

The nutrient pollution rate (or the rate at which nutrients must be removed from the wastewater) can be described by Equation 1, which is simply a mol balance over the hydroponic outlet:

$$R_i = C_i Q \quad (1)$$

Where, R_i is the discharge rate of nutrient i (in units of mol per time) from the hydroponic unit, C_i is the concentration of nutrient i in the solution (in units of mol per volume), and Q is the volumetric discharge rate from the hydroponic unit (in units of volume nutrient solution per time), which can be interpreted as the frequency of solution replacements.

Although this wastewater is often treated (via reverse osmosis, for example), the space and energy requirements of these treatment technologies incurs their own environmental costs. To minimize environmental impact, the objective would be to minimize R_i . Q can be reduced through more extensive recycling of the nutrient solution and C_i can be reduced via concentration control strategies. By controlling nutrient concentrations, Q is effectively reduced since the same nutrient solution can be used continuously until inert build-up dictates replacement. R_i is then dependent on the nutrient concentrations at the time of replacement. Therefore, to minimize R_i , concentration control of N and P at low levels is key.

Nutrient concentrations in the discharge solution (C_i) for N and P have been reported in the range of 15 to 21 mM and 1 to 3 mM, respectively (Gagnon *et al.*, 2010). These levels are around 2 orders of magnitude higher than the concentrations at which half of the maximum nutrient uptake rates would be observed (Michalis-Menten constant K_M) (Akhtar *et al.*, 2007; Kuzyakov & Xu, 2013; Le Deunff *et al.*, 2019; Lefebvre *et al.*, 1990; Wang & Shen, 2012). Therefore, N and P concentrations could theoretically be controlled at much lower levels (and hence achieve proportionally lower pollution rates) without affecting plant growth.

The most common method of nutrient concentration control is through using the electrical conductivity (EC) of the solution (Christie, 2014). The EC is near-proportional to the total amount of nutrients in solution and thus controlling the EC at a setpoint (by dosing additional nutrients) controls the total nutrient concentration. This strategy is relatively cheap and robust. However, the EC does not supply information about individual nutrients. Consequently, the build-up of salinity and inert species in the medium renders the EC signal less reliable. Also, the nutrients

being dosed to maintain the nutrient concentrations must be added in near-exact proportions to which the plants consume them, else some nutrients will deplete whilst others accumulate. As such, nutrient concentrations must be controlled at relatively high levels to ensure that individual nutrients do not deplete without warning. Therefore, although Q is minimised, C_i (for N and P) remains relatively high. Other strategies have made use of ion-selective-electrodes (ISE), which are able to measure individual nutrient concentrations (Cho *et al.*, 2018). For example, a nitrate ISE can directly measure the nitrate concentration in solution and hence additional nitrate can be dosed when the nitrate concentration drops below a setpoint value. Therefore, the nitrogen pollution rate can be minimized by minimizing the nitrate concentration in solution. However, ISEs are generally much more expensive and exhibit practical limitations such as signal drift, reduced accuracy over time, and interference from other ions in solution (Kim *et al.*, 2013).

The novelty of the current work lies in the use of pH measurement (instead of using EC or ISEs) as the sole controller-input to maintain N and P concentrations at low levels. Given the drawbacks of the EC and ISE methods (discussed in more detail in Chapter 2), a pH-based control system provides hydroponic operators with an alternative. pH measurement and control is standard protocol in most hydroponic systems and therefore these control systems are affordable and can be easily implemented. Control schemes were designed for phosphate (plant phosphorous source), nitrate and ammonium (plant nitrogen sources), individually. Phosphate concentration control is considered in chapter 4 exclusively, directly after the Theory and literature and Experimental chapters (chapters 2 and 3). The remaining three chapters deal with nitrogen. Nitrate is the most common nitrogen source supplied to plants and is considered in chapter 5 exclusively. The following chapter (chapter 6) concerns ammonium. Ammonium is important from an environmental perspective since most organic fertilizers contain large fractions of ammonium (Chan-Pacheco *et al.*, 2021). Wastewater from the anaerobic-digestion process is used as the model ammonium-rich wastewater (termed digestate) in chapter 6 (Koszel & Lorencowicz, 2015). Nitrification of ammonium is commonly carried out since ammonium is toxic to plants when supplied as the sole nitrogen source (Hachiya & Sakakibara, 2017). Also, nitrifying bacteria inevitably establish in hydroponic systems when ammonium is present and thus their effects cannot be ignored. When nitrification occurs within the hydroponic unit, the process is referred to as *internal* nitrification. This is the process considered in chapter 6. *External* nitrification is when ammonium is nitrified in a unit upstream to the hydroponic system. This method has many merits and therefore the following chapter (chapter 7) is

devoted to the external nitrification of ammonium as a pre-treatment step. The last chapter (chapter 8) is a general discussion and conclusion on the results from the previous chapters, specifically, the integration of the control schemes developed. The outline of the chapters discussed above is presented graphically in Fig. 0 below.

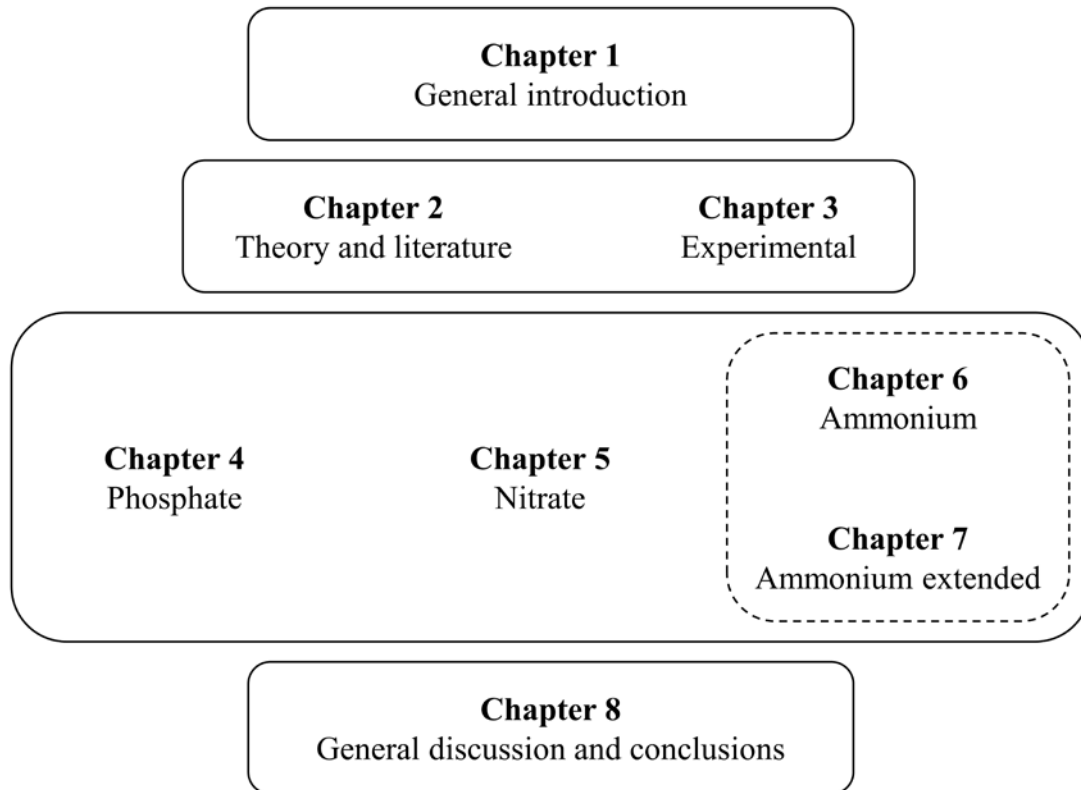


Fig. 0: Graphical outline of chapters.

CHAPTER 2 Theory and literature

2.1 Plant nutrition

The carbohydrate backbone of a plant is built during photosynthesis from carbon dioxide and water. Other elements are also required to build specialized molecules such as proteins (which contains nitrogen) and DNA (which required phosphorous). These elements are absorbed from the soil through the plant roots. Table 1 summarises these essential elements and the chemical forms in which they can be absorbed. The compounds of the first six elements (N, P, K, Ca, Mg and S) are known as macronutrients, since they are absorbed in much higher quantities as compared with the remaining compounds, known as micronutrients.

The first published hydroponic nutrient solution was formulated by Hoagland and Arnon (1938), which contained all the nutrients listed in Table 1 (in rough proportions to which plants absorb them). Nickel was not explicitly added as it was not yet known to be an essential plant nutrient (Brown, 1987). Its necessity was unnoticed due to its very low requirements and was likely present as a trace contaminant in the other nutrient salts used. Hoagland's solution contains N, P, K, Ca, Mg and S (macronutrients) in molar parts of 15 : 1 : 6 : 5 : 2 : 2. Other nutrient solution formulations followed (Hewitt, 1966; Cooper, 1979; Steiner 1984), but these varied only slightly in terms of the total nutrient concentration and the ratios between nutrients. Hoagland's solution is generally suitable for optimal plant growth and is still widely used. The macronutrient proportions of Hoagland's solution are shown in Fig. 1 (left-hand-side chart). Note that the ionic charges of each nutrient (see Table 1) are also shown in Fig. 1 (right-hand-side chart). This will be further discussed in later sections, but the importance lies in the choice of nitrogen source. Since nitrogen makes up around half of the total nutrients absorbed and can be supplied as either a cation or anion, the choice of nitrogen source dictates the overall charge

absorbed. As electrical neutrality must be maintained in the solution, the plant must exude the net charge absorbed (De Wit *et al.*, 1962). For example, if ammonium is supplied as the sole nitrogen source, the net charge absorbed is positive and the plant must exude additional cations (typically H^+) to maintain electrical neutrality. For this reason, nitrate is often supplied as the primary nitrogen source since a lower net charge is absorbed.

