### Remediation of nutrients runoff from feedlot by hydroponic treatment

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Abstract: Nutrient runoff from feedlot can contaminate surface and groundwater and may cause eutrophication if not managed properly. In this study, a cost-effective and environmentally friendly hydroponic treatment was compared with the standard Hoagland solution for treating feedlot runoff and evaluating plant growth and nutrient removal under greenhouse conditions. The hydroponic remediation method was evaluated in batches using 10 L of feedlot runoff and Hoagland solution individually applied in a plastic tub planted with water hyacinth (Eichhornia crassipes), water lettuce (Postia stratiotes), and sorghum (Sorghum bicolor) separately. Water samples were collected weekly and plant biomass were collected at the beginning and the end of the experiment. The sorghum plants grown in the Hoagland solution produced 3.0 and 2.6 times higher biomass than the runoff in first and second batch experiment, respectively, due to balanced nutrient availability, especially nitrogen. Nutrients from the feedlot runoff and Hoagland solution were reduced by all plants as they uptake nutrients, but sorghum outperformed other plants. Plants grown in feedlot runoff, reduced >90% ammonium nitrogen (NH4-N) through the root absorption. Total phosphorus (TP) reduction by sorghum ranged 52%-92% from the Hoagland solution and 70%-100% from the feedlot runoff. Water hyacinth reduced TP by 61%-74% from the feedlot runoff, but only 9%-33% in the Hoagland solution. The TP reduction by water lettuce ranged 49%-93% from the feedlot runoff, but its reduction was not significant in the Hoagland solution. Water lettuce reduced significantly more (75%) Total Kjeldahl Nitrogen (TKN) from the undiluted feedlot runoff than the sorghum (61%) and water hyacinth (66%). Overall, sorghum outperformed water lettuce and water hyacinth by taking up more TP, NH4-N, nitrite and nitrate nitrogen (NO<sub>2</sub>-N+NO<sub>3</sub>-N), ortho-phosphorus (Ortho-OP), and potassium (K) from the undiluted feedlot runoff in both batch experiments.

Keywords: Hoagland solution, water hyacinth, water lettuce, sorghum

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#### 1 Introduction

Livestock production is increasing globally due to the increasing demand of meat-based protein for the increasing global population. Animals raised in a feedlot generate large amounts of manure and runoff, which are rich in macronutrients (such as nitrogen, phosphorus and potassium), organic matter, pathogen, hormones, and antibiotics (Crane et al., 1983; Dillaha et al., 1989). Improper management of feedlot runoff may contribute to surface and groundwater pollution, particularly runoff of nitrogen and phosphorus may cause eutrophication and reduction of oxygen levels in surface water (Ansari et al., 2011; Hribar and Schultz, 2010).

Researchers are developing and testing different treatment options and technologies for safe discharge of feedlot runoff on natural streams to minimize environmental impacts (Connor, 2010; Tiemann, 2011). These include membrane filtration, advanced oxidation distillation, process, air floatation, nitrification, precipitation, ammonia stripping, electro-dialysis, etc. (Bensadok et al., 2011; Ilhan et al., 2008). These methods are complex, expensive, and require specialized technical knowledge for operations and maintenance (Crites et al., 2014) and often they might not be economically viable for livestock growers. Scientists

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and researchers are continuously seeking for alternative methods of treating feedlot runoff. Among different biological treatments, the hydroponic plant cultivation technique is one of the options that is used for treating industrial and municipal wastewater, but limited use in treating nutrient runoff from feedlots.

