Zheng, 2019; Rharrhour *et al*., 2022). Hence, this technique requires multidisciplinary knowledge in horticulture, statistics, aquaculture and aquaculture engineering.

Aquaponics is a quasi-closed food production system that can produce both proteins: fish and plants without the need of chemicals and pharmaceuticals; these systems can reduce 90% of agricultural water consumption **(Rharrhour *et al.*, 2022)**. However, this technique's drawbacks are presented, especially coupled aquaponics systems, in low economic profitability and inefficient availability of both macronutrients, especially potassium (K^+), calcium (Ca^{++}) and magnesium (Mg^{++}) in addition to micronutrients such as iron (Fe^{++}) **(Yep & Zheng, 2019)**.

Water quality presents a big challenge for aquaponics research, as it serves as a mechanical medium for both plants and fish. Given that an aquaponics system is a micro-ecosystem, water quality can affect, either positively or negatively, fish and plants' well-being. Conversely, the fauna and the flora of this system can, in turn, affect water physico-chemical parameters. Aquaponics water physico-chemical parameters must be balanced to ensure the well-being of fish, plants and bacteria **(Rharrhour *et al.* 2022)**. These three groups of organisms have, in general, the same tolerance ranges for most parameters **(Sommervile *et al*., 2014)**. **Rharrhour *et al.* (2022)** claimed that fish feed, pH, total nitrogen and temperature are aquaponics key elements, and pH is a significant water parameter that can affect directly aquaponics systems biocenosis, and the challenge is that these three groups have different optimal pH ranges **(Yep & Zheng, 2019)**. An acid pH range (5.5–5.8) is recommended for an optimal plant growth **(Bugbee, 2004)**, while neutral ranges are preferable by nitrifying bacteria **(Goddek *et al.*, 2015)**. On the other hand, nitrogen is another significant aquaponics parameter; it is primordial for plants growth, reproduction and storage **(Novoa & Loomis, 2005; McCauley *et al.*, 2009)**. Fish excrements are the principal nitrogen source in aquaponics systems; it offers 10 to 40% of nitrogen as ammonia **(Wongkiew *et al.*, 2017)**. Nitric nitrogen [NO_3^-] is the most essential form of nitrogen for plants, being a nutrient of prime importance for the growth of plants since nitrogen (N) is assimilated by these organisms, mainly by its nitrate ion form. Ammonium [NH_4^+] can be assimilated in small quantities too; however, it is oxidized into nitrate by nitrifying bacteria **(Canfield *et al.*, 2010)**. Generally, nitrate concentrations are harmless for aquaponic living components under 300mg/ l **(Graber & Junge, 2009, Hu *et al.*, 2015)**. Nitrite is the most toxic nitrogen form, its concentrations should be kept under 1mg/ l. Moreover, it can negatively affect both plants and fish species, as well as causing the lethal brown-blood disease for fish **(Timmons & Ebeling, 2010)**.

Aquaponics can be an alternative solution not only for its economic benefits but also for biodiversity threat and effects of climate change. Morocco's inland aquaculture is an epitome example for aquaponics potential to combat climate change and biodiversity threat; controlling reproduction and breeding of the Moroccan native species can be used to repopulate natural habitats alongside exploiting these species rationally in aquaponics to grow vegetables **(Rharrhour *et al*., 2022)**. This can be an effective solution for climate change effects, habitats degradation and overfishing of the Moroccan native species. Trout for example is represented in Morocco by numerous varieties, such as *Salmo akairos* and *Salmo multipunctata* as native species, and rainbow *Oncorhynchus mykiss* which was introduced in

1924. These species are rarely reared despite of its economic values, and their conservation must be the main goal of the Moroccan aquaculture to avoid its extinction as the case of *Salmo pallaryi* which populated Sidi Ali Lake **(Doadrio *et al.*, 2015)**. The blue barbell *Pterocapoeta maroccana* (Günther, 1902) is an another native species only found in Morocco, and it is the only representative of its genus **(Yahyaoui *et al.*, 2020)**. **Rharrhour *et al*. (2022)** correctly argued that this species needs to be protected and reared in order to repopulate its habitat. This shows that aquaponics has a great potential in the developing countries viz. Morocco although this approach can be a supply for biodiversity preservation. By rearing endemic fish species, aquaponics systems utilize their excrements to grow plants and protect these threatened species from extinction.

