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CONSERVATION OF AQUATIC ECOSYSTEMS BY CONSTRAINING NITROGEN POLLUTION THROUGH AQUACULTURE EFFLUENTS

Vasile Daniel GHERMAN[1], Vily Marius CIMPOIASU[2], Ioana Corina MOGA[3], Radu POPA[4, *]

[1] University Politehnica of Timisoara, Faculty of Civil Engineering, Department of Hydrotechnics, Spiru Haret Street, Timisoara, Romania.
[2] University of Craiova, Faculty of Horticulture, Biology and Environmental Engineering Department, A.I. Cuza Street, no 13, Craiova, Romania.
[3] DFR Systems S.R.L., Drumul Taberei Street, no 48, Bl. G2/3, Ap. 2, Sector 6, Bucarest, Romania.
[4] River Road Research, 2440 Sheridan Dr., Tonawanda, NY, 14150, United States of America.

Abstract

Recirculated aquaculture systems (RAS) have increased in preponderance in producing fish and shrimp protein. Yet, the economic sustainability of constraining RAS from negatively impacting aquatic ecosystems remains challenging. The future of RAS agriculture will eventually be settled by the relationship between water treatment costs and the impacts on downstream ecosystems. We present a user-friendly simulator of the costs of the treatment of water from RAS farms. This open-source freeware accounts for consumables and energy needed to protect the fish stock from ammonia and nitrite distress, as well as the cost of effluent treatments for specific nitrogen emission targets. This simulation platform uses information inflows about a RAS farm's layout, filters' performance, toxicity limits, and operational costs. It monitors the budget of water, ammonium, ammonia, nitrite, and nitrate, as well as the cumulative costs of management decisions for controlling nitrogen inorganics. In combination with local environmental regulations, such an assessment is essential for making business projections that correspond with acceptable impacts on downstream ecosystems. This simulator helps determine whether a specific RAS farm is both financially sound and environmentally sustainable. Such analyses are key to constraining pollution in the surrounding ecosystems and contributing to the conservation of biodiversity.

***Keywords**: Biodiversity conservation; Sustainable Aquaculture; Simulator; Biofilters; Nitrogen; Macrophytes; Ecosystem stability.*

Introduction

Large Recirculated Aquaculture System (RAS) farms can afford complex and expensive hardware and software systems to manage changes in water chemistry. But such systems are not affordable for smaller farms. In such farms, automation is frequently not cost-efficient and has to be replaced by frequent hands-on water testing and direct intervention. To such ends, software solutions are needed that are low-cost, easy to learn, and tailored to various system configurations. In aquaponic systems, management decisions are further complicated by additional operational costs, which can make the difference between a profitable and an

* Corresponding author: radu.o.popa@gmail.com

unsustainable aquafarm. We developed software predicting changes in water chemistry and the cumulative costs of these changes on small-scale RAS and aquaponic farms.

Of the numerous aquaculture parameters, this software focuses on the dynamics of water, nitrogen evolution, and energy and water costs. This manuscript implements this analytic capability on a virtual example of a freshwater salmonid RAS farm. Among others, the efficiency of RAS is influenced by the level of water recirculation. This is mostly due to an increase in the costs of constraining toxic physiological byproducts when more of the water is reused. Water plants biofilters lessen some of the cleanup burdens while adding others. Thus, there is a need for cost-benefit evaluations. E.g., if clean water were cheap and abundantly available (unrealistic in most cases), avoiding macrophyte biofilters and reusing little water may be the cheapest solution. Yet abundant sources of clean water for aquaculture are in short supply. This curb the capacity of salmonid RAS to keep fish stocks healthy and water treatment costs low as the water renewal percentage increases. The need exists for novel and more affordable means to treat the reused water. In water-constrained RAS operations, regeneration of filters (such as nitrification biofilters, active carbon, or zeolite scrubbers) is hard to afford. Macrophite biofilters have the potential to assist warm water RAS, but system' design is important, and it is still unclear how helpful macrophytes are in cold water used for salmonids.

Managing the soluble N waste in RAS is particularly challenging. Depending on how this is done, the economic sustainability of a RAS farm and its impact on the environment will vary. As the fish stock grows, the ammonium (NH_4^+) produced increases water toxicity and may force changes in the water renewal rate or treatment capacity. In RAS, most of the ammonium comes from fish excretion and, to a lesser extent, from the ammonification of organic wastes [1]. Increases in pH above 6.5 and incomplete nitrification turn NH_4^+ into more toxic chemicals such as unionised ammonia (NH_3^+) and nitrite (NO_2^-). The common solution to N-related toxicity and effluent pollution is integrating various means of management. Ultimately, the choice of methods influences the overall economic efficiency, as various N-processing methods have various costs/benefit ratios.

As a fish stock grows, the mismatch between ammonium production and its controlling costs will increase. Increasing the number, or dimension, of non-biological means to manage N (e.g., settlers, filters, pH buffering, scrubbers, N precipitation, or water replacement) is not always sufficient or economical. Situations will exist when the N-controlling means are overkill for the longest part of a growth cycle (i.e., when fish are small) but insufficient for a short period just before harvest. In large-scale RAS facilities, with fish grown in many parallel series and multiple harvests per year, a large-size water treatment facility can service more RAS circuits as they approach the high-NH_4^+ production stage. Such means are most often unaffordable for small farms.

Oxidising all the RAS' NH_4^+ to less toxic NO_3^- is insufficient. Without net removal of N from the system and within a single production cycle, the fish will excrete sufficient ammonium-N (NH_4^+-N) (e.g., 87g NH_4^+-N per 1,000g of fish produced) to make the derived [NO_3-N] toxic as well; the toxicity limit for nitrate is ~70mg/L [2]. On the other hand, gradually increasing the water renewal rate as toxic nitrogen accumulates is not always possible. Flushing the system with more water exposes the fish stock to external factors and leads to more nutrients being discarded downstream. In RAS, it is often not economically sustainable to lower the concentration of ammonium, ammonia, or nitrite to fish-safe levels by means common in water treatment plants (ion-exchange, denitrification, reverse osmosis, or mineralization to struvite).

As aquaculture grows, the RAS' economics will face a conundrum. On the one hand, the more water reused, the higher the fish production costs. On the other hand, more water means more pollution. Legal restrictions will likely constrain aquaculture outflows, along with more pollution taxation [3]. Though some low-tech methods (such as woodchip denitrification, constructed wetlands, and sand infiltration) were shown to achieve near zero pollution [4], such a high benchmark is seldomly reached by fish farms. The NH_4^+-derived toxicity of soluble N chemicals increases exponentially during a RAS cycle while global RAS activity increases. A crisis point is predicted to occur where many conventional methods to control the soluble N will simply become unaffordable for most farms.

Abiotic methods to limit soluble mineral N (minN) in water include larger water renewal rates, NH_4^+ scrubbers [5], magnesium-CSC composite filters [6], and reverse osmosis [7]. Biological means are phototrophic, autotrophic, or heterotrophic microbial reactors with bacteria or algae [8], fertigation [9], and macrophyte biofilters [10]. Microbial reactors can be based on nitrification or denitrification [11], phototrophic microbial mats and biofilms, planktonic microalgae [12], biofloc [13], and microalgae [14]. Macrophyte biofilters can be in the form of crop-centred hydroponics, aeroponics, or fogoponics [15], floating macrophytes [16], constructed wetlands and soil cultures [17], artificial wetland systems with helophites [18], submerged or floating macrophytes [10], and rice paddy aquaculture [19]. The fact that many methods exist to remove N doesn't mean that the RAS' N-management problem is easy. Many of these methods are costly (in terms of $/g of N removed to fish-safe levels).