The hydroponics require minimum energy, is inexpensive to implement, and it is environment friendly. Additionally, seedlings of hydroponic plants are likely to uptake soluble nutrients from runoff. When compared with other management options like vegetative filter strips (VFS) or other wastewater treatment, hydroponic treatments has a better soluble nutrient reduction capacity (Jamuna and Noorjahan, 2009). Thus, to reduce the soluble nutrients from wastewater, researchers use different hydroponic crops based on the adoption capacity of plants in wastewater, its biomass production, and nutrient reduction capacity (Gupta et al., 2012). Published literature suggested that water hyacinth (Brix and Schierup, 1989; Gupta et al., 2012; Ndimele and Ndimele, 2013; Spencer et al., 2006), water lettuce (Gupta et al., 2012; Kon é et al., 2002; Snow and Ghaly, 2008), and sorghum (Khan et al., 2010; Lobato et al., 2008; Oliveira Neto et al., 2009; Yang, Y. et al., 1990) are salt tolerant plants and they can be used to reduce soluble nutrients from wastewater. Additionally, plant biomass produced in wastewater can be used as an animal feed or other purposes such as making paper, fiberboard, ropes, baskets, charcoal briquetting, fertilizer, and fish feed (Gopal, 1987).

Generally, salinity of feedlot runoff varies according to rainfall amount, animal density, topographic conditions, and feedlot management. According to Rahman et al. (2013), the salinity level of feedlot runoff in North Dakota ranged from  $0.701 \pm 0.501$  to  $4.740 \pm 2.873$  mS/cm. Sweeten (1990) reported that the salinity level of feedlot runoff generated from a feed yard in Texas ranged from 6 to 8 mS/cm. Typically, feedlot runoff has high salinity, thus, hydroponic plants used for treating feedlot runoff must be salt tolerant. However, application of hydroponics technique is limited to treating feedlot runoff and it is important to evaluate hydroponic treatment effectiveness in minimizing nutrients from feedlot runoff. Therefore, the objectives of this research were: i) to evaluate and compare nutrient removal from feedlot runoff and standard Hoagland solutions by different crops (water hyacinth, water lettuce, and sorghum) grown under greenhouse conditions, and ii) to compare plant biomass using feedlot runoff and standard Hoagland solutions

#### 2 Materials and methods

# 2.1 Runoff sample collection and standard Hoagland solution preparation

This study was conducted in two batches comparing plant growth and nutrient removal in either feedlot runoff or standard Hoagland solution. The first batch of experiments with runoff was conducted using feedlot runoff effluent collected from a runoff retention pond. On the second batch, feedlot runoff was collected at a predetermined interval, immediately after the feedlot pen surface drainage by ISCO sampler (Teledyne ISCO Inc., Lincoln, NE, USA). In both cases, sufficient feedlot runoff samples were collected to set up experiments. All samples were stored at 4 °C until used and mixed thoroughly before using.

For the standard Hoagland solution experiments, Hoagland solution was prepared by mixing a predetermined quantity of chemicals with reverse osmosis (RO) water by following modified Hoagland nutrient solution preparation procedure (Hanan and Holley, 1974; Hoagland and Arnon, 1950). Hoagland solution was used as reference for ideal plant growing medium, in order to compare plant growth and nutrient removal capacity of different plants cultivated in Hoagland and feedlot solution under the same growing conditions.

#### 2.2 Greenhouse environmental condition

The first batch of experiments was conducted during June 13 to July 4, 2013. Day length, solar intensity, and ambient temperature information were downloaded from the North Dakota Agricultural Weather Network (NDAWN), which was within 3 km from the experimental site. solar hours ranged between 15-16 hours. The average daily ambient temperature ranged from 25°C to 29°C and outside solar radiation ranged from 1,169 to 1,243  $\mu$ mol s<sup>-1</sup> m<sup>-2</sup> during the first experimental period. Corresponding indoor temperature and solar radiation ranged from 25°C to 30°C and 525 to 610  $\mu$ mol s<sup>-1</sup>m<sup>-2</sup>, respectively. Similarly, the second batch of experiments was conducted between September 9 to October 14, 2013, when solar hours were shorter (11 to 13 h), and the average daily ambient temperature and solar radiation ranged from 6°C to 18°C and 346 to 600  $\mu$ mol s<sup>-1</sup> m<sup>-2</sup>, respectively. At the same time, greenhouse indoor temperature varied from 20°C to 25°C and solar radiation varied from 225 to 410  $\mu$ mol s<sup>-1</sup> m<sup>-2</sup>.