This study aimed to contribute to aquaponics development in Morocco through the technical study of aquaponics systems. A small scale aquaponics system was designed to measure the water physico-chemical parameters using a spectrophotometer in order to model the system and to grow both fish and plants, without the need for chemicals, antibiotics or fertilizers. Data of water temperature, electro-conductivity (EC), total dissolved solids (TDS), and pH were collected daily, while dissolved oxygen (DO), alkalinity (KH), ammonia (NH_3), nitrite (NO_2^-), nitrate (NO_3^-), phosphorus (PO_4^{3-}), potassium (K^+) and calcium (Ca^{2+}) concentrations were dosed twice a week.

The major contribution of this study were the following: development of an aquaponics system adapted to the Moroccan climate using simple materials available in the Moroccan market, application of spectrophotometry to measure chemical parameters of water and contributing to develop a mathematical model that can predict physicochemical parameters variation.

MATERIALS AND METHODS

1. Description and experimental setup

The present aquaponics system was implemented between September 2021 and December 2021 in BIOECOGEN laboratory of the Faculty of Sciences of Rabat, MED V University. The system design stage lasted for almost a month before the actual start-up and launch of the water circuit. The system was started on 09/19/2021 with a cycling phase that lasted 10 days to highlight the operation of the system and to evaluate the control measurements of the physicochemical parameters of the water. On 09/28/2021, forty-six (46) juveniles of the Nile tilapia *Oreochromis niloticus* were placed in a square glass aquarium (48 × 51 × 53cm, L, W, H) containing 71 liters of water. Juveniles of the tilapia with an initial average weight of 4.48g and an average length of 6.5cm were obtained from the National Center for Hydrobiology and Pisciculture (CNHP). The fish were fed a floating pellet feed made up of 37% crude protein once-daily; the feed was hand-delivered at a ratio of 2% of the total fish weight.

We adopted the floud-and-drain system (Fig. 1) in which aquaculture effluent was pumped, using a submersible pump with an electrical power of 25w and a maximum flow rate of 1750l/ h from the aquarium to the hydroponic unit consisting of a single plastic basin of

35cm long, 25cm wide, and 22.5cm high, with an area of 0.0875m² and a volume of 0.02m³, filled with volcanic gravel (Pozzolan). We used a hand-made bell siphon to ensure both good oxygenation of the hydroponic unit and the return of water to the aquarium. The aquarium was equipped with a heater having an electrical power of 300w, a mechanical internal filter and an air pump with an electrical power of 3w and a maximum flow of $3.5l.min^{-1}$.

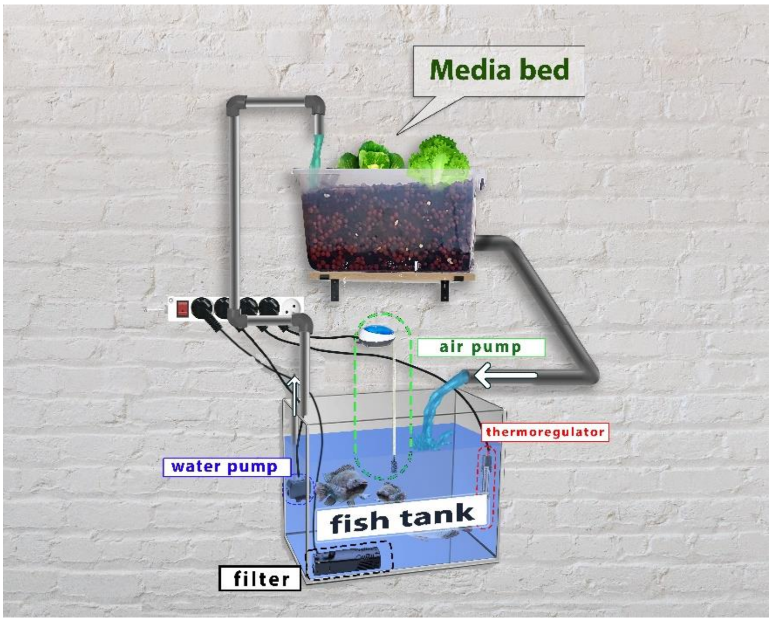

Fig. 1. Sketch of flood-and-drain system used in experiment

In this experiment we chose to grow three species of leafy vegetables: sweet basil *(Ocimum basilicum)*, and two lettuce *(Lactuca sativa)* varieties (madrilene and sucrine). This species choice was based on the Moroccan market demand and economic values of chosen species.