Aquaponics was often presented as a revenue-generating solution because vegetable crops, microgreens, or forage biomass could be produced as well. Yet to date, most hydroponic farms are either aquaculture or hydroponic operations. Aquaponics is a complex process with costly infrastructure, and its evolution is often unpredictable, requiring advanced skills or experience to understand, control, and optimise for profit. Very often, hydroponics-related expenses will not cover costs, especially when the control of water chemistry is not focused on plants' growth but on fish safety. There is no good rule of thumb for a fish-safe plant-to-fish ratio. This ratio is, in some cases, 2:1 (by weight). But phytomass density (in kg/m^2) is less than the fish mass density (in kg/m^2) in fish tanks. Hence, in aquaponics, more space is allocated to the plants than to the fish. At a ratio of 2kg of fish per m^2 of hydroponic system, adding a climatized indoor hydroponic facility to an indoor RAS operation will increase about tenfold the surface area requiring indoor climatization [18]. Hydroponics itself can be sustainable, but only when key factors are optimised for plant growth (such as nutrients, temperature, illumination regime, pH, Eh, and humidity). Hydroponic optimisation is also not trivial, and most fish farmers are not prepared to produce or sell vegetables. Most fish farmers would likely prefer hydroponics to be a secondary activity in support of the aquaculture operation. Reality is vice versa; hydroponics has to function at near 100% capacity in order to be profitable, and footprint-wise, it will take over most of a RAS farm's surface area. Hydroponic performance is not easily achievable if water treatment to protect the fish stock becomes a "make or break" objective.

An alternative to crop-centred indoor aquaponics is coupling RAS with low-cost, and thus less revenue-dependent, macrophyte biofilters. Examples include constructed wetlands, outdoor hydroponics, and floating islands [20]. The solution tested in the model presented here is called a hydroponic outdoor system (HOS). It consists of macrophyte biofilters with seasonal plants grown outdoors but in hydroponic setups. This is unlike plants grown in soil, constructed wetlands, ponds, marshes, lakes, or paddies. Most constructed wetland biofilters treat effluents rather than the recirculated RAS water. Also, most constructed wetlands and controlled macrophyte biofilters studied so far were developed for warm-water aquaculture systems. They are better for farming tilapia, catfish, or cyprinids. Floating island phytofilters are the closest in similarity to HOSs.

The goal of the programme developed for this study is to compare various RAS architectures, e.g., RAS alone, aquaponic RAS with hydroponic indoor biofilters (RAS/HIS), and RAS with hydroponic outdoor phytofilters (RAS/HOS) (Fig. 1).

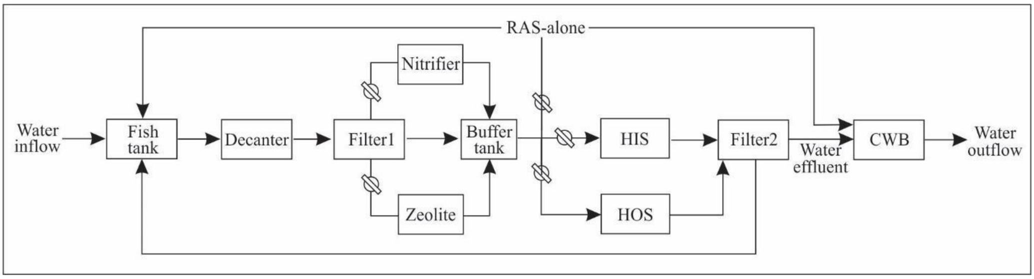

Fig. 1. Main stocks and flows of the dynamic simulation model: HIS = Hydroponic Indoor System; HOS = Hydroponic Outdoor System; CWB = Constructed Wetland Biofilter

Experimental part

Materials

An aquaponic system model was constructed in Stella Architect [21] (https://www.iseesystems.com/). This approach is common in behaviour-predictive studies of aquaculture systems [22]. Here, monitoring of changes in the water budget occurs as well as inorganic N chemicals and costs affected by various managerial decisions regarding the control of soluble inorganic N. In the model, the labels for the various N parameters are TAN for total ammoniacal nitrogen; NH_4^+-N for ammonium nitrogen; NH_3-N for un-ionised ammonia nitrogen; NO_2-N for nitrite nitrogen; and NO_3-N for nitrate nitrogen. Three system configurations can be selected: RAS-alone, RAS/HIS, and RAS/HOH, with or without connection with nitrification and zeolite filters (Fig. 1). The computer model is open-source and available at: https://uptro29158-my.sharepoint.com/:u:/g/personal/vasile_gherman_upt_ro/EcqsG41UkKJGgtfHpONQ6yABHR-k9esX9RmI6VH0bIleXA?e=cVOjN9. Readers and model users can change quantitative parameters, formulas, and growth assumptions; introduce or delete novel pathways; and create graphs and tables for results to be analyzed.

Methods (the model construct)

sers with "Stella Architect" installed on their computers can download the model file and run simulations directly. Those who do not can upload the model into "Stella Online" and run simulations online as follows:

a) Access: https://exchange.iseesystems.com/signup; isee Exchange™ (iseesystems.com).

b) "Login" or

c) "Create account".

d) Select "Add a model".

e) Go to the URL address (e.g., https://uptro29158-my.sharepoint.com/:u:/g/personal/vasile_gherman_upt_ro/EcqsG41UkKJGgtfHpONQ6yABHR-k9esX9RmI6VH0bIleXA?e=cVOjN9).

f) Download the file to the computer.

g) Upload the file into "Stella Online".

h) A window may appear after uploading the model: "Warning: This model exceeds the limitations of your account. This model will be presented to you in player mode". Close this window.

i) The model's input conditions and results can be found in the upper left corner.

j) To perform "Zoom-in" or "Zoom-out" functions, use the "Ctrl+" or "Ctrl-" keyboard combinations.

k) Select "Run model, press play" (i.e., the black triangle in the lower left corner).

l) On personal computers (and depending on specific system performance), a simulation representing 12 months of a RAS evolution at 1-minute intervals will take on average 14 minutes to complete.

m) NOTE: With "Stella Online, you can modify model parameters but cannot save them.

Model functioning features: these include key variables, the base for various formulas, and how data is interpreted.

Temperature

The current model analyses RAS and aquaponics at temperatures that are optimal for freshwater salmonids. Examples are 12–16 °C for rainbow trout (*Oncorhynchus mykiss*), 7–13 °C for Brook trout (*Salvelinus fontinalis*), 9–16 °C for brown trout (*Salmo trutta*), and 13–15 °C for Atlantic salmon (*Salmo salar salar*). For helophite macrophites, the submerged part of the plant will be exposed to the RAS water temperature, while the aerial parts will be exposed to air temperatures. The current model analyses water temperature, and changes are needed to account for indoor climatization. In salmonid RAS farms placed in warm climates, heat exchange between water and air must be limited as salmonids will not tolerate warm water (e.g., ≥ 20°C). The critical thermal maxima for rainbow trout mortality begin at 23°C [23]. This software focuses on costs associated with water and nitrogen management. It does not model the caloric budget of a RAS facility or heat exchanges between water and air.

Fish stock density

In trout aquaponics, a stock density of 12–15.2 kg for every 1,000 litres of water in the system is recommended (https://farmingaquaponics.com). This value will be modified in the model if other fish are used. As a rule of thumb, the largest fish-to-water ratio in this model is 14.8 kg of fish per mt. of water. Notably, in aquaponic systems, most of the water (66–80%) is in the plants' section, plus some in decanters, filters, buffer reservoirs, and conduits. In the fish tank, we used an upper density to limit diseases and aggressivity, e.g., $80 kg \cdot m^{-3}$ for rainbow trout (https://www.globalseafood.org/).