#### 2.3 Treatments

In the first batch, four treatments (as-is runoff, 1:1 and 1:2 runoff diluted with RO water, and a standard Hoagland solution) and three crops (water hyacinth, water lettuce, and sorghum) cultivated in hydroponics were arranged in a completely randomized factorial (4  $\times$  3) experimental design with three replications, totaling 36 experimental units.

A second batch of experiments was conducted to compare only as-is (undiluted) feedlot runoff with the Hoagland solution. Thus, in the second batch only 18 experimental units (2 treatments  $\times$  3 plant species  $\times$  3 replications) were used. Additionally, three buckets of RO water without plants were set up to measure the evaporation rate under the same experimental conditions.

#### 2.4 Experimental setup

Three types of crops (water hyacinth, water lettuce, and sorghum) were seeded in Hoagland, diluted, and as-is feedlot runoff solution. Four plants of water hyacinth and water lettuce with equal sizes were taken from a seedling preparation plastic tub. To measure plant fresh biomass, plant's water was drained for 10 min, soaked with paper towel to remove extra water, grouped together and weighted. Then, they were seeded, and grown in a rectangular plastic tub ( $400 \times 318 \times 152$  mm) (12-Quart black dishpan model 0657; Sterlite Corporation, Townsend, MA, USA) with 10 L of well-mixed feedlot runoff or Hoagland solution.

For sorghum plants, sorghum seeds were germinated on Rockwool cubes inside a plastic tray and seedlings were transferred and anchored in foam boards float in the plastic tub. In the first batch of experiment, row, and column spacing for sorghum plants were equal  $(45 \times 45)$ mm), but in the second experiment, row and column spacing were  $45 \times 90$  mm to provide additional space for plant growth. In this way, total 56 and 32 seedlings were placed in the first and second batch, respectively. In both batches, while the plants were in place, dissolved oxygen was supplied gently (at the rate 0.2 L per minute) at the bottom of the plastic tray from a centralized air compressor (QGV-75, Quincy Compressor, Quincy, IL, USA) using a vinyl tubing (3 mm internal diameter) (Cole-Parmer, Fargo, ND, USA) attached with an air stone (AA8; Pentair Aquatic Eco-system, Apopka, FL, USA). During an experimental period, evaporation rate, water temperature, and photosynthetically active radiation (PAR) were measured from buckets with RO water (Culligan AC-30 RO system; Culligan, Fargo, ND, USA), infrared thermometer (IRT 207; General Tools and Instruments, Secaucus, NJ, USA), and quantum sensor LI-COR, Lincoln, (LI-250A; Nebraska USA), respectively.

#### 2.5 Water management and sampling

Weekly water loss due to evaporation from each bucket was determined by measuring the amount of RO water added to bring the water level back to its original volume and measurement was delayed for one hour to allow homogeneous mixing and conditioning of water in the bucket. Before taking the weekly water samples, in-situ electrical conductivity (EC) and pH were measured using a handheld EC and pH meter (YSI Pro Plus; YSI Inc., Ohio, USA). Collected water samples were stored at 4 °C for later nutrient analysis.

#### 2.6 Sample Analysis

Twenty (20) mL unfiltered water samples were poured into crucibles and oven dried at  $105^{\circ}$ C for 24 h and Total solids (TS) was measured using the 2540B method (APHA, 2005). Nutrients such as ortho-phosphorus (ortho-P), total phosphorus (TP), ammonium-nitrogen (NH<sub>4</sub>-N), nitrate-nitrite (NO<sub>3</sub>-N+NO<sub>2</sub>-N), Total Kjeldahl nitrogen (TKN), and potassium (K) were measured using Lachet QuickChem (Lachat Instruments, Loveland, CO, USA) following the procedure summarized in Table 2. Before analyzing Ortho-P, NH<sub>4</sub>-N, and NO<sub>3</sub>-N, water samples were filtered using 0.45  $\mu$  filters (mixed cellulose ester membrane filters). For quality assurance and quality control (QAQC) in the QuickChem analysis method, calibration standards and blanks were analyzed in every ten samples.