The seeds of these plants were bought from local market, cultivated in potting soil ten days before transplantation to the hydroponic unit using plastic net cup pots.

2. Data collection and analysis

Water temperature, electro-conductivity (EC), total dissolved solids (TDS), and pH were measured daily from 09/30/2021 until the end of experience, using a multiparameter instrument “Hanna HI9814”. Eight other water quality parameters were dosed twice a week using specific spectrophotometer HI801 of Hanna instruments, such as dissolved oxygen (DO), alkalinity (KH), ammonia (NH_3), nitrite (NO_2^-), nitrate (NO_3^-), phosphorus (PO_4^{3-}), potassium (K^+) and calcium (Ca^{2+}). Salinity was calculated from conductivity and temperature using the formula of **Aminot and Kérouel (2004)**.

All data were analyzed using R Studio version 3.6.2. The Pearson coefficient and analysis of variance (ANOVA) were used to see if there were any significant differences between different variables. Moreover, cluster analysis (CA) was used to classify variables according to homogeneity and correlation. Additionally, principal component analysis (PCA) was employed as a qualitative analysis strategy, commonly used in environmental studies.

RESULTS AND DISCUSSION

1. Water quality parameters

The fluctuation of the values of the physico-chemical parameters recorded were within the standards recommended in aquaponics. The use of the thermoregulator to control the system temperature had a positive impact on maintaining a stable temperature in our system during the experiment with an average value of 27.2°C. The drop in temperature observed during the first days (Fig. 2) is explained by the modification of the setting of the thermoregulator which was set at the beginning of the experiment at 30°C to accelerate the metabolism of the bacteria, and then reduced to more or less 27°C after the addition of fish. As for pH, its values varied between 6 and 7.63 with an average value of 6.83 (Fig. 2). The pH concentrations have gradually decreased over time, and this comes down to various factors, such as high CO_2 levels, low photosynthetic activity, especially in the nocturnal phase, and fish metabolism, such as respiration (CO2 rejection) and urination.