Fish growth assumptions

At 14–16 °C, rainbow trout grow to 1kg in approximately 14–16 months (https://www.fao.org/fishery/). Based on Eq. 1, reaching 1,000g in 15 months from 0.1g juveniles requires a specific growth rate of approximately $0.002 g \cdot d^{-1}$.

$$\mathbf{r = [ln(Wf/Wi)]/N} \ldots\ldots\ldots\ldots\ldots\ldots (1),$$

where: r = specific growth rate ($g \cdot d^{-1}$); Wf = final weight (g); Wi = initial weight (g); N = number of days.

During simulations, the fish stock growth rate is constantly recalculated in step - 1.

Aquaponics-based model users will find that macrophyte biofilters are more valuable at reducing N in the last months of the RAS cycle, when the excretion-derived [TAN] increases. From such simulations, users will be able to forecast the best timing to connect the RAS with the hydroponic filter. The step-dependent fish size is derived from:

$$\mathbf{Wf = Wi \bullet e\hat{}(N \bullet r)} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2)$$

In the warm season, the TAN production by the fish stock will show exponential growth. As the model sets specific thresholds for N toxicity, users can predict at what individual body size the fish will become too large for the hydroponic system? Once this critical Wi size is identified, the step where the transition from RAS-alone to RAS/Hydroponics combination has to occur is calculated as:

$$\mathbf{N = [ln(Wf/Wi)]/r} \ldots\ldots\ldots\ldots\ldots\ldots\ldots..(3)$$

Feeding the fish

Here, it is assumed that trout will be fed a standard diet with 42% crude protein (an adjustable variable in the model). Commonly, such a diet is associated with a food conversion ratio (FCR) ≤ 1.0 for fish that are < 100g in weight, while between 100 and 1,000g, the FCR is between 1 and 1.3 (https://www.globalseafood.org/). Since this model focuses on chemical stress during the last 6–8 months of fish growth and because the aim is to secure sufficient biofilter means to keep the fish healthy, the FCR is set at 1.3 (also a modifiable variable).

Mineral N chemicals

We assumed that 25% of the N from the feed is recovered in the fish produced and that most of the excreted N ends up as dissolved ammonium [24]. In salmonids, between 47 and 87 kg of soluble N are generally excreted for each 1,000kg of fish produced [2]. Here, we selected the largest value (i.e., 87 kg) in order to prepare sufficient biofilter means, thus avoiding toxic N levels. Ammonification of organic N (Org-N) in decanters and other subsystems is also likely to occur in poorly managed systems, creating additional TAN. But, because in trout aquaculture the DO levels are generally high, the Eh values are positive, the BOD levels are low, and the organic N levels are kept low, ammonification is considered negligible. In this model, the oxidation of ammonium to nitrite and eventually nitrate occurs in nitrifying bioreactors. At pH = 7.2 (recommended for efficient nitrification) and 15°C (recommended for trout), the [NH_3-N] is 0.432% of the [TAN] [25]. Users can modify this percentage if the targeted pH and temperature are different. In this model, the N-related toxicity in the fish tank is controlled in five ways: water renewal, water recirculation, nitrification, zeolites scrubbing, and macrophytes. A sub-system for regenerating the nitrifying biofilms is assumed to also exist and be part of nitrification costs. Such regeneration systems are not coupled with the main water circuit from Fig. 1 and thus are not included in the water circulation subsystem.

Physical filters

Two physical filter units are present, labelled Filter 1 and Filter 2. Their role is to remove organics, mostly particulate and some soluble (e.g., faeces, uneaten food, plant litter, and DOC). These are combinations of coarse filters, fine filters, and carbon filters. Users must be knowledgeable of their filter performance and frequency of cleanup to input credible data into the model. In trout RAS, physical filters will lower the BOD to fish-safe levels and increase the efficiency of nitrification. If the [BOD] level is not lowered sufficiently upstream from the nitrification filters, clutter of the nitrification media will occur with heterotrophic biofilms. This means more frequent regeneration and increased nitrification costs. The cost of Filter 1 is not included in this model because this filter will always be present in all RAS configurations and thus will not matter for comparisons between RAS alone and RAS/hydroponic combinations. Yet, model users can add Filter 1 participation to their costs. The second physical filter (Filter 2) is present in all RAS/HIS and RAS/HOS configurations. Its scope is to remove particulates and organic matter released by the hydroponic section. Some plants in hydroponic-like setups, such as floating islands of reeds, cattail, bamboo, and papyrus, produce massive amounts of litter that has to be removed. This second filter is key for salmonids, whose health requires low DOC levels. The cost of carbon filtration can be quite high in RAS and hydroponic systems. Some very large aquaponic farms may afford the infrastructure needed to regenerate the carbon, but this is not likely on small farms. Here, the cost of Filter 2 was approximated at $1,000, with a lifetime of 20 years and a yearly cost of $100 for filter washing and carbon regeneration.

Nitrification costs

General recommendations for efficient nitrification in freshwater RAS are an inflow TAN ≤ 2mg/L, DO in water > 4mg/L, pH = 7.2 and [BOD] as low as possible in tens to a few hundreds of ppm range [26]. Here, the production of TAN is:

TAN = Feed • Protein • Nw • 0.16………(4)

where: TAN = total ammoniacal nitrogen produced ($g \cdot d^{-1}$); Feed = amount of feed added ($g \cdot d^{-1}$); Protein = crude protein in feed (%); Nw = nitrogen wasted from food (%) and 0.16 = g TAN/g Protein.

The volumetric TAN conversion rate (VTR) in nitrification biofilters is in g of TA $m^{-3} \cdot d^{-1}$. Moving-bed bioreactors (MBBR) are more efficient (per m^2 of media) than trickling bioreactors and are expected to become more common in RAS farms [26]. An MBBR' VTR may be 1.45-1.75$m^{-3} \cdot d^{-1}$ depending on various specific conditions and modes of usage [26]. The amount of MBBR media needed by the modelled system is:

BMV = TAN/VTR/%CBM………………(5)

where: BMV = biofilter media volume (in m^3); TAN = production of ammoniacal nitrogen (in $g \cdot d^{-1}$); VTR = volumetric TAN conversion rate (e.g., 350g TAN $m^{-3} \cdot d^{-1}$, at $200 m^{-2} \cdot m^{-3}$ and T = 25°C); % CBM = ratio between the media volume (in CBM) and the reactor' volume (≈ 70%).

For example, a RAS with 800kg of trout at 15°C and producing 1,010g of TAN d^{-1} will require 2.9m^3 of MBBR media in a 4.14m^3 bioreactor. MBBRs have a flow rate limit for peak performance. Systems with large water flows have to use MBBRs that can be rearranged in parallel or in series to adapt to the optimal flow rate. Though this technical detail is important in a RAS facility, it does not require changes in the model. Since the temperature is set at 15°C instead of 25°C (as reported for most nitrifying reactors), the true VTR is smaller. It was reported that the impact of temperature on a fixed film's nitrification rate is less than predicted by the van't Hoff-Arrhenius equation [27]. Under conditions of non-limiting [DO] and [TAN], the rate of change in nitrification efficiency ranged between 1.086 and 1.109% per °C [28]. This is equivalent to an MBBR that must be approximately 10% larger at a temperature of 15 °C relative to 25°C.

In water with high DOC levels, the nitrification media can become overcrowded with heterotrophic biofilms, which lowers the efficiency of nitrification bioreactors. Media recovery is generally done in separate MBBRs, where the regenerative (or recovery) treatment consists of flowing water with near-zero DOC and [TAN] ≤ 2mg/L. Nitrifying media are maintained in the "recovery-MBBRs" until heterotrophic biofilms have been removed and a stable nitrification rate is restored (up to 1,740 g TAN $m^{-3} \cdot d^{-1}$). In this model, we assume that the recovery MBBRs are approximately 30% higher than the RAS' main MBBRs. Estimated nitrification costs for a RAS producing up to 1,010g of TAN per day are: \$660 for the MBBR moving bed media (at \$220/CBM), \$2,000 for the MBBR reactors (including aeration equipment), plus 30% for the recovery MBBRs = \$2,660 + \$2,660 • 0.3 = \$3,458. Assuming a lifetime for the nitrifying equipment of 5 years, the nitrification expenses are \$8.6 per year for each kg of TAN processed. Model users can alter these cost assumptions for specific local conditions.