 Table 2 Method/protocol used to analyze Hoagland solution and feedlot runoff samples from hydroponic experiments (Rahman et al., 2013)

Parameter, mg/L	Methods /protocol used/ Measurement range
Ortho-P <sup>a</sup>	QuickChem Method 10-115-01-1-O (Lachat Instruments, Loveland, CO) Equivalent to EPA 365.1 method; 0-20
NH <sub>3</sub> -N <sup>a</sup>	QuickChem Method 10-107-06-1-J (Lachat Instruments, Loveland, CO) Equivalent to EPA 353.2 method; 0-20
$NO_2 + NO_3 - N^a$	QuickChem Method 10-107-04-1-R (Lachat Instruments, Loveland, CO) Equivalent to EPA 350.1 method; 0-20
K <sup>b</sup>	Hach Method 8049 (Tetraphenylborate); 0-7
$TP^b$	Hach Method 10127 (Molybdovanadate Method with Acid Persulfate Digested); 1-100
TKN	APHA 2005 4500-N C (Semi Micro Kjeldahl Method)
TN <sup>b</sup>	Hach Method 10072 (Acid Persulfate Digestion); 2-150

Note: <sup>a</sup>Equivalent EPA methods. <sup>b</sup>USEPA approved for reporting.

Additionally, mineral composition such as calcium (Ca), magnesium (Mg), sulfur (S), zinc (Zn), manganese (Mn), copper (Cu), molybdenum (Mo), boron (B) and iron (Fe) in feedlot runoff and plant tissue (before and after experiment) were analyzed. All minerals were measured by the inductively coupled plasma spectroscopy (ICP) using 2010-11-15 Standard Method in the Wet Ecosystem Laboratory at the North Dakota State University.

Plant growth was visually inspected and plants' net biomass were calculated and processed as described by Itoh and Barber (1983), where plants were removed and roots were washed carefully with running water. Following raw plant collection, dry weight of plants was determined using the APHA (2005) oven drying method (Method 2540B) by drying samples at 105°C for 24 h or until a constant weight was reached. Later on, net plant biomass and nutrient removal were calculated.

#### 2.7 Data analysis

Analysis of variance (ANOVA) was used to compare mean concentrations of pH, EC, TP, OP, NH<sub>4</sub>-N, NO<sub>3</sub>-N+NO<sub>2</sub>-N, TKN, and K among treatments and plants. All statistical analysis was done using SAS software version 9.3 (SAS Institute Inc., Cary, NC, USA) using the PROC means procedure at the 5% level of significance. The null hypothesis tested was that mean pollutant concentrations and nutrient removal efficiencies among treatments and plants were equal.

#### **3** Results and discussion

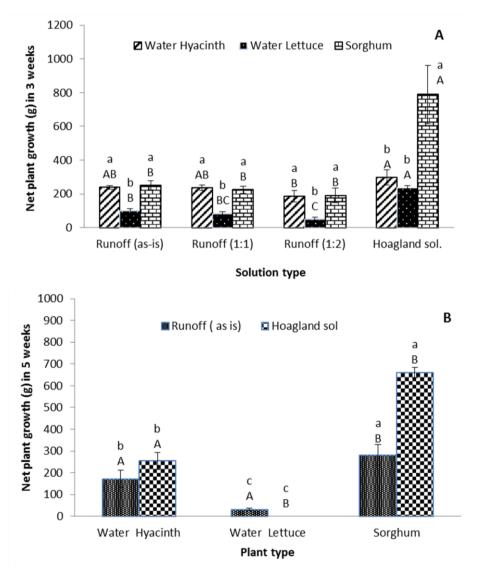
# **3.1 Feedlot runoff and Hoagland solution characteristics**