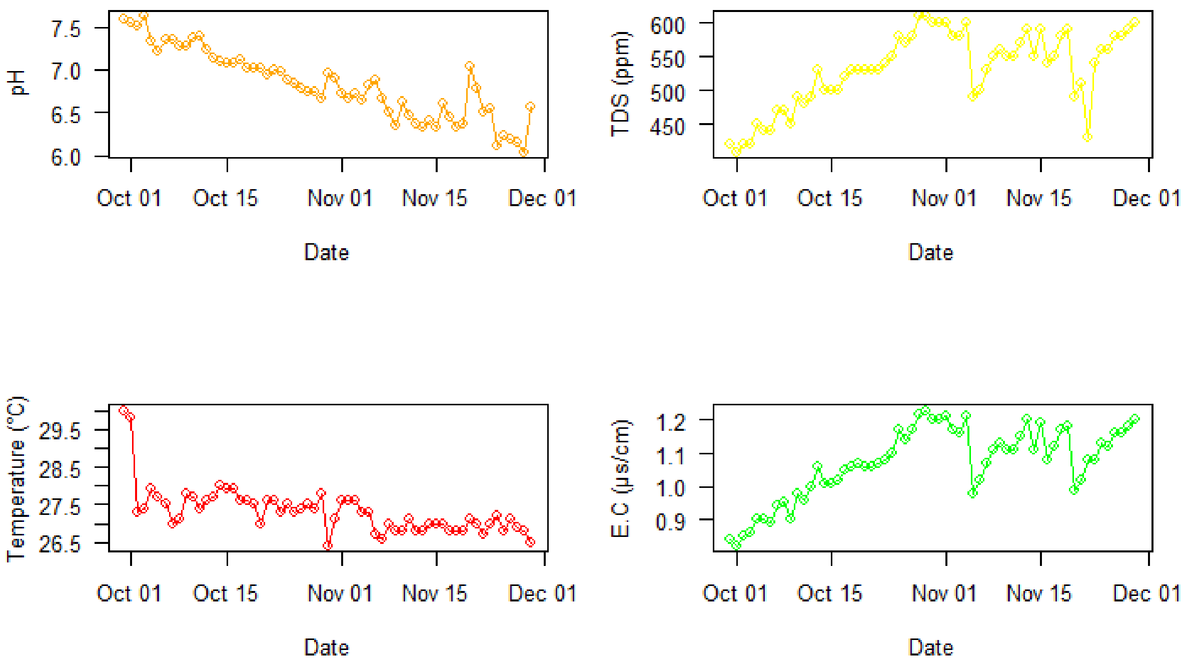

Fig. 1. Variation of pH, temperature, TDS and E.C during the experiment

For the total dissolved solids in the water, we noted that the values increase as the experiment progresses, going from about 400ppm at the start of monitoring to 600ppm by the fifth week (Fig. 2), with brutal falls that correspond perfectly to the weeks of water renewal. The average total dissolved solids (TDS) value for our system is 527ppm. The TDS variation presented in Fig. (2) can also be explained by the increase in the quantity of fecal matter generated by the fish; we also noticed that this parameter is correlated with the EC, while it varies inversely parallel to pH. The variation of electro-conductivity was perfectly parallel to the TDS, we noticed that the curve of the E.C. presents a strong correlation with the curve of the TDS, and we then deduced that the EC varies proportionally with the TDS. Same thing for salinity, which was calculated according to temperature and EC, which explains why it varies proportionally with the latter. At the start of the experiment, the EC values did not exceed 1ms; however, as the fish grew and the fecal load on the system increased, the values of this parameter continued to rise over time. By the end of October (Fig. 2), the EC reached a maximum value of 1.23ms, while undergoing significant decreases during water renewal. The decrease can also be explained by the absorption of nutrients by the plants; however, in our system, the conductivity increases due to the uptake of nutrients by the plants being lower than the concentration resulting from waste from the aquaculture unit.

2. Spectrophotometer analysis

The dissolved oxygen (DO) levels varied between 4 and 7.7mg/ l, with a control value of 8.8mg/ l and an average level of 5.84mg/ l (Fig. 3). The level of DO during the experiment remained above the recommended standards for aquaponics. This fluctuation is mainly due to the flow of water falling back into the fish tank since the flow of the air pump is generally stable. Maintaining this parameter at the right concentration ensured the well-being of fish and avoided deficiencies related to DO in plants.

The concentrations of nitrites are generally correlated with those of ammonia; they fluctuated between 19 and 889µg/ l during the experiment (Fig. 3), recording a sharp drop on the twenty-sixth day of monitoring. The variations shown in Fig. (3) are related to fluctuating temperature, pH, and ammonium and ammonia concentrations. Nitrites, being a very harmful form of nitrogen for both plants and fish, must generally be kept below the recommended standard in aquaponics. A high concentration of this parameter can lead to necrosis in plants, as well as stress for the fish, which explains the periodic renewal of the water, aiming to keep this concentration below the standard.

The concentration of NO_3^- started to increase significantly with the introduction of fish to the system until it stabilized around the fifth week at about 130mg/ l (Fig. 3). The appearance of nitrates is due to the phenomenon of nitrification. Given the abundance of waste from the aquaculture unit and following other processes (mineralization and oxidation), ammonium/ammonia are quickly transformed into nitrates, which explains the significant increase in nitrate concentration in our system. On the other hand, its stability at a value that does not exceed 140mg/ l can be explained by both the assimilation of plants and the maximum nitrification capacity of the bacterial community.

Since the addition of the fish to the systems, the ammonia has not ceased to increase progressively according to the feces generated, but a peak was recorded at the seventh week of monitoring with a value of 3.65mg/ l (Fig. 3). A week later, a sudden drop in concentration was recorded, and this is explained by the renewal of water. We also observed a return to the peak around the sixty-second day of monitoring. The concentrations of ammonium and ammonia are associated; the variation of ammonium leads to the variation of ammonia. The chemical equation that determines the relationship between ammonia and ammonium is: $NH_3 + H_2O \leftrightarrow NH_4^+ + OH^-$. Ammonia concentrations are influenced by temperature and pH since NH_3 is non-ionized. In general, at room temperature with a pH lower than 6, the portion of NH_3 is very low, and almost all the ammoniacal nitrogen is present in the form of NH_4^+.