Table 1: Essential elements required by plants together with the molecular form in which they can be absorbed (Mahler, 2004).

Essential element	Absorbable forms
N	NO_3^- , NH_4^+
P	$H_2PO_4^-$, HPO_4^{2-} , PO_4^{3-}
K	K^+
Ca	Ca^{2+}
Mg	Mg^{2+}
S	SO_4^{2-}
B	H_3BO_3 , $H_2BO_3^-$, BO_3^{2-}
Cl	Cl^-
Cu	Cu^{2+}
Fe	Fe^{2+}
Mn	Mn^{2+}
Mo	MoO_4^{2-}
Zn	Zn^{2+}
Ni	Ni^{2+}

2.2 Nitrogen

Proteins differ from carbohydrates by the presence of a nitrogenous functional group (amine). Most organisms higher up on the food chain consist primarily of proteins (besides water), most of which (specifically, the amino acids building blocks) were originally produced by plants. The absorption of nitrogen by plants to produce proteins (including other specialized molecules such as DNA and RNA) is therefore key to life of earth. Unlike the other nutrients listed in

Table 1, nitrogen originates from the atmosphere (N_2). Plants cannot use this abundant source of nitrogen directly and instead rely on microbial processes that convert the N_2 into NH_4^+ (Burris, 2001). These bacteria are called nitrogen fixing bacteria and they have various relationships with plants. The bacteria depend on the organic carbon produced by the plants and in turn fixates N_2 for the plants (Miles *et al.*, 1992). A direct symbiotic relationship exists between nitrogen fixing bacteria and legumes, where the bacteria establish within the plant root (Wang *et al.*, 2018). The plant feeds the bacteria with carbonaceous molecules and the bacteria releases NH_4^+ for the plant to use. Other plant species have less direct relationships with plants, such as associative nitrogen fixation. In these relationships, the bacteria are found on the roots or in the root vicinity (rhizosphere) and consume the organic carbon exudated from the roots (Miles *et al.*, 1992).

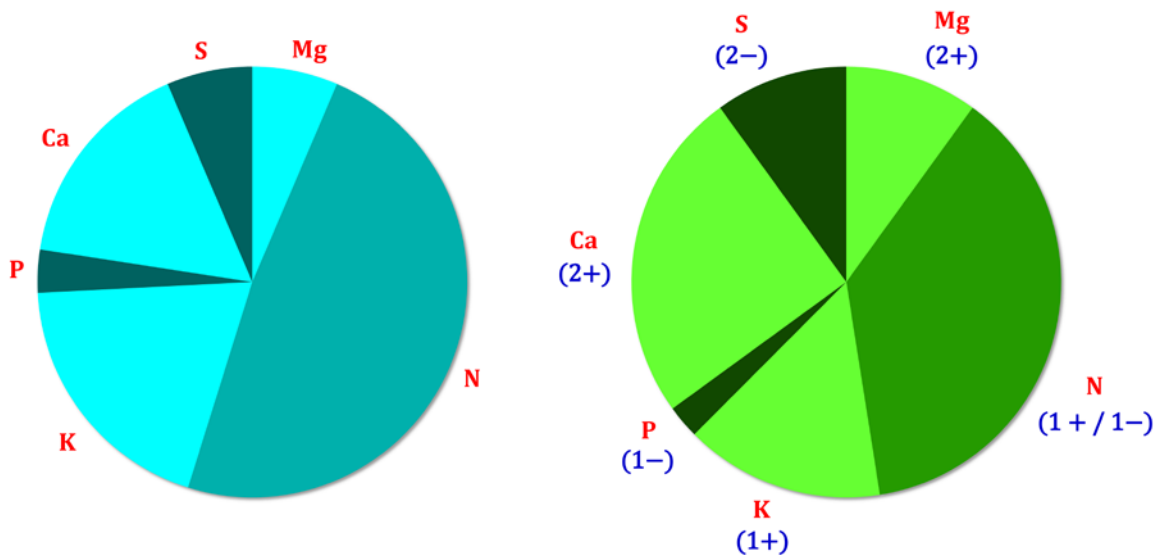


Fig. 1: Left-hand-side pie chart: Molar fractions of the macronutrients in Hoagland's solution, which are roughly in proportion to the rates at which plants absorb them. Actual molar concentrations are 15, 1, 6, 5, 2, 2 mM for N, P, K, Ca, S and Mg, respectively. Right-hand-side pie chart: Valency/charge fractions (absolute, regardless of $+/-$) of the macronutrient. For example, Hoagland's solution contains 5 mM Ca, which equates to 16 mol % of the total macronutrients as shown in the left-hand-side pie chart. Ca has a charge of 2+ and therefore constitutes 10 mM valence electrons, which is 25 % of the total valences as shown in the right-hand-side pie chart. Note that P is given the charge of -1 since $H_2PO_4^-$ is predominantly absorbed (see Table 1).

used, producing 10 000 Lux at the canopy. Kale seeds (*Brassica oleracea* var. *Sabellica* or Vate's Blue Curled Kale) were purchased from Raw™. The main recirculation pumps (responsible for the ebb-and-flow mechanism) were purchased from Xylem™ (“Flojet Diaphragm Electric Operated Positive Displacement Pump, 3.8 L min⁻¹, 2.5 bar, 12 V DC”). For seedling propagation, aeroponic systems (Aeroponic Cloner) purchased from hydroponic.co.za™ were used. DoPhin® “14 in 1 nitrifying bacteria” was used as an inoculum. Evolution Aqua Kaldnes® K1 Media was used for biofilter packing. Four Regent® 9500 air pumps were used for sparging. Chemical analysis of the nutrient solution was done via liquid samples of 2 mL taken daily. Phosphate, nitrate, nitrite, and/or ammonium analysis was done on these samples (depending on the aims of the run) using Merck™ photometric cell tests (phosphate test: PMB 0.0025 – 5.00 mg/l PO₄-P, Spectroquant® nitrate test: DMP 0.10 - 25.0 mg/l NO₃-N, nitrite test: 0.002 - 1.00 mg/l NO₂-N, and ammonium test: 2.0 - 150 mg/l NH₄-N Spectroquant®). The absorbance was measured in a spectrophotometer (Agilent Technologies™, Cary 60 UV-Vis, G6860A) at 690, 340, 525, and 690 nm for phosphate, nitrate, nitrite, and ammonium.

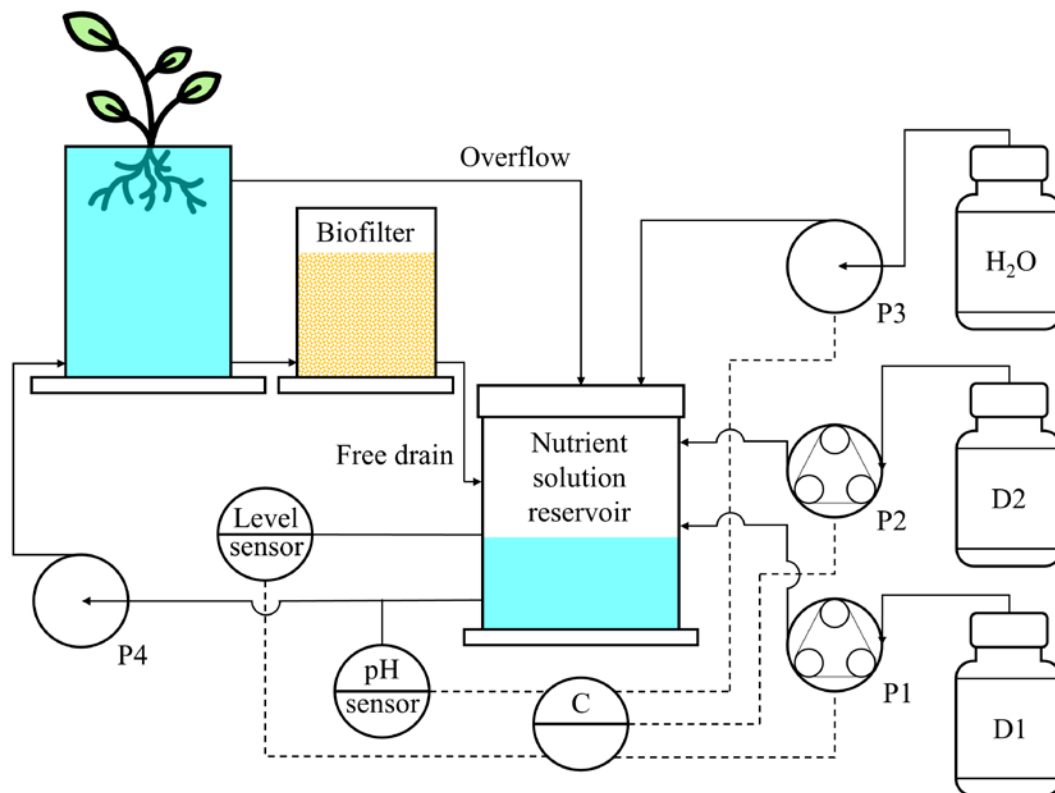


Fig. 6: Simplified process flow and instrumentation diagram of the experimental setup (one of the four) showing major control elements (controller “C”, sensors, and pumps), vessels (plant vessel, biofilter and nutrient solution reservoir), and dosing reservoirs (D1, D2, H₂O).