The characteristics of initial feedlot runoff are presented in Tables 3 and 4. In general, the concentration of nutrients in feedlot runoff was higher in the second batch than those with the first batch except for NO<sub>3</sub>-N+NO<sub>2</sub>-N (approximately five times lower than the first batch). Electrical conductivity (EC) and concentrations of TS, TP, OP, TKN, NH<sub>4</sub>-N, and K in the second batch feedlot runoff experiment were approximately 5, 4.5, 6, 2, 6, and 14 times higher than those measured in the first batch, respectively. This was likely due to differences of sampling locations and timing (feedlot runoff from retaining pond vs. runoff from immediately after the pen surface drainage). The pH was similar between two batches. The Hoagland solution was prepared based on the recommended doses of chemicals required for plant growth (Hoagland and Arnon, 1938, 1950). Thus, nutrient concentrations between the two batches of Hoagland solution were

almost similar. Between feedlot runoff and Hoagland solution, the Hoagland solution had higher TP, TKN, NH<sub>4</sub>-N, NO<sub>3</sub>-N+NO<sub>2</sub>-N, and K concentration than the feedlot runoff.

#### 3.2 Net plant biomass

Water hyacinth and sorghum plants grew 200% and 300% more compared to water lettuce, in both batches of experiments with feedlot runoff and Hoagland solution (Figure 2). Sorghum produced the most biomass and water lettuce produced the least biomass, while water hyacinth growth was in between.



(A) in the first batch experiments during 3 weeks, (B) second batch experiments during 5 weeks Figure 2 Net plant biomass of water hyacinth, water lettuce, and sorghum. The bars with the same capital letter and the same plant type are not significantly different over the experiment period. Similarly, the same small letter for the same feedlot runoff type and different plants are not significantly different from each other at p<0.05.

Overall, average net plant biomass of water hyacinth, water lettuce, and sorghum in the Hoagland solution was 134%, 304%, and 344% more than the feedlot runoff (Figure 2). The lower biomass in the feedlot runoff were likely due to lower NH<sub>4</sub>-N and NO<sub>2</sub>-N+NO<sub>3</sub>-N concentration in the feedlot runoff than the Hoagland solution. Greater biomass is likely to contribute to greater pollutant removal from feedlot runoff and Hoagland solution. The net biomass of water hyacinth, water lettuce, and sorghum was about 11%, 17%, and 11% less in the 1:1 diluted feedlot runoff with RO water and 22%, 50%, and 24% less in the 1:2 diluted feedlot runoff with RO water, respectively, than the plants grown in undiluted (as-is) feedlot runoff (Figure 2A) in the first batch experiment. The lower growth rate of plants in diluted feedlot runoff was due to the dilution of nutrients present in feedlot runoff.

The net water hyacinth, water lettuce, and sorghum biomass produced in the first batch of feedlot runoff was 28%, 68%, and 99% higher than the second batch experiment, respectively, and in the Hoagland solution was 14%, 12.3%, and 16% higher than that of the second batch experiment, respectively (Figures 2A and 2B), although ammonium nitrogen was 87% (Figure 3), potassium was 93% (Figure 5), ortho-phosphorus was 40% (Figure 6) and total phosphorus was 83% less (Figure 7) in the first batch of feedlot runoff than the second batch of feedlot runoff. The plant biomass difference was likely due to differences in the longest solar day (June-July) and intensity of photosynthetically active solar radiation as pointed out in the greenhouse environmental section. Therefore, this study demonstrated that undiluted (as-is) feedlot runoff may be used for growing hydroponic plants to minimize nutrient runoff via plant uptake. Therefore, there are potential to grow hydroponic plants in runoff water; however, plant biomass is likely to be affected by the environmental conditions.

#### 3.3 pH change

Feedlot runoff samples were slightly alkaline (pH =7.45) and the Hoagland solution was slightly acidic (pH =5.76) throughout the study period (Table 3). No noticeable changes of pH were observed for any plant grown in feedlot runoff, however, the pH of the Hoagland solution seeded with sorghum resulted in either increase in pH or remained the same. A similar trend was also observed by Tarre and Green (2004) for the Hoagland solution. In the Hoagland solution, NO<sub>3</sub><sup>-</sup> was about four times higher than the NH<sup>+</sup> and the sorghum plants' remarkable uptake of  $NO_3^-$ .  $NH_4^+$  or  $NO_3^-$  is assumed to release one  $H^+$  or one OH, respectively. Therefore, plants uptake of  $NO_3^{-1}$  from the Hoagland solution by sorghum is likely to increase the pH value of the Hoagland solution (Dejaegere et al., 1984; Jeong and Lee, 1996). Similarly, slight fluctuation of pH value in feedlot runoff was likely due to the nitrification process of bacteria.