HOS biofilter costs

We have estimated the costs of a HOS biofilter in the form of a flatland with raised earth margins (dikes) lined up with EPDM foil. This biofilter is also enclosed (i.e., with metal posts and chicken wire) and shaded (by means of UV-resistant greenhouse cloth). Assuming a lifetime of 30 years for an EPDM-lined wetland, we estimated the HOS costs to be approximately 1.4 \$ $m^{-2} \cdot yr^{-1}$. Model users will input new data for each situation, as many types of outdoor phytofilters may exist.

Water inflow and water recirculation costs

Agricultural water rights (i.e., the price of water) were estimated to be \$0.01m^{-3}, but this value will vary among farms. The cost of pumping water towards or within the system is:

$$\mathbf{CP = (Q \bullet g \bullet H \bullet time) / Peff / (3.6 \bullet 106) \bullet E\$} \text{......................(6)}$$

where: CP = cost of pumping water [$]; Q = water flow ($kg \cdot h^{-1}$); g = gravitational acceleration ($9.81\ m \cdot s^{-2}$); H = pumping head [m]; Peff = the pump's efficiency (%); time = duration of operation (hrs.); E$ = the price of electricity (e.g., $0.12/kWh).

Pumping efficiency will vary with the pump type. It ranges between 35–40% in airlift pumps and 80–85% in impeller pumps. Centrifugal (radial flow) pumps have intermediate efficiency, controlled mostly by the motor used.

Zeolite costs

Zeolites are sometimes used in aquarium systems as an emergency measure. The model assumes that zeolites remove on average 15mg $NH_4^+ \cdot g^{-1}$ [29]. If trout excrete 87g of TAN until they reach 1kg in weight, and if only 10% of this NH_4^+-N is extracted with zeolite, up to 464kg of zeolite are needed by a RAS facility producing 800kg of trout. Though it can be recycled many times, at a price of $2kg^{-1} zeolite alone would cost $928. If the zeolite material will last 10 years, the cost of zeolite in these simulations is then $92.8yr^{-1}. Commercial zeolite granules for aquaculture have a density of approximately 0.88kg/L and a void space close to 50%, meaning that 1kg of zeolite will have 0.57L of void space. Regeneration of the zeolite is done with a solution of 0.5M HCl, followed by dH_2O washes [30]. We assumed that each zeolite regeneration cycle required two acid treatments and ten water washes. In these conditions, regenerating 1kg of zeolite requires 1.14L of 0.5M HCl, equivalent to 47mL of conc. HCl. At $53 per 2.5L of 37% ACS grade HCl, the acid needed to wash the zeolite is 1\cdot kg^{-1}$. The dH_2O produced by distillation costs approximately $70 per 1,000L. If 5.66L of dH_2O is needed to wash 1kg of zeolite, then $0.4 worth of dH_2O is needed. From the above, the estimated cost of regenerating the zeolite is ≈$1.4/kg. As 1kg of zeolite will remove 15g of RAS NH_4^+-N, zeolite costs are ≈$0.093 per g of ammonium removed, which adds $0.81kg^{-1} of fish produced. For these reasons, zeolite is not considered a dominating means to remove ammonium waste in RAS, yet it can become important as an emergency measure, namely to "shave off" toxic NH_4^+-N spikes and overall remove only a few % of the NH_4^+-N produced in a RAS farm.

Effluent cleanup costs

Associating the management of N from RAS wastewater with a dollar amount can be done either through the benefits of N fertigation or through the costs of N removal. If liquid dispersion towards local cultures is feasible, then one would inquire about the price of the commercial fertilisers being replaced. E.g., rice fields need approximately 50kg of N per hectare between the green ring phase and the 1.3cm internode elongation phase [31]. One can estimate the worth of the effluent N relative to the price of conventional fertilisers (e.g., $0.86kg^{-1} of urea-N). In this model, a RAS-alone facility producing 800kg of trout will release a total of approximately 70kg of Min-N (i.e., TAN + NO_2-N + NO_3-N). This N is worth $60 as fertiliser, sufficient to fertigate a 1.4ha rice field. Though appealing on paper, the direct application of aquaculture wastewater to field crops is not trivial. The timing of fertilisation is also important, as are the concentration of other nutrients, field proximity, and water transfer costs.

For N removal from wastewater, most small RAS farms cannot afford high-tech water treatment plants. In such farms, N cleanup from effluents can yet be done with simpler systems such as mechanical filters, biofloc reactors, algae, or constructed wetlands [4]. The size of a constructed wetland biofilter (CWB in Fig. 1) depends on the amount of nutrients to be removed, seasonal changes, and the wetland plants used. In this model, the cost of effluent treatment in CWB is derived from the total amount of Min-N released from RAS, the cattail's biofilter efficiency in the CWBs (approximately $52.5m^2 \cdot kg^{-1} N \cdot yr^{-1}$ in USDA Hardiness Zone 6a continental temperate conditions), the price of lining up the CWB with bentonite (1\cdot m^{-2}$) and the estimated lifetime of a CWB system (approximately 30 yrs).

Results and discussion

Results and Discussions are organised in three sections:
(i) Model verification in the RAS-alone state
(ii) Advantages and limitations of indoor hydroponics
(iii) Combining RAS with outdoor hydroponics

Here, costs are only estimated; they are only helpful at interpreting trends in response to N-management activities. E.g., large differences can exist in the cost and availability of freshwater, in the type and price of energy, in the technical specifications of the nitrification biofilters, and in local law enforcement and environmental limits. Without local specifics, one cannot generalise from simulations or venture economic sustainability analyses. Instead, one can ask, how would a type of activity influence costs? Can it be efficacious, relevant, or unaffordable? The ultimate goal here is to analyse whether the coupling of salmonid RAS farming with HOS biofilters can at the same time be affordable and constrain pollution.

RAS-alone simulations as Negative Control

The model file "Model Plants in Cold Aquaponics" (see Materials and Methods) follows 15 months of fish growth at one-minute intervals (i.e., 657,007 steps). It produces 800 kg of trout in a facility with $22m^3$ of water in a RAS-alone configuration and $54m^3$ in a RAS/hydroponic configuration. Water is distributed as follows: $10m^3$ in the fish tank, $5m^3$ in a decanter/clarifier, 2-$4m^3$ in 1-2 physical filters, $5m^3$ in a buffer tank, and $30m^3$ in the hydroponic unit in RAS-alone runs. Variables that mostly influence the costs of controlling MinN are water flow, nitrification, zeolite-based ammonia scrubbing, and temperature. The fish amount, feed amount, FCR, and TAN excretion will not change between these simulations.