CHAPTER 5 Nitrate

5.1 Introduction

Plants can absorb nitrogen in the form of nitrate and/or ammonium. In hydroponic systems, optimal growth is often achieved when nitrate is supplied as the primary nitrogen source and many nutrient solutions are formulated with nitrate as the sole nitrogen source (Cooper, 1988; Hewitt, 1996; Hoagland & Arnon, 1938; Steiner, 1984). This is partly because high ammonium fractions can be detrimental to hydroponic plants due to their high ammonium affinity, resulting in excessive amounts of ammonium being absorbed (Pitts & Stutte, 1999). Also, the anionic nature of nitrate results in more-equivalent charge uptake (see Fig. 1), thus promoting balanced nutrient uptake and requiring less cation exudation to maintain electrical neutrality (Bugbee, 2000; Le Bot, 1998). This chapter considers nitrate when supplied as the sole nitrogen source. A pH-based control algorithm was developed to control the nitrate concentration at low levels in solution. As discussed in Section 2.6, nitrate uptake is accompanied by hydroxide exudation which requires acid dosing to control the solution's pH. The premise of this chapter is that a relationship exists between the acid dosing rate required for pH homeostasis and the nitrate uptake rate of the plant. Similar to the approach used by Bugbee (2004) in which the plants' transpiration rates were used to infer the nutrient uptake rates (see Section 2.6), here the proton (acid) dosing rates are used to infer the nitrate uptake rates. With an established relationship, the nitrate concentration can be controlled by dosing additional nitrate at an equal rate to the inferred uptake rates. Since the nitrate concentration is not measured directly, drift in the nitrate concentration (slow accumulation/depletion) is bound to occur. As such, the control scheme was calibrated to effect slow depletion of nitrate in solution. Since depletion ultimately results in extinction, a nitrate extinction prevention algorithm was included, in which nitrate extinction

5.3 Results and discussion

5.3.1. Relating nitrate absorption to proton dosing

Run 1 was conducted using standard Hoagland's solution (high nitrate concentration). The results are given in Fig. 13, in which the hypothetical nitrate concentrations, and proton dosing rates (required for pH homeostasis) are plotted. The hypothetical concentrations are the actual (measured) concentration minus the added amounts of nitrate (manually with a pipette to maintain high concentrations). Fig.13 (c) is a plot of the ratio of HCl dosing to nitrate absorption (η_1 , see Nomenclature), which indicates a constant ratio of proton dosing required for pH homeostasis and nitrate absorbed by the plant ($\eta_1 \approx 0.5 \text{ mol mol}^{-1}$). This relationship provides the means of inferring the nitrate absorption rate from the proton dosing rate.

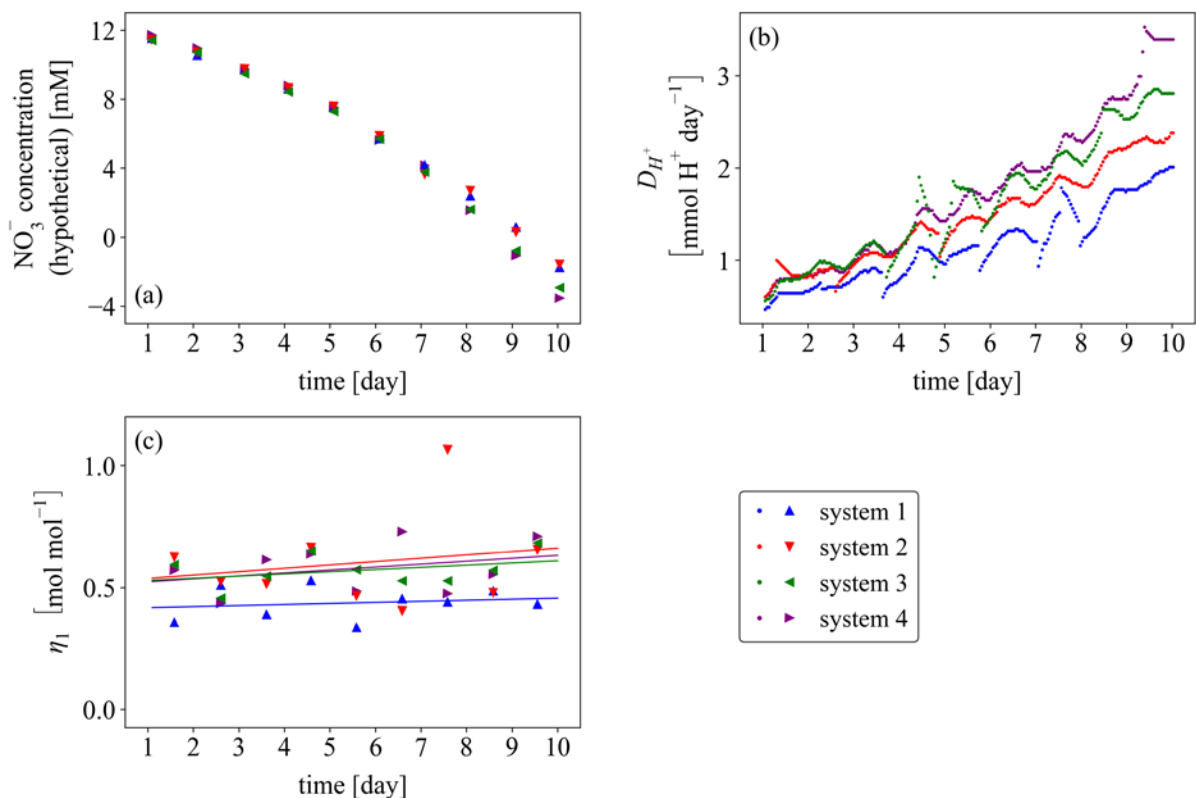


Fig. 13: Results from run 1 in which plants were cultivated in standard Hoagland's solution (nitrate as the sole nitrogen source). The pH was controlled at an average value of 6.1 ($\sigma = 0.07$, $n = 997$). The hypothetical nitrate concentrations (a) and the HCl dosing rates required for pH homeostasis (b), are shown. The hypothetical concentrations are the actual (measured) concentrations minus the added amounts of nitrate (manually with a pipette to maintain high concentrations). Subplot (c) plots the ratio of proton dosing to nitrate absorption (ratio of (a) to (b)), in which a fitted value of $\eta_1 \approx 0.5 \text{ mol mol}^{-1}$ is obtained.

If this ammonium dosing strategy is used, it can be deduced from Fig. 18 that the ammonium dosing rate will be 25 % of the nitrate uptake rate by the plant (by doing a proton balance). This is assuming that $\eta_1 = 0.5$ and $\eta_3 = 2$ according to Fig. 13 and 19, and that negligible amounts of ammonium is absorbed by the plant due to high ammonium oxidation rates. As a result, the nitrate concentration in the solution will decrease over time. Consider however, if instead of dosing ammonium only, a mixture of hydroxide and ammonium was dosed. If the ratio of hydroxide-to-ammonium (δ) does not exceed the value of η_3 (≈ 2), the effect of dosing will remain acidic but higher dosing rates will be realized since the effect is less acidic. Therefore, the decrease in the nitrate concentration in solution will be slower with increasing δ values. It can hence be deduced from Fig. 18 that a δ value of 1.5 will result in ammonium dosing rates being equal to plant nitrate uptake rates. Since all ammonium is converted to nitrate, the nitrate concentration will remain constant (assuming all ammonium dosed is rapidly oxidized).

Run 3 was performed to test this control strategy. Plants were cultivated in the system together with the established biofilter. The pH was controlled by dosing ammonium and hydroxide in a ratio of 1.5 mol mol⁻¹. Ammonium as (NH₄)₂SO₄ and hydroxide as KOH was dosed individually from separate dosing reservoirs (D1 and D2 in Fig. 5 and 6). Hoagland's solution was charged initially with nitrate as the sole nitrogen source. A sequential function chart is given in Fig. 20 to convey the control algorithm. From trial runs, it was found that a constant feed of ammonium was required to maintain the vitality of the nitrifying bacteria. Therefore, the controller dosed 1 mmol day⁻¹ ammonium and 1.5 mmol day⁻¹ hydroxide at 30 min intervals (thus, 1/48 mmol ammonium per instance), regardless of the pH. This is shown as "Low" at the bottom of Fig. 20 in which $D_{NH_4^+}$ is the ammonium dosing rate and D_{OH^-} is the hydroxide dosing rate. When the pH rose above the specified setpoint SP , higher amounts of ammonium and hydroxide was dosed (still in a ratio of 1.5). These rates are abbreviated as "High" in Fig. 20. The "Low" nitrogen dosing rate (1 mmol day⁻¹) is equal to the nitrogen uptake rate of a small Kale plant weighing about 10 g. Therefore, larger plants (as compared with runs 1 and 2) were employed to better observe the control action. The two outermost branches at SP_L and SP_H were added primarily as safety features in case of disturbances or biofilter inactivity. These branches did not play a significant role in the upcoming runs and can thus be neglected, but they are nonetheless recommended.