 Table 3 Characteristics of feedlot runoff and Hoagland solution used to grow plants hydroponically in the first and second batch of experiments

	First batch		Second Batch				
Parameters	Feedlot runoff			<ul> <li>Hoagland Sol.</li> </ul>	Feedlot runoff		
	As-is	1:1	1:2		As-is	Hoagland Soln.	
рН	7.45±0.03 c <sup>*</sup>	7.51±0.02 b	7.63±0.08 a	5.76±0.01 d	7.97±0.03 a <sup>*</sup>	6.21±0.04 b	
EC, mS/cm	0.80±0.004 b	0.48±0.003 c	0.368±0.005 d	1.55±0.03 a	3.99±0.03 a	1.47±0.03 b	
TS, mg/L	0.75±0.026 b	0.42±0.009 c	0.32±0.015 d	1.07±0.034 a	3.41±0.183 a	0.97±0.577 b	
TP, mg/L	16.53±3.69 b	6.07±1.78 b	4.23±0.83 b	175.92±23.88 a	95.70±11.37 b	168.17±2.70 a	
OP , mg/L	8.23±1.20 b	4.47 ±0.359 c	2.51±1.051 c	66.47±5.83 a	13.69±1.48 a	64.83±4.15 b	
TKN, mg/L	53.5±2.04 b	45.93±7.63 c	40.27 ±0.23 c	64.93±8.57 a	217.20±22.17 a	99.87±9.39 b	
NO <sub>3</sub> -N+NO <sub>2</sub> -N, mg/L	$1.43\pm0.49$ b	1.01±0.84 b	0.59±0.01 b	117.64±6.29 a	$0.28{\pm}0.01~b$	123.50±1.323 a	
NH <sub>4</sub> -N, mg/L	$4.26{\pm}0.376~b$	1.91±0.237 c	1.26±0.026 c	29.97±1.258 a	32.20±0.78 a	34.05 ±7.50 a	
K, mg/L	57.23±11.66 b	47.13±11.66 b	30.60±5.10 c	144.77±15.43 a	777.70±26.72 a	134.67±5.83 b	

Note: \* Values are mean ±SD. Same letter within a row and same parameter are not significantly different at P≤0.05

#### 3.4 Electrical conductivity (EC)

Average EC values in the first batch of experiment for undiluted feedlot runoff, 1:1 and 1:2 diluted runoff, and Hoagland solution are presented in. Whereas, the EC value of undiluted runoff and a Hoagland solution in the second batch of experiments are presented in Table 3. This difference in EC values between batches was due to sampling locations and sampling timing. The EC values of feedlot runoff and the Hoagland solution seeded with plants decreased gradually over the experimental period, which was likely due to plant uptake of salt and nutrient ions (Table 3).

The salt tolerance threshold levels for the sorghum is 6.8 mS/cm (Tabatabaei and Anagholi, 2012), water hyacinth is 2.85 mS/cm (Rotella, 2010), and water lettuce is 2.9 mS/cm (Haller et al., 1974). The corresponding lethal EC limits for sorghum, water hyacinth, and water lettuce are 12.0, 7.8, and 4.0 mS/cm, respectively (Rotella, 2010; Rani et al., 2012; Gupta et al., 2012). In this study, the salinity of the Hoagland solution were within the threshold EC level, but the EC value in feedlot runoff in the second batch was 26% and 27% higher than the threshold EC values for water hyacinth and water lettuce, respectively (Table 3). In both batches of experiments, the EC values were lower than the lethal limit for any of the plants, and these plants can be seeded to treat feedlot runoff.