Normally, phosphate is the limiting plant growth factor in traditional agriculture and hydroponics, but for our system, orthophosphates have been widely available. Phosphate levels varied between 1 and 3.1mg/ l during monitoring (Fig. 3), with a slight decrease in the second half of the experiment (introduction of plants). Orthophosphates also come from fecal matter, which explains their abundance in our system. The decrease in the concentration of phosphate is explained by the absorption of this nutrient by plants. The hydroponic unit did not show phosphate deficiencies, which proves the stability of this parameter above the norm.

The lowest value recorded for potassium is that of the control (cycling phase without fish) at 6mg/ l (Fig. 3), while the highest value is recorded on the thirty-third day of monitoring at 20mg/ l. In general, potassium levels fluctuated between 6.5 and 15.5mg/ l. Potassium is very important for the well-being of plants; it helps to strengthen cell walls and to increase both leaf area and the chlorophyll content of leaves, playing a role in photosynthesis. Our system did not present a deficiency in this nutrient.

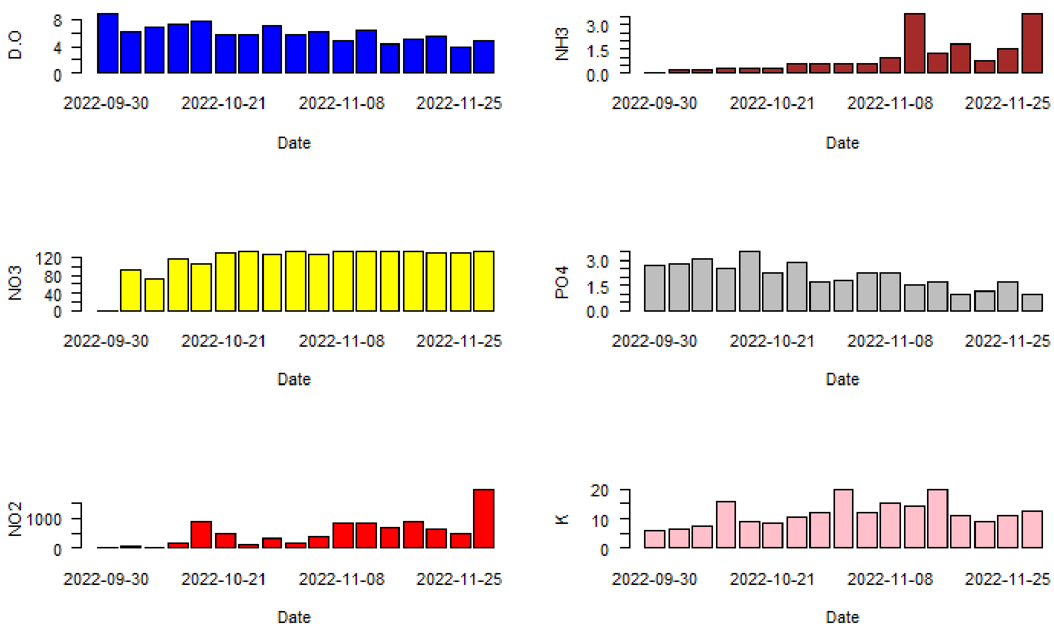

Fig. 2. Spectrophotometric results. Codes: Dissolved oxygen = D.O, Nitrates = NO3, Nitrites = NO2, Ammonia = NH3, Phosphates = PO4, and Potassium = K

3. Correlation measures

- The Pearson coefficient

In order to measure the relationship between two variables, we chose to calculate the Pearson coefficient between all parameters (Table 1).