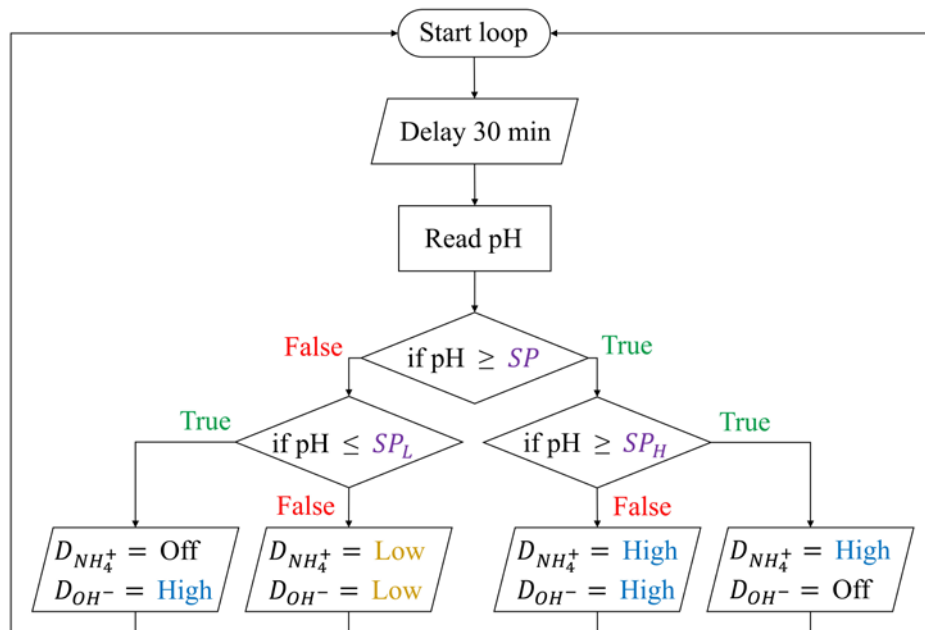


Fig. 20: Sequential function chart of the control algorithm. Online pH measurements were taken every 30 min as shown. High dosing rates of both chemicals were actuated in the constant ratio of 1.5 mol mol^{-1} (as discussed in Section 3.2) when the pH was above the setpoint value $SP = 6.1$, but below a higher setpoint value $SP_H = 6.4$. The “High” dosing rates were proportional to the setpoint error (proportional control action), specifically, $D_{NH_4^+} = \text{“High”} = 30 \times (\text{pH} - 6.1) + 1 \text{ mmol day}^{-1}$ and $D_{OH^-} = 1.5 \times D_{NH_4^+}$. If the pH rose above SP_H (safety feature), hydroxide dosing was halted and $D_{NH_4^+}$ was dosed at a high rate of 7 mmol day^{-1} . If the pH was below $SP = 6.1$ but not as low as $SP_L = 5.8$, low dosing rates were actuated (also in the constant ratio of 1.5). This was necessary to maintain the vitality of the bacteria and prevent inactivation. $D_{NH_4^+} = \text{“Low”} = 1 \text{ mmol day}^{-1}$ and $D_{OH^-} = \text{“Low”} = 1.5 \text{ mmol day}^{-1}$. If the pH was below $SP_L = 5.8$ (another safety feature), ammonium dosing was halted, and high hydroxide rates were actuated (also proportional control), specifically, $D_{OH^-} = 20 \times (5.8 - \text{pH}) + 5 \text{ mmol day}^{-1}$.

The results of run 3 are reported in Fig. 21. Subplot (a) gives the nitrate concentrations of the four systems (triangle markers). No ammonium was detected in the solution (measurements were at the calibration error of 0.1 mM), thus rapid ammonium oxidation was confirmed. The highest measured nitrite concentration was 0.002 mM ; hence ammonia oxidation remained the rate limiting step during nitrification. A slow downward drift in the nitrate concentration profiles is observed. The nitrate depletion rate is around $19 \text{ } (\pm 6 \text{ } \%)$ of the rate in [run 1 Chapter](#)

5 (Fig. 13), in which no additional nitrogen was added/dosed (hence 81 % of the plant's nitrogen in run 3 was dosed as ammonium). Predictions of the nitrate concentrations, if no additional nitrogen was dosed, is also shown for comparison. These predictions are based on exponential fits of the nitrate concentrations in run 1 Chapter 5 (standardised based on initial plant mass). A higher δ value may be employed to reduce the downward drift in the nitrate concentrations of run 3. However, inevitable drift (either up or down) is expected because small errors in pump calibration and genetic variations in the plants and bacteria (regarding the proton/hydroxide to nitrogen uptake characteristics) are bound to exist (Van Rooyen & Nicol, 2021). These parameters are likely dependent on the system and plant species. Therefore, calibration would be required if implemented. Subplot (b) shows the pH profiles for each system. Tight pH control is observed indicating the success of the control strategy. The ammonium and hydroxide dosing rates increased with plant plants size. The results shows that the nitrate concentration in nitrification-hydroponic systems can be maintained at relatively constant levels by controlling the pH with an alkaline ammonium solution at a specific hydroxide-to-ammonium ratio. Similarly, for an ammonium solution at the same pH as that of the hydroponic system, 1.5 mol of hydroxide must be dosed together with 1 mol of ammonium. As an added benefit, this strategy reduces the amount of base required (hence cation build-up) compared to using pre-nitrified ammonium fertilizer only (nitrification occurring in an external unit from the plant growth unit), which requires 2 mol of hydroxide to oxidise one mol of ammonium. In addition, feeding this pre-nitrified solution to the hydroponic system would require additional acid dosing (hence anion build-up) to control the pH.

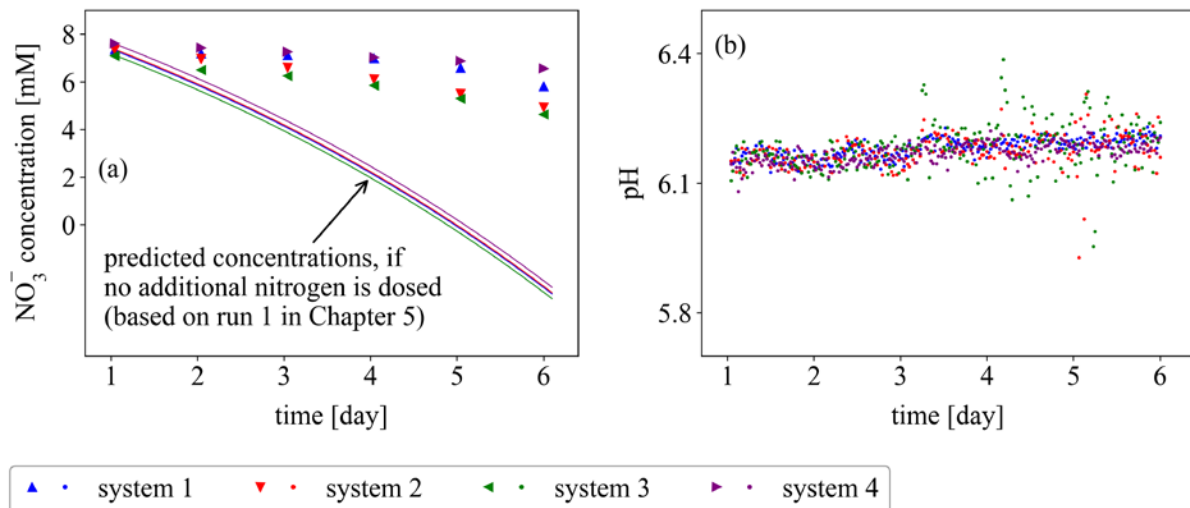


Fig. 21: Results from run 3 in which the nitrate concentration was controlled by controlling the pH with ammonium and hydroxide dosing in a ratio of 1.5 (mol hydroxide per mol ammonium). An overview of the control algorithm is given in Fig. 20. Subplot (a) shows the nitrate concentrations in solution (triangle markers) together with predicted concentrations (based on [run 1 Chapter 5, Fig. 13](#)) if no additional nitrogen was dosed. Subplot (b) shows the online pH measurements (raw) for each of the four systems. All ammonium and nitrite measurements were at the calibration limits of the analytical tests (0.1 mM and 0.001 mM, respectively), which confirms rapid ammonium oxidation and complete nitrification (all ammonium is converted to nitrate).

6.3.3 Controlling the nitrogen concentration at lower levels

Since the nitrogen concentration can be maintained at a relatively constant value (given a slow downward drift in concentration when $\delta = 1.5$), lower operating concentrations may be employed (thus minimizing R_i in Equation 1). Lower nitrogen concentrations can be employed without affecting plant growth or nutrition (discussed in Chapter 1), but lower nitrogen levels mean higher risk of nitrogen extinction in solution. Therefore, good controller performance and robustness is required. Run 4 was performed to determine the feasibility of operating at lower nitrogen concentrations using the proposed control strategy outlined in Fig. 20. Run 4 was conducted under the same conditions as run 3 (also employing a δ value of 1.5), except for charging a lower initial nitrate concentration of 1 mM. The results are reported in Fig. 22 in the same format as in Fig. 21. Like run 3, no ammonium or nitrite was detected.

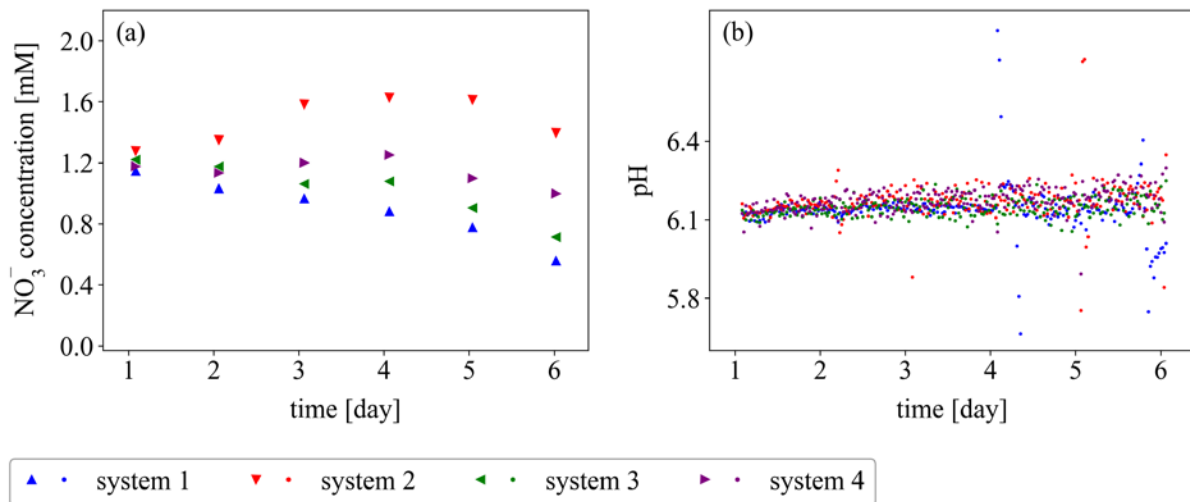


Fig. 22: Results from run 4 in which the nitrate concentration was controlled by controlling the pH with ammonium and hydroxide dosing in a ratio of 1.5 (mol hydroxide per mol ammonium). An overview of the control algorithm is given in Fig 20. Subplot (a) shows the nitrate concentrations in solution (triangle markers). Subplot (b) shows the online pH measurements (raw). All ammonium and nitrite measurements were at the calibration limits of the analytical tests (0.1 mM and 0.001 mM, respectively), which confirms rapid ammonium oxidation and complete nitrification (all ammonium is converted to nitrate).