#### 3.5 Ammonium nitrogen (NH<sub>4</sub>-N) reduction

The average  $NH_4$ -N concentration in the first and second batch of feedlot runoff was 4.26 and 32.2 mg/L,

respectively. Similarly, the NH<sub>4</sub>-N concentration in Hoagland solution was 29.97 and 34.05 mg/L in the first and second batch, respectively (Figure 3). Irrespective of feedlot runoff or Hoagland solution, NH<sub>4</sub>-N concentration was reduced significantly by all plants towards the end of the experimental period and most of the NH<sub>4</sub>-N reduction occurred within the first week of experiment initiated (Figures 3A and 3B) when maximum plant growth was noticed. However, the differences in NH<sub>4</sub>-N concentration reduction among plants were not significant. Young plants prefer to utilize NH<sub>4</sub>-N than nitrate (NO<sub>3</sub>-N), especially water hyacinth absorbs ammonia by their roots to incorporate it in their biomass (Gupta et al., 2012). As a result, the significant NH<sub>4</sub>-N reduction was likely to occur within the first and second week of the experimental period. Due to dilution of the feedlot runoff (1:1 and 1:2), there were no significant differences in NH<sub>4</sub>-N reduction and plants' growth. Irrespective of treatments, once again sorghum uptake was significantly higher NH<sub>4</sub>-N than water lettuce, but not significantly different from water hyacinth.

Thus, sorghum reduced most (92%) of the NH<sub>4</sub>-N concentration. This was expected since sorghum produced the most biomass and water lettuce produced the least amount of biomass, while water hyacinth growth was in between (Figures 2A and 2B). Therefore, any of the plants used in this study may be seeded to reduce NH<sub>4</sub>-N from wastewater, but sorghum outperformed others.

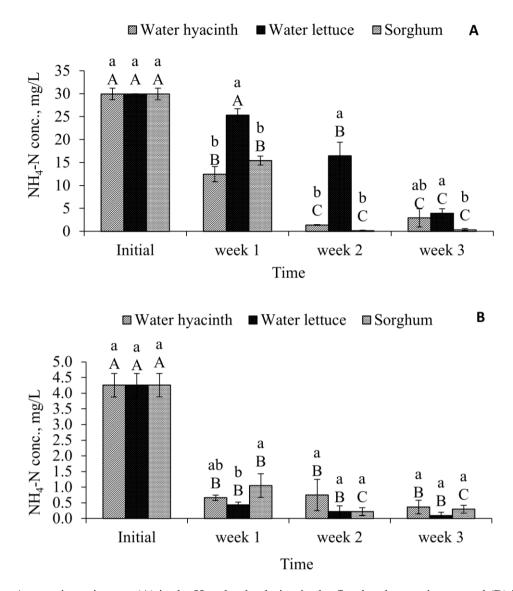


Figure 3 Ammonium nitrogen (A) in the Hoagland solution in the first batch experiment, and (B) in runoff (undiluted) in the first batch experiment. The bars with the same capital letter and the same plant type are not significantly different at each sampling week over the experimental period. Similarly, the same small letter for the same sampling date and different plants are not significantly different from each other at p≤0.05. The same Y-axis scale was not used due to concentration differences.

#### 3.6 Nitrate and nitrite nitrogen

Initial NO<sub>2</sub>-N+NO<sub>3</sub>-N concentration in feedlot runoff was 1.43 and 0.03 mg/L in the first and second batch, whereas this concentration in the Hoagland solution was 117.64 and 123.5 mg/L in the first and second batch, respectively. Since, NO<sub>2</sub>-N+NO<sub>3</sub>-N concentration in the second batch was not significant (data not shown). The NO<sub>2</sub>-N+NO<sub>3</sub>-N concentration in the feedlot runoff was almost negligible as compared to Hoagland solution (Figure 4). Therefore, the Hoagland solution is likely to provide most of the nitrogen required by plants than the feedlot runoff throughout the experimental periods. For better plant growth, a sufficient concentration of  $NO_2-N+NO_3-N$  is necessary with  $NH_4-N$ . If the Hoagland solution is considered as the ideal plant growing medium, then lower net plant biomass (Figures 2A and 2B) production in the feedlot runoff as compared to Hoagland was due to the lack of required amount of  $NO_2-N+NO_3-N$  concentration in runoff along with lower solar intensity and duration during the second batch of this study.