Table 1. Pearson coefficient of correlation of pH and temperature with other parameters

Variable	pH	Temperature
Electroconductivity	- 0.8017813	- 0.5354016
Salinity	- 0.8187477	- 0.6029348
Total Dissolved Solids « TDS »	- 0.7502766	- 0.479805
Dissolved Oxygen « DO »	0.7814588	0.78601
Alkalinity « KH »	0.9571932	0.7602357
Nitrates « NO_3^- »	- 0.8067207	- 0.8300247
Nitrites « NO_2^- »	- 0.554988	- 0.5126913
Ammonia « NH_3 »	- 0.5998536	- 0.4914349
Phosphate « PO_4^{3-} »	0.7501727	0.481593
Potassium « K^+ »	- 0.5600303	- 0.2625376

"r" varies between -1 and 1; when "r" is equal to 0, the two variables are therefore independent, whereas if "r" is equal to -1 or 1, the two variables are totally dependent.

We have measured the correlation between pH and temperature (r = 0.6063314). Generally, when “r” is positive, the correlation is said to be positive, which means that when one variable changes, the other changes in the same direction. This explains why these two parameters correlate in the same way with the other parameters. pH and temperature have shown positive correlation with dissolved oxygen “DO” and phosphate “PO_4^{3-}”, while it was negative for the rest.

- **Two-way ANOVA**

We have used multiple linear regression (MLR) to test whether there is a link between the quality parameters and the four variables which are pH, temperature, conductivity and TDS. Table (2) provides “The *P*-values (significant if $P< 0.05$)” and “estimate values” from results.

The results show that under 5% of risk, dissolved oxygen has a statistically significant association with pH and temperature, with significant $P< 0.05$, which means that when pH increases by 1 degree, DO value increases by 2mg/ l, and when temperature increases by 1°C, DO value increases by 0.8mg/ l. These two variations are additive, provided that there is no synergy of potentiation between the two explanatory variables on DO values; the test of verification shows a $P> 0.05$; therefore, there is no interaction between the two variables on OD, and then the OD values increase by 2.8mg/ l as a function of pH and temperature. Phosphate values show statistically significant correlation to pH; its values increase by 1.4mg/ l as a function of pH ($P< 0.05$).

Table 2. Summary of main statistics and two-way ANOVA results (95% confidence)

	D.O	NO_3^-	NO_2^-	NH_3	PO_4^{3-}	K^+
Mean	6.018	115	530.3	1.007	2.1	11.78
SD	1.242	34.2	482.7	1.106	0.7	4.13
Max	8.800	132.8	1970	3.650	3.5	20.00
Min	4.000	0	19	0	1.0	6.00
2 way ANOVA (p values)						
pH	0.03149*	0.615	0.305	0.101	0.0482*	0.371
Temperature	0.02396*	0.011*	0.424	0.504	0.6709	0.330
E.C	0.22550	0.128	0.651	0.282	0.4834	0.642
TDS	0.31926	0.708	0.645	0.317	0.3536	0.658

Statistically significant correlations ($P < 0.05$) are marked with asterisks (*).

Nitrates only shows significant negative correlation to temperature when temperature increases by 1°C, NO3- values decrease by 8.8mg/ l. Other water quality parameters show $P >$ 0.05, which means that those four explanatory variables have no effect on their values.