Similar to run 3, a slow depletion rate in the nitrate concentrations is observed (with a small initial increase in system 2). However, the depletion rates are 3 % (± 3 %) of those in [run 1 in Chapter 5 \(Fig. 13\)](#) which are significantly slower than those of run 3 (which were 19 % (± 6 %) of run 1 in Chapter 5). These rates are reported in Fig. 23 which is intended to compare the growth characteristics between the runs. The relative growth rates (*RGR*) and nitrogen content of the plants are also compared in Fig. 23. As can be seen from Fig. 23, all the *RGR*s are around 0.2 day^{-1} . A significant decrease in the plant nitrogen content is observed for run 5 (ANOVA, $p = 0.06$), which is believed to be due to the low nitrogen concentrations employed. When comparing the nitrate depletion rates of runs 4 and 5, the rates appear to decrease with the nitrate concentration in solution (ANOVA, $p = 0.006$). To explain this phenomenon, mathematical modelling of the flux model depicted in Fig. 18 was undertaken. The model and simulation results are given in the following section, which agrees with the observed phenomenon in which the rate of nitrate depletion decreases with the nitrate concentration. The simulation predicts a steady state nitrate concentration which demonstrates controller robustness. This steady state concentration was not achieved in the current system, however, which is believed to be due to imperfect mixing and system instabilities as observed in the pH and dosing profiles

of the previous runs. However, it was shown that the control strategy effectively safeguards against nitrate extinction in solution.

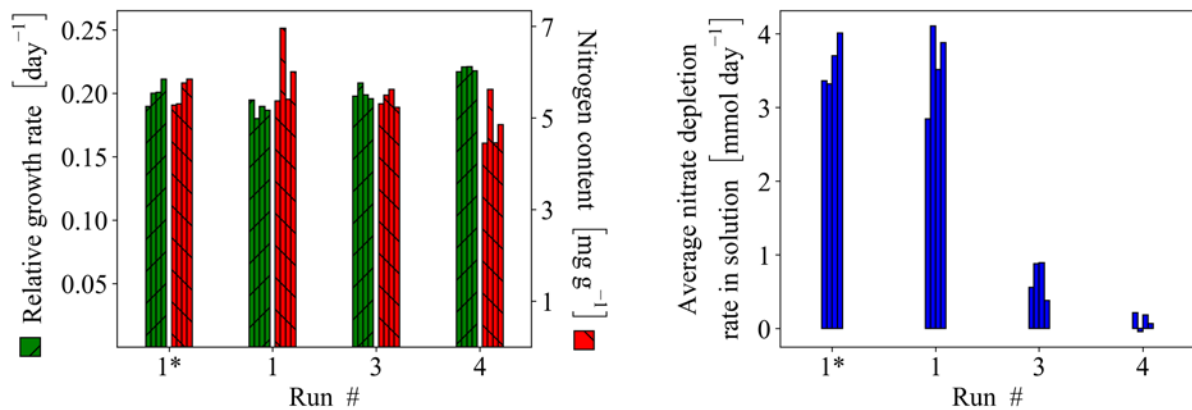


Fig. 23: Comparison of the relative growth rates (*RGR*) of the plants, the nitrogen content of the plants, and the average nitrate depletion rates in solution between runs. Run 1* is from Chapter 5 (Fig. 13) and runs 1, 3 and 4 are from this chapter. The average nitrate depletion rates are standardized based on initial plant mass. The nitrogen content was calculated from a nitrogen mass-balance over the solution and change in plant fresh mass (thus units of mg elemental nitrogen per change in plant fresh mass).

From Fig. 23, it can be seen that plants in runs 1* (Chapter 5) and 1 (this chapter) had similar nitrogen contents. The only difference between the runs was the nitrogen source, with nitrate being supplied in run 1* and ammonium in run 1. This shows that the influx of total nitrogen is regulated by the plant, rather than separate uptake mechanisms for nitrate and ammonium. If the influx of total nitrogen was not regulated by the plant, double the amount of plant nitrogen content would be expected if both nitrate and ammonium is supplied in sufficiently high concentrations. Therefore, efforts to model plant nitrogen uptake should consider the total nitrogen concentration in solution rather than model nitrate and ammonium uptake separately. A suggested correlation is given in the following section (Equation 10), which is intended to explain the observed phenomenon of slower nitrate depletion rates at lower nitrate concentrations.

6.4 Mathematical modelling and simulation

It was shown in the previous section that the rate of nitrate depletion is related to the concentration of nitrate in solution. This may be the result of higher ammonium uptake rates by the plants at lower nitrate concentrations (since the ammonium-to-nitrate concentration ratio in the

solution is higher). To demonstrate, consider the conditions employed in run 3. A δ value of 1.5 resulted in slow depletion of nitrate in solution while the ammonium concentrations remained low. As ammonium uptake relative to nitrate is related to the concentration ratio of ammonium-to-nitrate in solution (Imsande, 1986), higher ammonium uptake rates by the plants are expected when the nitrate concentration approaches that of ammonium. From Fig. 18, it can be shown that if the pH and nitrogen concentrations remain constant: $\delta = (\eta_1 + \eta_2 - \eta_3)F_{NH_4^+}^P + (\eta_3 - \eta_1)$. Where, $F_{NH_4^+}^P$ is the fraction of the total ammonium dosed which is absorbed by the plant (the rest being taken by the bacteria). This function is plotted in Fig. 24 (a) in which $\eta_1 = 0.5$, $\eta_2 = 1$, and $\eta_3 = 2$. Since ammonium and hydroxide dosing is intended to produce an acidic effect (lowering the pH when it rises above a setpoint), high δ values will result in nitrate accumulation and low δ values will lead to nitrate depletion in solution. Therefore, if the δ required for constant nitrate concentration is lower than the actual δ being dosed, nitrate will accumulate in solution and vice versa. So as the nitrate concentration decreases, $F_{NH_4^+}^P$ increases and the employed δ becomes “too high” which tends towards nitrate accumulation. This may be difficult to conceptualize initially and thus it is shown mathematically below by modelling the flux diagram depicted in Fig. 18.

The nitrate balance over the solution at constant volume, assuming all ammonium consumed by the bacteria is converted to nitrate (zero nitrite accumulates and negligible amounts of nitrogen is used to produce bacterial biomass):

$$V \frac{d[NO_3^-]}{dt} = r_{NH_4^+}^B - r_{NO_3^-}^P \quad (6)$$

Where, $[NO_3^-]$ is the concentration of nitrate in solution (mM), $r_{NH_4^+}^B$ is the ammonium oxidation rate by the bacteria which is assumed equal to the nitrate production rate by the bacteria (mmol day^{-1}), $r_{NO_3^-}^P$ is the nitrate uptake rate by the plant (mmol day^{-1}) and V is the solution volume (L) (assumed constant).

The ammonium balance over the solution at constant volume:

$$V \frac{d[NH_4^+]}{dt} = D_{NH_4^+} - r_{NH_4^+}^B - r_{NH_4^+}^P \quad (7)$$

The relationship given in Equation 12 provides the means to develop pH-based nitrification control systems. The objectives of these control systems would be: (1) to maintain operation at the microbial maximum nitrification rate (v_{max}), (2) achieve high conversion of ammonia to nitrate, and (3) adapt accordingly to accommodate variations in microbial activity (robustness). Nitrification can be carried out in various reactor types and the choice depends again on process variables. Therefore, control algorithms were designed for three common reactor types, namely, batch, fed-batch and continuous reactors.

7.2 Method

The main experimental setup shown in Fig. 5 and 6 was used for all experiments. No plants were cultivated since nitrification is considered in isolation (independent of plant growth). The same microbial culture (initially cultivated in Chapter 6) was used. One of the four units failed during run 1 and thus the remaining 3 were used in all remaining experiments. Thus, triplicates are presented instead of quadruplicates as in the previous chapters. The dosing reservoirs, D1 and D2, shown in Fig. 5 and 6 contained 0.2 mM $(\text{NH}_4)_2\text{SO}_4$ (representing digestate) and 0.3 M KOH, respectively. A drain pump (not shown in Fig. 5 and 6) was incorporated to each system to allow for a continuous liquid throughput (CSTR configuration), which was employed in the Section 7.3.3.

Run 1 involved nitrification control in a batch setup with a proposed control strategy (discussed in the following section). The same nutrient solution used in runs 1 and 2 of Chapter 6 was charged, except for $(\text{NH}_4)_2\text{SO}_4$, where 4 mM was charged initially. The pH was controlled autonomously (at 6.5) by dosing hydroxide from the 0.3 mM KOH reservoir via proportional-integral (PI) control (from online pH measurements taken every 30 mins). Run 2 employed a different control strategy in a fed-batch system. The same nutrient solution was supplied with zero $(\text{NH}_4)_2\text{SO}_4$ initially. Ammonium was dosed at a calculated value according to a second proposed control strategy. The pH was also controlled autonomously via PI control. Run 3 was near-identical to run 2, but instead, a constant throughput of deionised water was employed at a dilution rate of 1 day^{-1} (thus, operation in a CSRT instead of a fed-batch).