In this set of simulations, the hydroponic paths (HIS and HOS) are turned OFF, the water renewal rate is $10\%\cdot d^{-1}$ relative to the fish tank, and the water turnover time in the fish tank is $0.25h^{-1}$. As this is a type of discrete (i.e., stepwise) modelling, the capacity of the water reservoirs and the time intervals should not be changed arbitrarily. The step-interval ($1min\cdot step^{-1}$ is already long relative to the recirculation flow rate (i.e., > 666L $step^{-1}$) and the capacity of some reservoirs (some only 1,000L). If fluctuations exist in the model, calculating the concentration of chemicals may on occasion lead to division by zero, and simulations will freeze. In this model, fish are grown for 15 months (456.3 days) from Wi = 0.1g to Wf = 1,000g, equivalent to a specific growth rate of $1.4E^{-4}\cdot g\cdot step^{-1}$ (Eq. 1). Before simulations begin, the following parameters have to be checked:

- the converter “Months per growth cycle” = 15;
- the converter “Wi = Initial weight (g/fish)” = 0.1;
- the converter “Wf = Final weight (g/fish)” = 1000;
- the stock “Weight per fish (g)” = 0.1;
- the stock “Fish stock (g/tank)” = 80;
- the stock “Minutes from beginning” = 0;
- the converters “ON/OFF Zeolite” and “ON/OFF Nitrifier” = 0 for OFF or 1 for ON;
- the system begins with initial concentrations at 0.001 mg/L for [TAN], [NO_2-N] and [NO_3-N]; this means:
- an initial value of 0.01 for the stock ”TAN in Fish tank (g)”. In simulations where the HIS and HOS biofilters are active the initial TAN values are 0.03 g in the stocks ”TAN in HIS (g)” and ”TAN in HOS (g)”;
- an initial value of 0.01 for the stock ”NO_2-N in Fish tank (g)”. In simulations where the HIS and HOS biofilters are active, the initial NO_2-N values are 0.03 g in the stocks ”NO_2N in HIS (g)” and ” NO_2-N in HOS (g)”; and
- an initial value of 0.01 for the stock ”NO_3-N in Fish tank (g)”. In simulations where the HIS and HOS biofilters are active the initial NO_3-N values are 0.03 g in the stocks ”NO_3-N in HIS (g)” and ” NO_3-N in HOS (g)”.

- In the main menu verify under ”Model”, “Run specs” verify that “Start time” = 1, “Stop time” = 657007 and “DT” = 1.

Users can select which parameters to monitor in either table or graph format. One simulation run will take approximately 14 minutes. As predicted, the total amount of TAN produced through fish excretion is 69.6kg. If the nitrification biofilter and the zeolite scrubber were inactive, 56.4kg pf TAN would have been released into the environment, with 19% of TAN still resident in the system at the end. Such a protocol is unsustainable for a fish farm because if the buildup of ammonium is not controlled, the [TAN] would reach fish-unsafe levels (i.e., > 1.3mg/L) during the 226,531th step during the 6th month, when fish are only < 4g in weight and only < 3.2kg of fish have been produced. If the nitrifying biofilter is turned ON, the 1.3mg/L [TAN] level is reached at steps 615,630, during the 14th month of growth (29 days away from the planned harvest). In this case, the [NO_3-N] will also rise above the 70ppm toxic level during the 12th month at step 513,037 (i.e., 100 days before harvest). Since capturing 10% (0.7kg) of the NH_4^+-N produced with zeolite adds ≈ \$84 in zeolite regeneration costs, the zeolite filter was set to only function in the last month and to only absorb up to 1% of the TAN ever produced. In order to avoid wasting zeolite unnecessarily when the concentration of ammonium is not toxic, the zeolite was set up to start operating only when the [TAN] reaches the > 1.3ppm threshold. At uniform 10%·d^{-1} water renewal, the cost of freshwater was approximately \$9.82 + \$6.42 = \$16.24. Yet, even if both nitrification and zeolite circuits are open, the RAS water in the negative control simulations will become toxic before harvest. As the number of fish, or feed, cannot be decreased without a loss in revenue, the only alternative to further limit the toxicity of N compounds is to gradually increase the water renewal rate. But this would not solve the environmental pollution problem.

Indoor Aquaponics (Hydroponic Indoor System) as a Positive Control

Combining RAS with a hydroponic biofilter is thought to influence the cost of N management by reducing the need for renewal water, lowering zeolite costs, requiring fewer additional filters downstream, and reducing effluent cleanup. Here, RAS/HIS is a positive control to better understand the N-management of RAS/HOS. Hence, expenses particular to indoor hydroponics, such as hydroponic fertilisers, artificial illumination, indoor climatization, pest control, labour, and plant nurseries, are not discussed. The RAS-alone unit was first verified under various conditions to identify the best timing to open the RAS/HIS connection. This also produced a baseline for N-related costs in RAS and hydroponics.

Most simulation conditions are similar to Section 1, but the HIS flowthrough path opens when "Step to couple the HIS" = ≥1. When the HIS path opens, the NC path is automatically turned off. In this section, the HOS path is closed by inputing a value of 657,008 for the converter "Step to couple the HOS". Unlike in RAS-alone, in RAS/HIS water will also be lost by evaporation from plants (in cattail, this is approximately $1.93E^{-5}$·L·m^{-2} of HIS min^{-1} = 100T·ha^{-1}·yr^{-1}). The amount of water lost by evapo-transpiration is not large here relative to the water inflow, but the capacity to include such losses in the model has to be present. The capacity of HIS to remove MinN (i.e., "TAN uptake HIS", "NO_2-N uptake HIS," and "NO_3-N uptake HIS") was set to 0.5. This ratio can be changed based on the specifics of each fish farm, plant used, and climate. Here, 0.5 means that 50% of the TAN, NO_2-N, or NO_3-N entering the hydroponic section is absorbed every time water flows through it.

For real-life applications, the surface area of the HIS and its biofilter capacity have to be derived based on the plant used, phytomass density, growth rate, and amount of crop harvested. As a model plant for indoor trout aquaponics, we used broccoli, which grows best at 12.8–18.3 °C with an optimum at 15°C and prefers a pH of 6.0–6.5 (https://hydrobuilder.com/learn/hydroponic-broccoli/). Based on the productivity of field broccoli (i.e., 13T·ha^{-1}), 86.4% moisture content, and 35% protein per DW (i.e., 5.6%g N/DW), we derived that 100m^2 of a broccoli crop equivalates to approximately 1kg of N uptake. This underestimates the N uptake of a broccoli HIS culture, as in broccoli only the inflorescence is harvested and because, depending on design, hydroponics can be more space efficient than field cultures. In climate-controlled broccoli hydroponics, plantings are frequently staggered at 3–4-week intervals to generate a continuous harvest. We assume this will happen in our model as well. In our model, the broccoli HIS will be in a continuous and season-independent production mode, whether RAS-coupled or not. This is because indoor aquaponics only makes sense if the rather expensive hydroponic section is also highly productive. We also assumed that the hydroponic broccoli culture was constant in phytomass growth rate and biofilter capability and that hydroponic fertilisers could be augmented as needed. Hence, the HIS from our model has an upper limit biofilter capacity relative to its surface area, and this biofiltering capacity is independent of the RAS/HIS coupling history. Since the HIS-biofilter capacity is capped, we must provide sufficient surface area of HIS relative to the largest MinN released by the RAS. It appears obvious that the RAS/HIS combination will be under a different management paradigm compared to RAS/HOS, whose biofiltering capability per surface area is planned to increase exponentially to mimic the increase in the fish's excretion (Fig. 2).

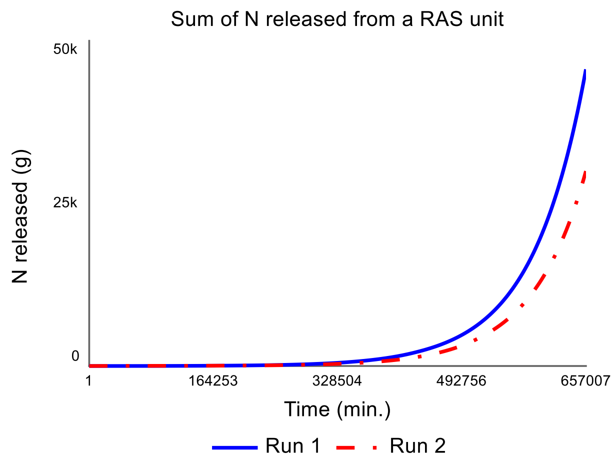

Fig. 2. Total amount of N released in the environment from a RAS system with or without coupling with an indoor plant biofilter (i.e., HIS module in the model). (Run 1) RAS alone and no plant biofilter released a total of 45.6 kg of N into the environment. (Run 2) With HIS plants, coupled to the RAS, released a total of 30 kg of N into the environment. Temperature controlled in the HIS plants module to optimise plant growth efficiency and nitrogen uptake. The 0X axis represents time (15 months of RAS operation with fish growing from fingerling stage to harvest size in 657,000 steps). The 0Y axis represents the total amount of nitrogen released in g.