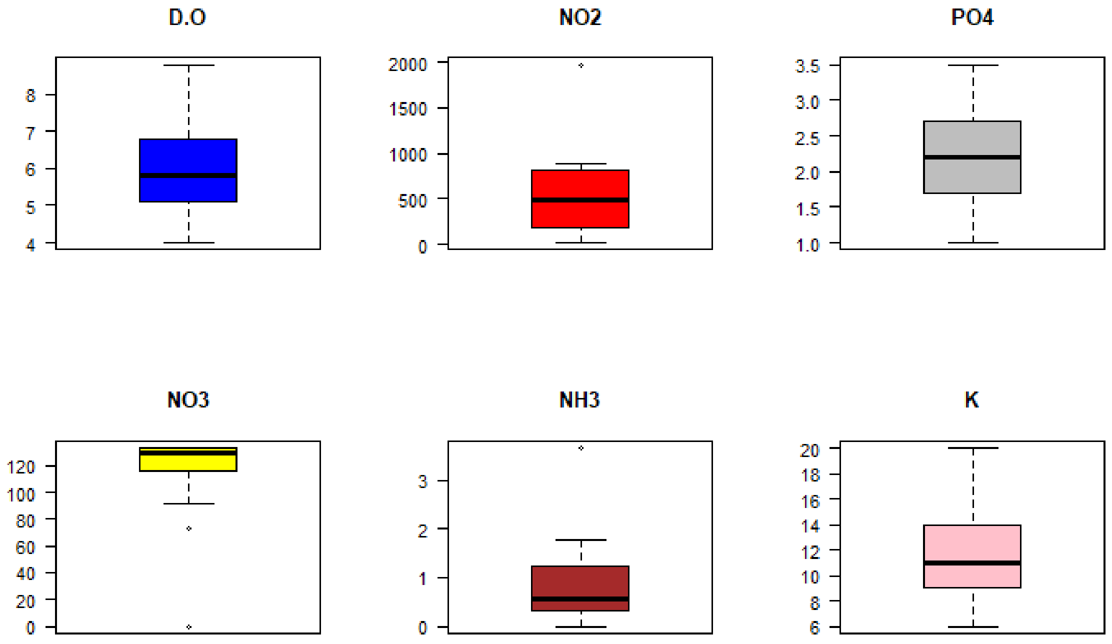

Fig. 4. Graphical effect of pH, temperature, E.C and TDS on the average results (two-way ANOVA) of water quality parameters (average values and confidence intervals calculated using the least significant difference, 95% confidence). Codes: Dissolved oxygen = D.O, Nitrates = NO3, Nitrites = NO2, Ammonia = NH3, Phosphates = PO4, and Potassium = K.

- **Principal component analysis (PCA)**

To understand the structure of our system, we used the principal component analysis (PCA) which create instruments to analyze things that cannot be measured directly, and condense information from a large number of variables into a small set with minimal loss. The variation of values of the concentrations of the physico-chemical parameters is presented in Fig. (5).

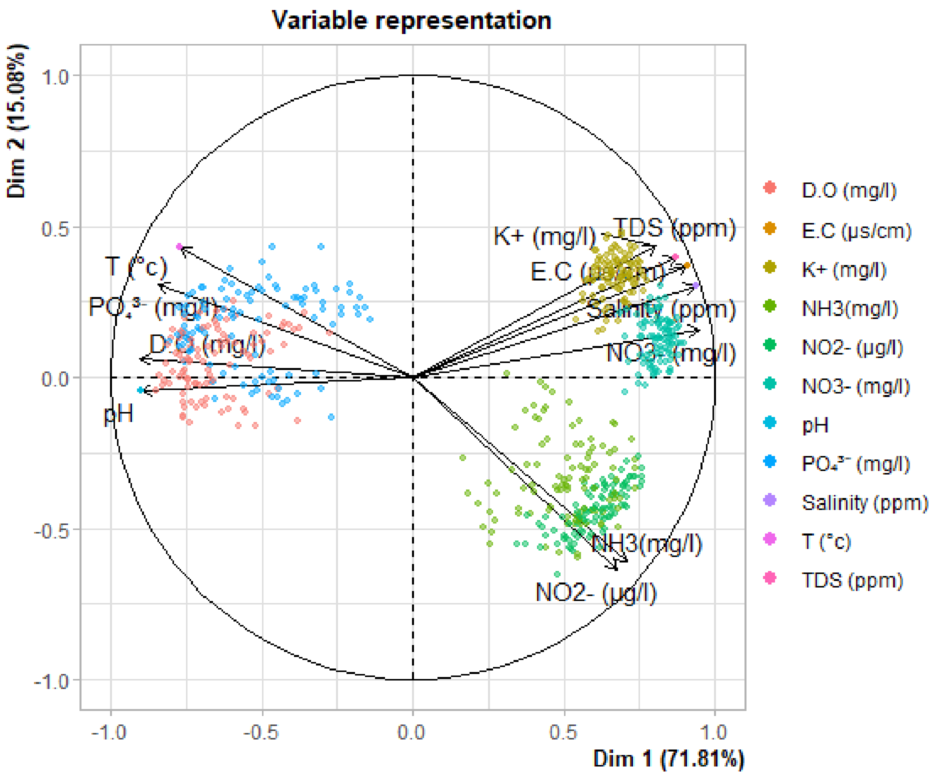

Fig. 3. Component plot

By applying the principal component analysis (PCA) on the water quality parameters, three groups of physico-chemical parameters are observed; the first follows the dim1 axis positively and gathers TDS, E.C., potassium, salinity and nitrates; the second follows the dim1 axis negatively and gathers temperature, dissolved oxygen, phosphates and pH; while the third is a set composed of ammonia and nitrites which correlates positively.