7.3 Results and discussion

7.3.1 Batch nitrification

Batch systems have the advantage of operating at v_{max} (objective 1) since substrate (ammonium) is consistently available in the bulk solution. Also, complete conversion can be achieved (objective 2). To maintain high production rates, draining and refilling the of the batch system is required soon after complete consumption of ammonium. This is also necessary to maintain the health (maintenance energy) of the microbial community. If the reactor is devoid of ammonium for an extended period, high death rates can be expected. Therefore, control systems able to detect ammonium extinction and drain-and-replace the medium quickly (or dose additional ammonium) are central to batch nitrification units.

Since the hydroxide dosing rates are an inferential measurement of the ammonium oxidation rates, ammonium extinction will be accompanied by a cessation in hydroxide dosing. This provides an online indication of ammonium extinction and enables the controller to take immediate action. This is similar to the approach used in Section 5.3.3, where nitrate extinction was inferred from a reduction in the rate of change of pH. As mentioned in Section 5.3.3, this approach has also been used in wastewater nitrification (Andreottola *et al.*, 2001; Hajsardar *et al.*, 2016; Kim & Hao, 2001). However, in these systems, the pH is not controlled at a set point and oscillates due to alternating nitrification/denitrification processes. Relatively low ammonium concentrations are dealt with (a few mM) and hence changes in pH are sufficiently small as not to affect microbial performance. Digestate, however, contains high ammonium concentrations (often over 100 mM) which would result in critically low pH levels well before complete ammonium conversion. Therefore, the pH must be controlled to achieve efficient nitrification. Thus, the pH remains constant, and the hydroxide dosing rates (resulting from the pH control system) may provide information regarding the ammonium oxidation kinetics instead of changes in the measured pH values.

This control hypothesis was investigated in run 1. Each of the 3 systems was charged with 4 mM ammonium and the pH was controlled at 6.5 via feedback proportional-integral control. More specifically, hydroxide is constantly dosed to the system but the rate at which it is dosed is adjusted (increased/decreased) by the pH controller every 30 min. This adjustment is based on the error signal received by the controller (difference between the pH reading and the pH setpoint). The control algorithm is given in the Sequential-Function-Chart shown in Fig. 25.

The degree of reduction in the hydroxide dosing rate (indication of ammonium extinction) was determined by comparing the recent dosing rate (average over the past 3 hours to reduce noise) to the maximum dosing rate (maximum of all past dosing rates), as described in Fig. 25. Ammonium extinction was assumed from a 50 % or larger reduction in the recent dosing rate compared to the maximum dosing rate, which upon detection, actuated additional ammonium dosing.

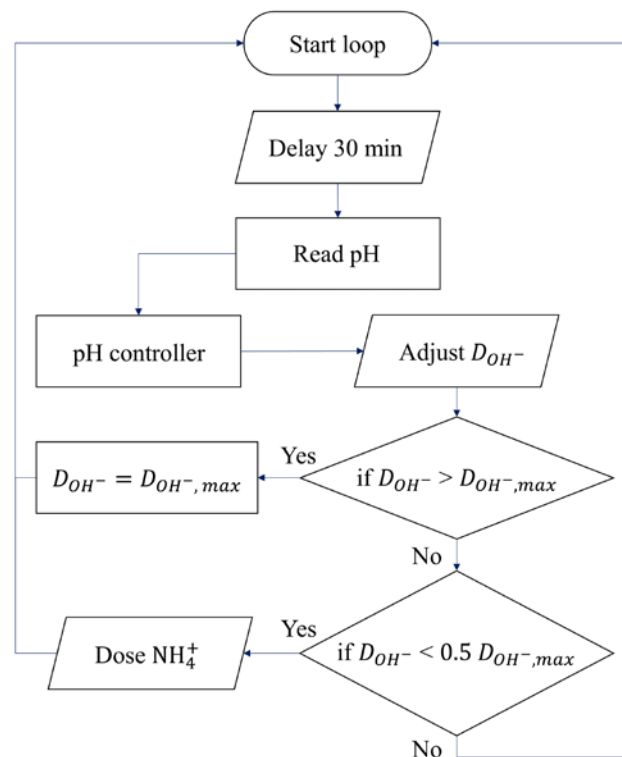


Fig. 25: Sequential-Function-Chart of the batch control algorithm designed to infer ammonium extinction from a 50 % reduction in the hydroxide dosing rate. The pH controller is a feedback proportional-integral controller. $D_{OH^-,max}$ is the maximum D_{OH^-} value since the start of the run. To reduce experimental noise, a six-point running average of the dosing rates were used (average over 3 hours) in the two “if” statements shown in the diamond boxes.

The results from run 1 are given in Fig. 26 for each of the 3 systems. Sharp declines in the hydroxide dosing rates (blue lines) are observed at ammonium extinction, which is confirmed by ammonium concentration measurements (red dots). Vertical green dashed-lines indicate ammonium dosing instances, which were actuated when a 50 % reduction in the ammonium dosing rates occurred. Fast recovery (increase in the hydroxide dosing rates) is observed after

the ammonium dosing instances. Note that the dosing rates are running averages over the past 3 hours (6 points), which was required to prevent false extinction readings resulting from experimental noise. This however caused ammonium dosing to occur in succession since the running average of the dosing rates took longer to increase (slower response time) than the immediate dosing rates. The immediate dosing rates dropped to zero soon after ammonium extinction and returned to their original values equally soon after additional ammonium was dosed.

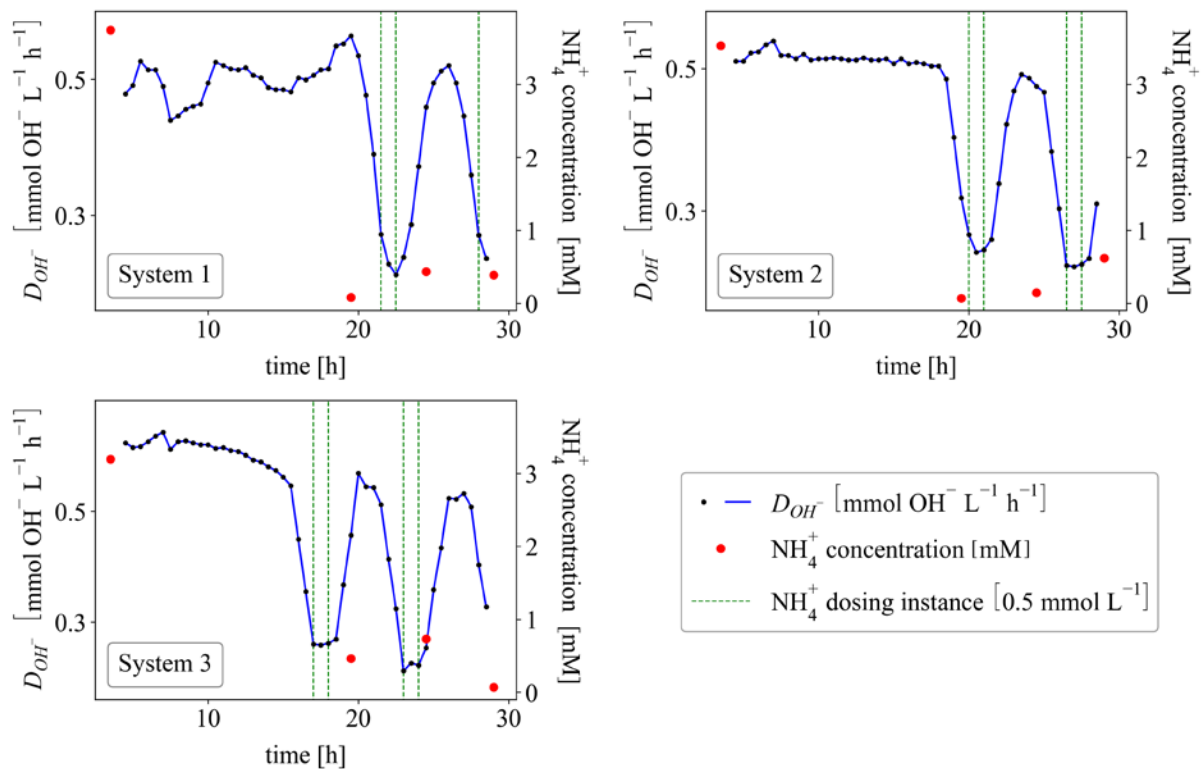


Fig. 26: Results from run 1 in a batch setup. Ammonium extinction was inferred from a 50 % reduction in the hydroxide dosing rates (6 point running average). This was accomplished with the control algorithm presented in Fig. 25. The hydroxide dosing rates are given as blue lines for each of the 3 systems. Ammonium dosing instances are shown as vertical green dashed-lines (which occurred upon a 50 % reduction in the hydroxide dosing rate). Ammonium extinction was confirmed by measurement of the ammonium concentrations in solution (red dots).

Maximum nitrification rates were maintained, and complete conversion was achieved in the batch setup. Controller adaptability (objective 3) is not applicable since ammonium is constantly available in the liquid, and thus variations in microbial activity do not affect controller performance. Although all objectives were satisfied, drawbacks to the batch setup exist. One such drawback is a potential false-alarm of ammonium extinction. For example, if a drop in

temperature occurred such that a 50 % reduction in the nitrification rates resulted, the controller would mistake this as an ammonium extinction event. If large variations in microbial activity are expected, the control strategy can be modified to guard against this by comparing the current dosing rate (which is the average over a 3-hour window) against a longer running average (such as over a 12-hour window), instead of comparing the current dosing rate to the maxing dosing rate over the whole time-span. Inferring ammonium extinction from a reduction in the more-recent dosing rate, compared to all past dosing rates will prevent false ammonium extinction readings if nitrification rates decrease relatively slowly.