Simulations began with [TAN], [NO_2-N], and [NO_3-N] at 1μg/L and the water inflow rate was flat (10%·day^{-1}). While the concentration of Min-N chemicals remains low and non-toxic to the fish, there is no benefit to connecting the HIS yet. Without zeolite and HIS, the first time a Min-N toxic threshold occurs is during the 11th month for NO_3-N (at 70mg/L). Unless changes in water chemistry or water renewal rate are made, the onset of the 11th month (i.e., step 438,006) is the latest moment to open the HIS circuit. Then, if the HIS' surface area is sufficiently large, the [TAN], [NH_3-N], [NO_2-N], and [NO_3-N] will not reach toxic levels till the end of the fish's growth cycle. Model users can change the HIS surface area to study effects on safety and costs. Because [TAN] remained < 1.3mg/L throughout the simulation, the zeolite scrubbing system was also not triggered, and its regeneration (≈ \$115kg^{-1} of TAN) did not add to costs. More than 95% of the min-N produced in the system was trapped in the plant's phytomass. Results indicate that adding HIS may also lower expenses for effluent treatment. To summarise, with HIS connected during the last 5 months of a 15-month-long RAS cycle, the costs of N-management were reduced by approximately \$100. Of the remaining costs, approximately 78% came from nitrification and water recirculation (actually not attributable to

HIS per se). Though the HIS is only used for a fraction of the RAS cycle, the HIS biofilter cannot be sterilised after each usage (unlike abiotic filters). This means that a HIS cannot serve as a biofilter for a succession of RASs without posing health risks for subsequent fish stocks.

The main observation from this section is that HIS biofilters can reduce N-related toxicity and will influence the costs of N-management through less water renewal, the elimination of zeolite scrubbers, and less effluent pollution. Yet, in our opinion, indoor aquaponics makes two assumptions that require more research before claiming sustainability. The first is that commercial hydroponics remains profitable even when ammonia and nitrate concentrations are below what is common hydroponic practice if these chemicals are continuously added from RAS. The second is that the biofiltering capability of hydroponic cultures will remain efficient and predictable after long-term exposure to low nutrient concentrations.

Outdoor Aquaponics (Hydroponic Outdoor Systems)

The main challenge of indoor aquaponics is generating sufficient revenue to cover the added costs, which are rather high. This is difficult to achieve because, among others, the increased concentrations of [TAN] and [NO_3-N] necessary for good plant yield are toxic for the fish. However, if we discuss trout, fish-safe levels of [TAN] and [NO_3-N] will not limit the growth of many cold-tolerant wetland plants. In this section, we ask whether simpler outdoor macrophyte biofilters with such plants can solve some of the limitations of indoor aquaponics. HOSs are cheaper than HISs and also simpler to operate; they are less dependent on large revenue streams and, arguably, more robust to nutrient fluctuations. The current HOS plant of choice is broadleaf cattail (*Typha latifolia*), a common inhabitant of wetland environments, from marshland to riparian habitats and from subtropical to cold temperate zones. This model contains differences between HOS and HIS applications. For example:

- The air temperature is constant in HIS and variable in HOS. Hence, while fully climatized HIS may be used as a biofilter throughout the year, HOS will only work during the outdoor plant growing season. For example, cattails survive but do not grow below 10°C. This means that in a temperate climate, a cattail HOS biofilter will only be usable for 6–8 months per year.

- The need for economic sustainability keeps HIS in a high production mode with a constantly high abundance of phytomass and a high growth rate. Relative to HOS, a HIS will likely have a depressed ratio between the smallest and largest biofilter potentials. This narrow performance range can become problematic when estimating the HIS surface area needed for a RAS/HOS combination. While the [Min-N] release from RAS grows exponentially over a very broad range (in $dMin\text{-}N \cdot d^{-1}$), the biofiltration capability of HIS (i.e., $dMin\text{-}N \cdot d^{-1} \cdot m^{-2}$) will increase less. The problem here is that undersizing a HIS can endanger the fish stock, while oversizing it can make the HIS biofilter unaffordable. In contrast, on a given surface area, the biofilter potential of a HOS can start very low and increase exponentially to very large values during an outdoor growth season. This supports the hypothesis that RAS/HOS combinations are tolerant of the HOS's surface area as long as it is above the recommended minimum. Though this will require a good match between the growth rates of the fish and the plants, We predicted that calendar/season matching is an important controller for the sustainability of RAS and HOS systems.

- In this model, the three most important factors that control the growth of the HOS' phytomass are the HOS's surface area, temperature, and N availability. The HOS' surface area is a model input parameter. The evolution of temperature and how the minima and maxima influence growth were explained in Materials and Methods. Nutrient availability is the difference between the concentration of a chemical and its C·min value for a specific type of filter. During each simulation step, the model determines which factor (either T or N) limits phytomass growth during the next step and acts accordingly. Hence, HOS plants will not grow faster than what temperature or available nutrients allow; hence, during a fish or plant cycle, the plants' growth is sometimes T-limited, other times N-limited.

This section includes three types of simulations: (a) RAS/HOS evolution for comparisons with the RAS/HIS results from Section 2; (b) the effect of the HOS' surface area on the RAS/HOS functioning; and (c) the effects of calendar shifting to understand how seasonality and the timing of management decisions influence the RAS/HOS' efficiency.

(a) Here, it is assumed that cattails were planted on March 1st at a density of 100g DW m^{-2} and that the RAS/HOS connection was made immediately. If the fish's growth cycle ends on Sept. 1st (step 657,007), then the RAS/HOS coupling has occurred at the beginning of the 9th month of the RAS cycle (i.e., step 394,205). The HOS' surface area is 500 m^2 (see converter "Hos surface area (m^2)"). This secures sufficient space for the plants to grow relative to the amount of MinN produced in the system (≈ 69.5kg) at a phytomass density peak of 10.2kg DW plants m^{-2}. The Cmin values for both [TAN] and [NO_3-N] were set at 0.1mg/L after [32]; i.e., converters "C·min TAN (mg/L)" = 0.1 and "Cmin NO_3-N (mg/L)" = 0.1. Improved predictive models will require measuring Cmin values more precisely for combinations of various physical-chemical parameters (T, pH, the ratio between [Min-N] and [PO_4-P], and the [dissolved O_2] in the hydroponic root system). This set of factors is a major contributor to N uptake, according to [14] and [22]. Simulations indicated the capacity of HOS biofilters to constrain the TAN, NO_2^- and NO_3^- levels within fish-safe levels under favourable temperature conditions. Plant growth may still occur past the point of RAS harvesting (i.e., September 1st at an average air temperature of 23.5°C). Plant growth was slowed during April due to limited N availability. For the rest of the year, however, it was temperature and not N that constrained phytomass growth.