- **Cluster analysis**

The cluster analysis confirms (PCA) results. The resultant dendrograms (Fig. 6) shows that indeed there were three main clusters, identified as follows: The first cluster gathers temperature, dissolved oxygen, phosphates and pH at distance 9 (correlated in PCA); the second cluster contains ammonia and nitrites at distance 5 (correlated in PCA), and the third cluster gathers TDS, E.C., potassium, salinity and nitrates at a distance 10.

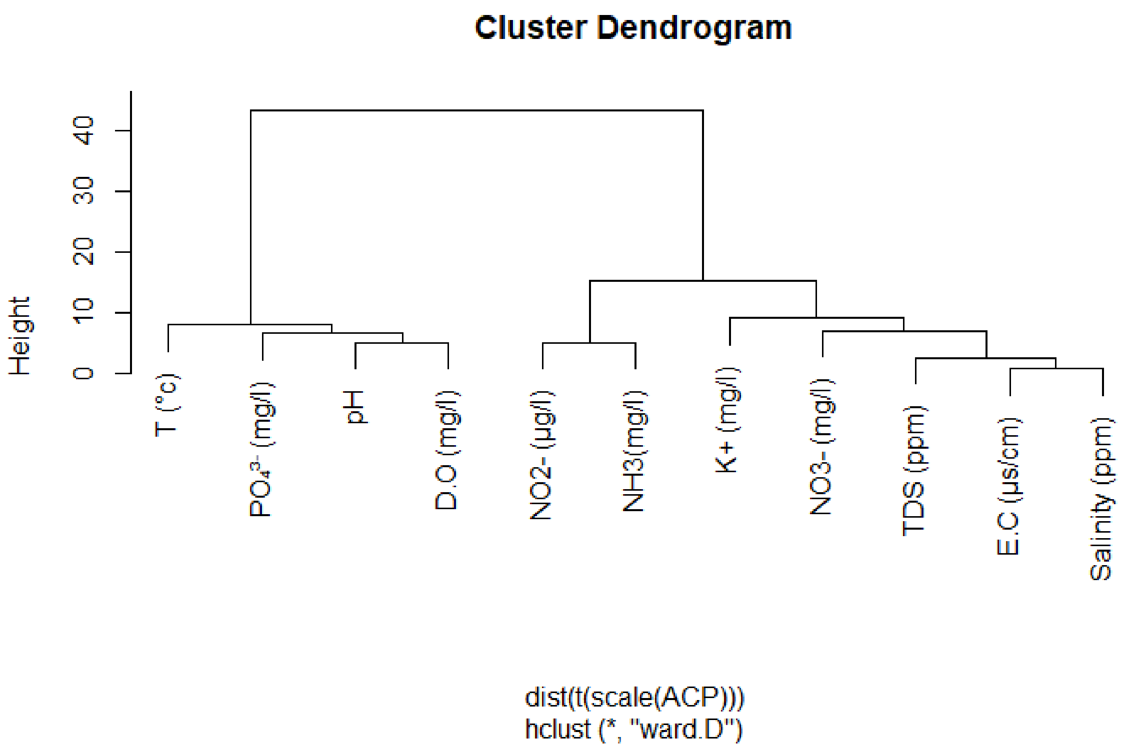

Fig. 4. Dendogram obtained by hierarchical clustering analysis for aquaponics water parameters

CONCLUSION

Given the fact that the only source of nutrients for plants was aquaculture effluent, results are promising showing that aquaponics indeed might be an alternative solution for the Moroccan agricultural problems. The observed correlations might be useful to both predict water quality parameters variations in order to develop a model adapted to Moroccan weather, and to have optimal control of the system, especially with the pH challenge represented in different optimal ranges for three different organisms (fish, bacteria and plants). However, further research and experiments are needed in order to compare different aquaponics set-ups, and to test different plant and fish species, especially the native ones.

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