7.3.2 Fed-batch systems

Although the batch control system satisfied all the objectives laid out in Section 7.1, other reactor configurations may be desired based on production specifications and drawbacks associated with batch systems, such as substrate inhibition (Kim *et al.*, 2006). Thus, a control algorithm was designed for fed-batch systems next. To meet objective 2 (which is to operate at v_{max}) a control strategy is required which feeds ammonium at v_{max} under conditions in which v_{max} varies (objective 3 of adaptability). To understand the mechanism of such a control strategy, consider Fig. 27 which plots the ammonium dosing rate ($D_{NH_4^+}$) against the corresponding hydroxide dosing rate (D_{OH^-}) required to control the pH. The ‘kink’ in the curve is the point at which the ammonium dosing rate equals v_{max} . Left of this point, ammonium is fed at a slower rate than v_{max} and thus the hydroxide dosing rate is 2 times the ammonium feed rate (Equation 12), since all ammonium is consumed. To the right of this point, ammonium is fed at a faster rate than the bacteria can consume it. Under these conditions, ammonium is still oxidized at v_{max} and the hydroxide dosing rate remains constant at $2v_{max}$. Therefore, if the controller doses an arbitrary amount of ammonium, the resulting hydroxide dosing rate provides information regarding the region of operation (either to the left or right of the targeting operating point). Specifically, if $D_{OH^-}/D_{NH_4^+} < 2$, the ammonium dosing rate is too large and must be reduced (operation is to the right of the target operating point). Alternatively, if $D_{OH^-}/D_{NH_4^+} = 2$, ammonium dosing is less than or equal to v_{max} . In this case, no information exists regarding how much lower the ammonium dosing rate is than v_{max} and thus ammonium dosing should be increased to prevent drifting away from the target operating point. This control strategy is conveyed in Fig. 28, in which the ammonium dosing rates are adjusted incrementally based on the operating regime. Note that a specification of $D_{OH^-}/D_{NH_4^+} < 1.9$ is

employed instead of $D_{OH^-}/D_{NH_4^+} < 2$. This is required to account for experimental errors such as pump calibrations or dosing solution concentrations. If, for example, this specified constant is larger than the actual value (for example, if 2.1 was specified, when the actual value is 2), the controller would continue to decrease the ammonium dosing rates until the ammonium dosing rates equal zero. Therefore, to safeguard against this, the specified constant (1.9 in this case) should be slightly below the calibrated value to allow for errors. Operation will thus occur slightly to the right of the target operating point in Fig. 27. This will result in slow accumulation of ammonium in solution but can be minimized through accurate calibration.

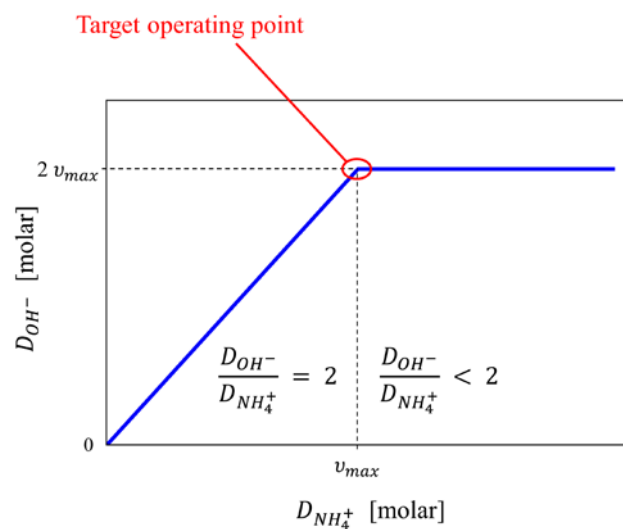


Fig. 27: Plot of the hydroxide dosing rate (D_{OH^-}) required for pH control as a function of the ammonium dosing rate ($D_{NH_4^+}$). At lower ammonium dosing rates, which are below v_{max} , the hydroxide dosing rate is double the ammonium dosing rate (see Equation 12) since all the ammonium dosed is consumed. When the ammonium dosing rate is higher than v_{max} , the hydroxide dosing rate remains constant at $2v_{max}$ regardless of $D_{NH_4^+}$.

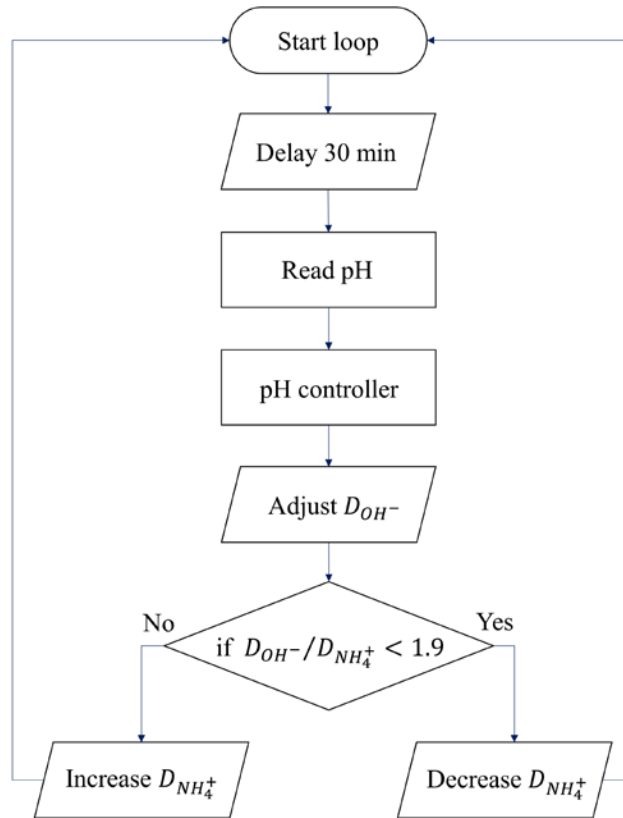


Fig. 28: Sequential function chart of the control algorithm used in the fed-batch system. Designed to operate slightly to the right of “Target operating point” shown in Fig. 27, the ammonium dosing rate is decreased when it is larger than v_{max} and increased when its smaller than v_{max} . A constant of 1.9 is specified (instead of 2) to account for calibration errors as discussed in the text.

Run 2 was conducted to test this control strategy in which the control algorithm shown in Fig. 28 was employed in a fed-batch system with zero ammonium initially. An arbitrary amount of ammonium was dosed by the controller to “kick-start” the algorithm. The results are given in Fig. 29 for each of the three systems. It can be seen that the ammonium dosing rates (red lines) “follow” the hydroxide dosing rates (blue lines). Slow accumulation of ammonium in solution occurred indicating that operation was maintained slightly to the right of the “Target operating point” shown in Fig. 27 (ammonium dosing was slightly higher than v_{max}). The cumulative amounts of ammonium dosed per litre solution (not shown) were around 10 times higher than the ammonium concentrations in solution (thus, around 90 % conversion was achieved). These results show that nitrification can be accomplished at maximum production rates with 90 % conversion under adaptable control. Thus, all objectives were satisfied. However, the

accumulation of ammonium may become significant if the solution is not replaced regularly. To address this problem, the same control strategy was employed in a CSTR setup.

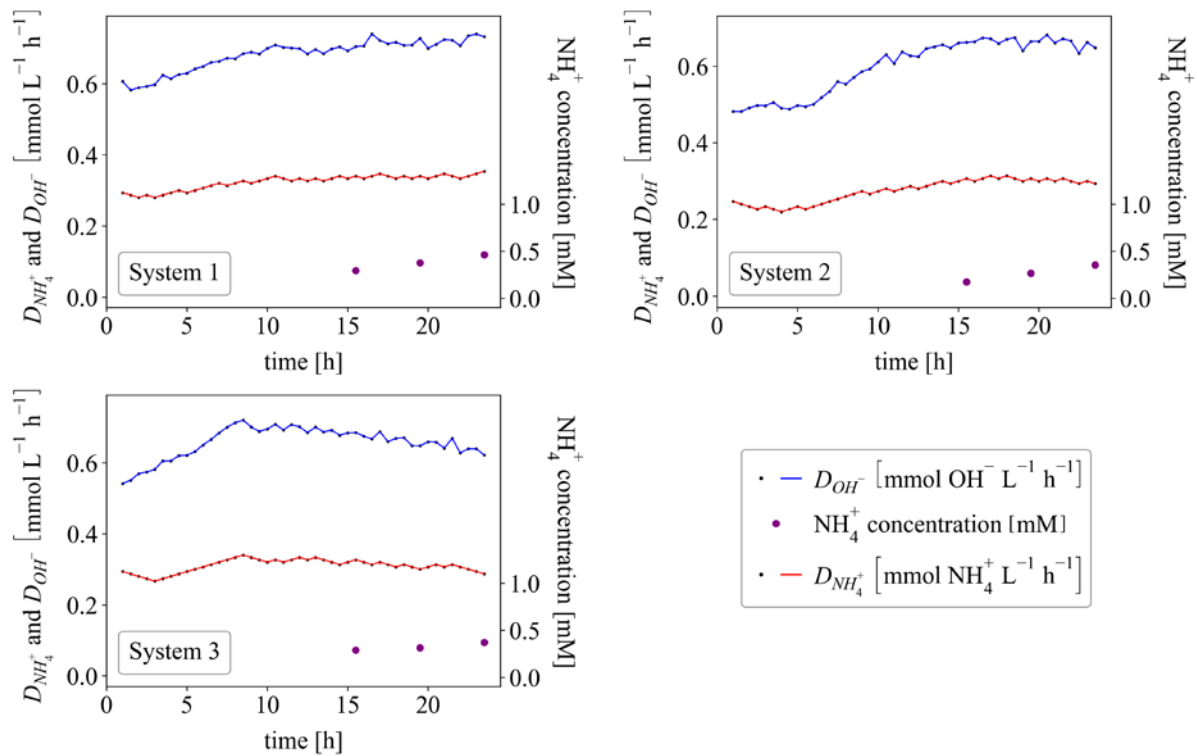


Fig. 29: Results from the control strategy shown in Fig. 28, employed in a fed-batch system (Run 2). Hydroxide dosing rates are shown as blue lines and the ammonium dosing rates are shown as red lines. The ammonium concentrations in solution were measured and shown as purple dots.