(b) In earlier simulations, HOS plants had unrestricted growth space (i.e., 500m^2) yet only 65.5kg (i.e., 94%) of the MinN produced by RAS was removed by the HOS biofilter. Of all the N released in the environment (i.e., 4kg), approximately 3kg were released after the HOS was coupled. Nitrogen was removed more efficiently by HOS if the water renewal rate was slowed down after the HOS biofiltering became active. Irrespective of how the HOS biofilter was managed, however, approximately 1kg of N was still released into the environment during the early months of fish growth while the RAS/HOS connection did not yet exist. This, however, represents only about 1.4% of all the MinN produced by RAS. In this set of simulations, the HOS' surface area was also modified to verify that "better occupancy of the HOS with phytomass and less N pollution can be further achieved with even lower costs". At 410m^2 of HOS surface area, the RAS water remained fish-safe, albeit HOS occupancy with plants did not exceed 45.3% due to a lack of N. Also, more N was discarded in the environment (= 36.7kg) than biofiltered (= 32.8kg). Although the HOS biofilter was able to constrain toxic levels of MinN chemicals, two problems were observed. (1st) Increasing the HOS' surface area (from 400 to 1,000m^2) was not well correlated with a reduction in the amount of N in the effluent. This led to the need for larger constructed wetland biofilters (CWB) to remove the excess N in effluents. Because it was T, and not N, that limited phytomass growth, a sizable portion of the bio-available N was not picked up by the plants. (2nd) Increasing the HOS surface area (up to 1,000m^2), to compensate for a poorer growth rate has corrected the N pollution problem but also added costs (at a rate of approximately 1.4 $\$\cdot m^{-2}$ of $HOS\cdot yr^{-1}$). Two solutions may solve this N-pollution problem. Either start HOS with a larger phytomass density, or a calendar shift of the RAS cycle, for better matching between the fish and the plant's growth rate.

(c) In this model, the biofiltration performance of a HOS is proportional to the plants' vegetative growth. In turn, vegetative growth is an outcome of the phytomass' stock and its growth rate. In the fall, as T decreases, a decrease in biofiltration efficiency is also expected, even if phytomass is not yet abundant. Consequently, this set of simulations is a calendar frameshift. The HOS surface area was kept constant (=420 m^2) while RAS/HOS coupling occurred at different moments in the RAS's growth cycle. The HOS vegetation is always planted on March 1st, but the RAS cycle will stop sooner or later during the year. The simulations' scope here is to determine whether a calendar shift may improve the RAS/HOS system in three respects: (1) protection of the fish stock from toxic N chemicals; (2) pollution with N in effluents; and (3) N-management costs. The results of 11 simulations are summarised in Table 1.

Table 1. Results of simulations to analyse the effect of a calendar shift in the RAS' endpoint when the timing of the RAS/HOS connection was time-shifted relative to the plants' growth season

Timing of the RAS/HOS coupling (simulation step & month of the RAS cycle)	RAS end-point	Duration of RAS/HOS coupling (months)	Was a toxic limit achieved? (Y/N) Chemical & Month	N toward the environment. (kg)	N-management related costs ($)
219,003 (6th mo.)	Dec. 31	10	(Y) NO_3 (14.6)	48.1	$1,650
262,804 (7th mo.)	Nov. 30	9	(Y) NO_3 (14.7)	31	$1,630
306,604 (8th mo.)	Oct. 31	8	(N)	10.5	$1,600
312,079 (8.125 mo.)	Oct.27	7,875	(N)	5.52	$1,590
317,554 (8.25 mo.)	Oct.23	7.75	(N)	1.43	$1,580
323,029 (8.375 mo.)	Oct.19	7.625	(N)	1.74	$1,580
328,504 (8.5 mo.)	Oct.15	7.5	(N)	2.19	$1,580
350,405 (9th mo.)	Sept. 30	7	(N)	6.69	$1,600
394,205 (10th mo.)	Aug. 31	6	(N) (NO_3) 40ppm	36.1	$1,660
438,006 (11th mo.)	Jul. 31	5	(N) (NO_3) 68ppm	52.2	$1,700
481,806 (12th mo.)	June 30	4	(Y) (NO_3) 14.8	59.3	$1,720

Results indicated that the timing of this coupling does affect the ability of outdoor macrophytes to protect the fish from toxic N-inorganics and the amount of N pollution, but not the costs of N-management. In this model, the time window to achieve fish safety was about three months (with the RAS endpoint situated between August 1st and October 31st). The time-frame window to limit N pollution was, however, narrow. A performance of < 5% pollution (relative to the MinM produced) only occurred when the calendar timing was accurate within a two-week interval. The RAS/calendar frameshift did not influence the N-management costs by more than 8.2%. Best results were obtained when the RAS/HOS connection was made at the onset of the plant growth season (March 1st), during the 317,554th step of a 15-month-long RAS cycle, and the RAS/HOS connection was maintained for 7.75 months. Under this model's cost assumptions, N-management costs were distributed as follows: 37.9% for nitrification; 37.2% for HOS; 13.4% for water recirculation; and 9.5% for secondary filters. Two potential means to lower the costs of managing N in a RAS/HOS system were identified: (a) extracting revenue from the HOS as well (these costs are about $1.4 m-2); and (b) bringing the water level in the fish section as close as possible to the water level in the hydroponic section. This will lower the cost of water recirculation.

(d) In this series of simulations, comparisons were made between a RAS-alone system and a RAS/HOS combination. The file name for the RAS-alone simulation is "Model Plants in Cold Aquaponics NC". In this instance, hydroponic systems are not yet coupled to RAS, and the zeolite filter is connected but will only be active when [TAN] >1.3 ppm. Again, to constrain expenses, the amount of zeolite is limited so that this filter can only absorb up to 1% of the TAN produced in the system. As shown in Section 1, the least costly means to consume water was the exponential growth of the water renewal rate as the excretion of TAN increased. Hence, in this series, we set the converter "Water regeneration" for an increase from 0.1 to 24 in

657,007 steps, using an exponential growth model with a "STELLA" curvature of 5. Under these conditions, the model predicts that the total cost of constraining the inorganic N chemicals within fish-safe limits will be \$1,870. These were distributed as follows: 27.5% to purchase water; 18% to pump the freshwater inward; 17.4% for water recirculation; 27% for nitrification; 2.7% for zeolite; and 7.4% for effluent treatment. The total water consumption was $4.52 \bullet 10^7$ litters. Near-toxic levels for ammonium were reached in the last quarter of the 15^{th} month but kept under control by the zeolite scrubber. In total, 69.1kg of Min-N were released in the effluent, which is 99.3% of all the TAN produced in the system. Treating this effluent required 3,600m^2 of CWB at an estimated cost of \$121.

These results were compared to those from Table 1 (RAS/HOS). The file name for this simulation is "Model Plants in Cold Aquaponics HOS". For this run, the converters "Step to Couple the HOS" and "Planting Date HOS" were set at 317554 (Fig. 3).

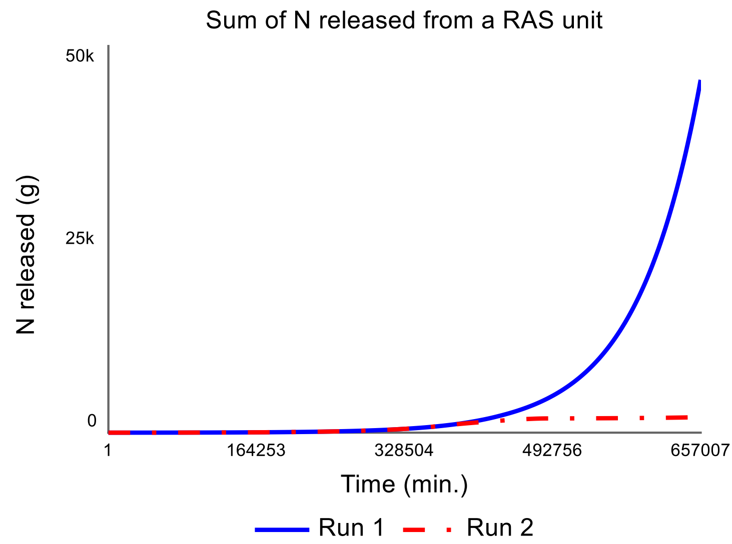

Fig. 3. The effect of coupling the RAS unit with an outdoor plant biofilter. The field was planted mid-March (at step 323,029 at a temperature average of 5.5 °C), and the plants were coupled with the RAS on April 1st (at step 350,405 at a temperature average of 12.8°C). (Run 1) Plant biofilters uncoupled, releasing 45.6kg of N into the environment. (Run 2) Plants biofilter coupled, releasing 2 kg of N into the environment. The temperature averages were typical for continental temperate climates at 45° parallel, with the following average high temperatures: March 5.5; April 12.8; May 21.1; June 23.9; July 26.7; August 25; September 21.1; October 15.5; November 10.

This is equivalent to a RAS functioning alone for 7.25 months (from a total 15-month cycle), then coupled with the HOS on March 1^{st} and fish harvested in the last week of October. The converter "ON/OFF zeolite" was set to 1, to allow the zeolite to function when [TAN] > 1.3ppm. The "water regeneration" converter was set at $0.1 p \cdot day^{-1}$, equivalent to a renewal rate of 1,000L per day. In these conditions, total costs of N management were 15% lower than in RAS-alone simulations (i.e., \$1,580) and distributed as follows: 1.13% to purchase water; 0.74% to pump in the water; 13.4% for water recirculation; 37.9% for nitrification; 0% for zeolite; 0.16% for effluent treatment; 9.5% for Filter 2; and 37.2% for HOS. The total water consumption was only 3.8% relative to the RAS-alone system. Toxic levels of N chemicals were never reached, and the automatic zeolite scrubbing was not triggered. In total, 1.42kg of Min-N was released in the effluent, or 2% of all TAN produced. Treating this effluent required 75m^2 of CWB at an estimated cost of \$2.5.

Without field data from a specific RAS or HOS operation, one cannot draw definitive sustainability conclusions from such models alone. However, this model showed that savings from HOS biofilters can be larger than the HOS' costs and that a sizable benefit of RAS/HOS combinations may come from constraining toxic N chemicals within fish-safe limits, with the potential to eliminate ammonia scrubbers and sizable reductions in renewal water needs and downstream pollution. In our opinion, with proper planning and management, outdoor hydroponic biofilters can be beneficial for both RAS and environmental protection.

Simulation results have shown that increasing the water renewal rate in RAS but without plant biofilters is water wasteful and pollutes the environment with N. Because results will vary with the design choices of each individual farms, it is not possible to draw definitive conclusions about economic success, but only to identify trends in expenses and in the costs to conform with local pollution limits. In RAS/HIS farm configurations, the main sustainability concern is generating sufficient revenue to cover expenses of indoor climatization. In such cases, extracting additional revenue from vegetal crops is a must, and phytofiltration becomes an added benefit. Due to the need for plant growth optimisation, the sustainability of aquaponic RAS with HIS requires supplementation with fertilisers and adjusting the NH_4^+/NO_3^- and N/P ratios. The phytofiltration potential of HIS will increase with nutrient availability but will seldom follow the exponential growth profile needed to mimic the RAS's ammonia emission. In temperate climates, HOS-cultured plants will start small and less active in the spring, and the phytofiltration capacity will increase exponentially towards the summer's end. The time window to maintain a RAS/hydroponics connection open will vary between RAS/HIS and RAS/HOS architectures. In temperate climates, this window of time may be extended for RAS/HIS combinations and remain seasonal for all RAS/HOS combinations. Relative to HIS, which is better amenable to climatization, in HOS the air temperature will be more variable than water temperature. Since HOSs do not involve climatization (for weather protection, illumination, temperature, and humidity), HOS facilities will cost less, and the needs to cut costs will also be diminished. This also makes the phytofilter function of the HOS plants a more realistic priority.

Both HIS and HOS can benefit RAS farms by overall decrease in water consumption, by eliminating the need for ammonia scrubbing, and by reducing N pollution through effluents (up to tenfold in some cases). One notable disadvantage of HIS is the space needed relative to RAS. We caution that it is not safe to reduce the space needed for HIS by reusing the same HIS as a biofilter for a time succession of different RAS batches. Such strategy is likely to increase the risk of spreading fish pathogens among batches. Other disadvantages of HIS include sizable investment costs, high operational complexity, stringent cultivation conditions needed to achieve economic sustainability, high expenses of indoor climatization, and mismatch between the [TAN] and [NO_3N] levels that are at the same time fish-safe and plant-optimal. Simulations indicate that indoor hydroponics may not be at the same time profitable and an efficient biofilter with regards to curbing effluent pollution.

In the case of HOS, if no revenue from plants is aimed at, the question of sustainability of a RAS or HOS operation reduces to "financial benefits vs. costs". Tangible benefits of HOS phytofiltration include reduced water usage and reduced need for effluents cleanup. The RAS/HOS management must focus on two problems: decreasing water renewal and pollution reduction. The efficiency of a HOS biofilter largely depends on the plants' planting calendar. Though this timing will vary among geographic locations, our simulations (for temperate, continental, and cold water conditions to grow freshwater salmonids) indicated that differences between "an efficient RAS/HOS" and a "RAS/HOS not worth doing" may require a calendar matching within +/- 2-4 weeks accuracy. In a cold temperate climate, exponential increase in a HOS biofilter's performance over 7.5-8 months may not be achievable with a single plant species. Achieving such performance may require combining two or more hydroponic cultures. For example, lettuce and spinach in the spring; cattail and water mosses in the summer; and broccoli, winter cabbage, and other late cruciferous crops in the fall. Yet, combining different plant cultures for an exponential biofiltration effect is also challenging and will require more in depth analyses.

Of interest for salmonid RAS/HOS farms are plants that are "physiologically divergent" with regards to the temperature preferences between subaerial and aerial parts. Riparian subalpine plants, such as butterbur and wasabi, are good experimental examples for this phenotype. The plants used for salmonid RAS/HOS will prefer cold and steady root temperatures, and their aerial parts should be more tolerant to variable temperatures. Some plants, such as wild rice, mountain bamboos, nasturtium, cattail, willows, and hemp, will tolerate lower root temperatures and grow faster during summer months.

Conclusions

We modelled patterns of nitrogen management costs in salmonid RAS farms with the scope of aligning business sustainability with the need to constrain the environmental impact of aquaculture wastewater. This was meant to make fish farming environmentally friendly and to protect biodiversity and downstream ecosystems from eutrophic pollution. This series of simulations compared the merits of indoor vs. outdoor hydroponic-type filters i.e., HIS vs. HOS, respectively. Both HIS and HOS biofilters can have the capacity to lower the toxicity of N chemicals to fish-safe limits. Simulation results indicated that RAS/HOS architercures have *two environmental benefits: (a) saving water and (b) reducing downstream pollution thus contributing to the conservation of biodiversity*. Under comparable simulation conditions:

(a) The fish stock from RAS/HOS is safe from toxic [NH_4^+] and [NO_3^-] with a 26-fold reduction in water renewal needs relative to RAS-alone. However, achieving such level of reduction in water consumption is greatly controlled by how the plant biofilter is managed.

(b) The second benefit comes from releasing wastewater with fewer macronutrients (nitrogen in this case). The HOS biofilters may be able to remove up to 98% of the mineral nitrogen derived from fish excretion. In the last 6-7 months of a RAS cycle with a HOS biofilter the concentration of NH_4^+ and NO_3^- in the effluent can be close to the Cmin values for the plants to be used. Because the concentrations of soluble N (ammonia, nitrite, and nitrate) in liquid effluents are low, the most efficient means to treat RAS/HOS wastewater is putatively constructed wetlands rather than water treatment stations.

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