7.3.3 Continuous systems

To eliminate ammonium accumulation, a dilution rate of 1 day^{-1} was employed (using deionised water), thus converting the fed-batch reactor into a CSTR. Run 3 was performed to test the control algorithm presented in Fig. 28 in the CSTR. The results are shown in Fig. 30, reported in the same format as in Fig. 29 but including the nitrate concentrations in the reactor/effluent (measured via analysis of liquid samples). Ammonium concentrations stabilized at values below 1 mM and nitrate concentrations at around 7 mM. This corresponded to a conversion of 93 % (± 1 %). The control algorithm performed well to consistently feed ammonium at just above v_{max} , under varying v_{max} conditions (adaptable control).

The ammonium-to-nitrate ratio in solution has great agricultural significance and an optimum ratio typically exists for each plant species (Cytryn *et al.*, 2012). This ratio is often around 1/3 molar, thus requiring a 75 % conversion of ammonium to nitrate (Tabatabaei *et al.*, 2006). As such, if a higher ammonium-to-nitrate ratio is desired, operation should occur further to the right of the “Target operating point” shown in Fig. 27. This can be accomplished by decreasing the constant in the “if statement” of Fig. 28 (currently equal to 1.9).

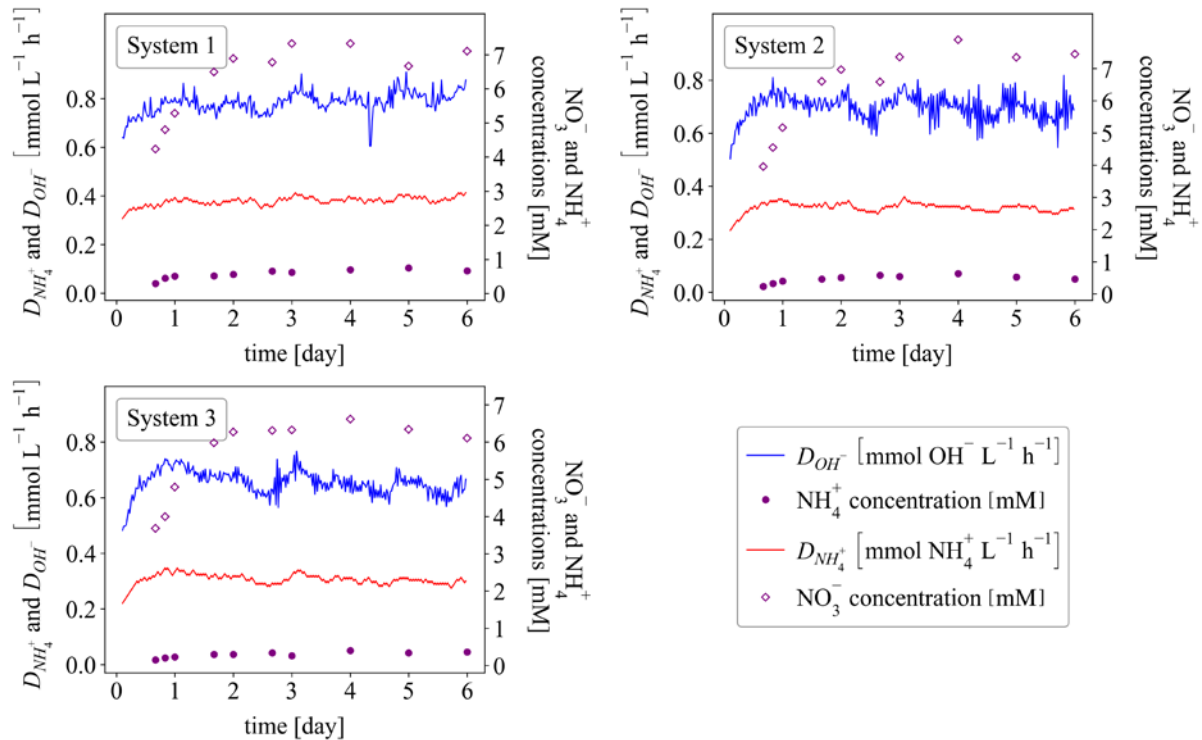


Fig. 30: Results from the control strategy shown in Fig. 28 employed in a CSTR setup with a dilution rate of 1 day^{-1} . Hydroxide dosing rates are shown as blue lines and the ammonium dosing rates are shown as red lines. The ammonium and nitrate concentrations in solution were measured and are shown as purple dots and diamond markers, respectively.

The CSTR setup requires knowledge of the ammonium concentration in the feed. If this concentration is unknown or varies during production, the batch control scheme may be a better alternative. The solution in the batch system can be drained and replaced immediately upon ammonium extinction, thus realising a semi-continuous process.

7.4 Conclusions

The objectives specified in Section 7.1, namely, operation at v_{max} , high conversion and controller adaptability was satisfied using the presented control schemes in batch, fed-batch and CSTR units. Although all the objectives were satisfied in all three reactor configurations, the CSTR system appeared as the most attractive method since seamless production of a high nitrate (93 %) liquid-fertilizer was realised. Drawbacks of each setup was discussed and thus the choice of system will depend on process specifications.

CHAPTER 8 General discussion and conclusions

It was shown that N and P discharge from hydroponic systems can be reduced by around an order of magnitude using the presented control strategies with pH as the sole measured variable. This was accomplished by controlling the N and P concentrations at lower levels and incorporating recycled nutrients (digestate) as feed. Although plant growth rates were maintained, slight decreases in plant N and P content were observed. Stress responses to low nutrient concentrations are typically dependent on plant species and thus the choice of operating concentration should consider the crops being cultivated.

Using EC as the measured variable is the traditional approach to control nutrient concentrations. Although EC has many merits, the drawbacks of using EC (discussed in Section 2.5) may be problematic in some cases (if the feed has a high salinity content, like digestate, for example). pH-based control systems may therefore serve as a much-needed alternative. In general, better nutrient management can likely be achieved by using a combination of EC and pH as input variables to a controller. This work has introduced the use of pH as a new method of nutrient concentration control to the literature. Since pH is routinely measured and controlled in hydroponic systems, pH-based control systems will likely find practical application for better management of fertilizer nutrients.

Although N and P were focused on (given their pollution potential), other nutrient levels could also be controlled using pH. This was accomplished using the same approach as in Chapters 5 and 6, where a constant proton-to-nitrate ratio of around 0.5 mol mol^{-1} was utilized to control the nitrate concentration. Since plant nutrients are generally absorbed in proportion to one another, other nutrients could be added together with nitrate in proportional amounts. For example, in Chapter 5, the nitrate concentration was controlled by controlling the pH with an acid

solution consisting of a proton-to-nitrate ratio of around 0.5 mol mol^{-1} . If it is known that the plant's potassium uptake rate is $1/3$ of its nitrogen uptake rate, then the potassium concentration will also be controlled if the acid dosing solution consists of a potassium to nitrate ratio of $1/3$. As a rough starting point, Hoagland's solution may be composed with additional HCl at a proton to nitrate ratio of 0.5 mol mol^{-1} . Using this solution as the acid dosing solution for pH control will result in total nutrient concentration control (to some degree of accuracy). It was found that for the model plant used (*Brassica oleracea* var. *Sabellica*), an acid dosing solution composed of a proton-to-nutrient ratio of $[0.5 : 1 : 0.3 : 0.2 : 0.1 : 0.075 : 0.05]$ molar parts $[\text{H}^+ : \text{NO}_3 : \text{K} : \text{Ca} : \text{SO}_4 : \text{PO}_4 : \text{Mg}]$ resulted in constant EC over a 10-day run. An advantage of this approach is the chemical stability of the acid dosing solution. The acidic conditions conveniently prevented any nutrients from precipitating (at an acid strength of 0.1 M H^+) and inhibited any microorganism infections such as algae. Disadvantages (as compared to EC) include the absence of an online estimate of the total nutrient concentration. A combination of this pH-based approach and the EC method is likely to yield the best results if total nutrient concentration is opted for.

Emphasis has been placed on ammonium wastewaters (digestate in particular) as feed (chapters 6 and 7) since the use of recycled nutrients is attractive from an environmental perspective. Only nitrogen management was investigated even though digestate consists of all the nutrients required for plant growth. If digestate is supplied as the sole fertilizer, N must be the limiting nutrient else other nutrients (the limiting ones) will deplete in solution. However, if N is the limiting nutrient, the other nutrients will accumulate in solution to some extent. This is particularly concerning for P, since the aim is to minimize N and P. To control N and P simultaneously, P must be the limiting nutrient and N the second-most limiting nutrient. If the N concentration is controlled (using the strategy in Chapter 6, for example), P will deplete in solution over time and additional P must be added, while the remaining nutrients (other than N) will remain in excess. As demonstrated in Chapter 4, P can be inferred from the pH-buffering capacity of the solution. Although the rate of change of pH as caused by the plant was used, the system can be configured differently such that the buffering capacity is measured independent of plant growth or nutrient uptake (since the buffering capacity is inherently independent of the plants). Therefore, N can be controlled using the strategy presented in Chapter 6 (or a combination of the strategies in Chapter 7 and 5) using a digestate in which P is limiting relative to N, and the P concentration can then be maintained by dosing additional P (synthetic) in response the solution's pH-buffering capacity, thereby controlling N and P simultaneously. This

however requires that P dominates the buffering capacity of the solution. It was shown in Chapter 4 that P dominated the buffering capacity of Hoagland's solution (synthetic), but this may not be the case for digestate. As such, the degree to which P dominates the buffering capacity of a digestate which is intended to be used as feed should be determined prior to application.

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