The NO<sub>2</sub>-N+NO<sub>3</sub>-N concentration in the feedlot runoff fluctuated during the study period (Figure 4). These fluctuations of NO<sub>2</sub>-N+NO<sub>3</sub>-N concentrations were likely due to nutrient uptake by plants and nitrification caused by microbes in feedlot runoff as also indicated by other researchers (Sooknah and Wilkie, 2004). Typically feedlot runoff is slightly alkaline (pH= 7.5-8.5). The decrease in NH<sub>4</sub>-N concentration and an increase in NO<sub>2</sub>-N+NO<sub>3</sub>-N concentration in feedlot runoff supported the nitrification process of the feedlot runoff both in undiluted (Figures 3 and 4) and diluted feedlot runoff samples (1:1 and 1: 2) used in this study.

 $NO_2$ -N+NO<sub>3</sub>-N concentration fluctuated in feedlot runoff seeded buckets and significant differences in  $NO_2$ -N+NO<sub>3</sub>-N concentration reduction was observed towards the end (Figure 4, p<0.05). The fluctuation of  $NO_2$ -N+NO<sub>3</sub>-N concentration was likely due to nitrification and denitrification process, nutrient uptake by plants, and biomass production (Kruzic et al., 1990). The highest  $NO_2$ -N+NO<sub>3</sub>-N concentration at week one and two was likely due to measurement or sampling error. Therefore, plants used in this study are not the best option to reduce  $NO_2$ -N+NO<sub>3</sub>-N concentration.

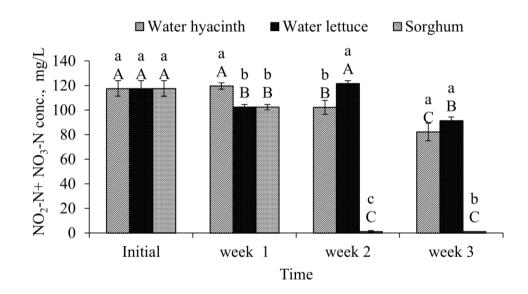


Figure 4 Nitrate nitrogen concentration in the Hoagland solution in the first batch experiment.
The bars with the same capital letter and the same plant type are not significantly different at each sampling week over the experimental period. Similarly, the same small letter for the same sampling date and different plants are not significantly different from each other at p≤0.05. The same Y-axis scale was not used due to concentration differences. plants are not significantly different from each other at p≤0.05. The same Y-axis scale was not used due to concentration differences.

#### 3.7 Potassium (K)

The typical K requirement for the plants ranged from 10 to 83 mg/L depending on the plant species (Gupta et al., 2012). The K concentration in the feedlot runoff was reduced by 2.6 and 3 times by water hyacinth and sorghum, respectively, as compared to the water lettuce during the first batch (Figure 5A). In both batches, sorghum (78%) showed significantly higher K uptake than the water hyacinth (59%) and water lettuce (34%) (Figure 5B) (first batch shown only). Gamage and Yapa (2001) also found a similar K reduction (64.4%) using hyacinth in textile effluent. The measured K concentration in the second batch feedlot runoff was 778 mg/L (data not shown), which was about 13 and 5 times higher than that measured in the first batch from the feedlot runoff (undiluted) and the Hoagland solution, respectively, due to sampling location. The K uptake pattern of all the three plants for undiluted and diluted feedlot runoff was virtually similar in the first batch. In the Hoagland solution, the K uptake was 77% higher for sorghum than water lettuce, but similar for water hyacinth at the end of the first batch (Figure 5A, p=0.05). When plants were seeded in feedlot runoff, the sorghum resulted in the highest K uptake, followed by water hyacinth (Figure 5B). Water lettuce, however, did not show any significant K uptake from feedlot runoff in the second batch experiment (Figure 5b). The sorghum reduced 161% and 220% higher K towards the end than the other two plants in the Hoagland solution in first and second batch experiments, respectively.

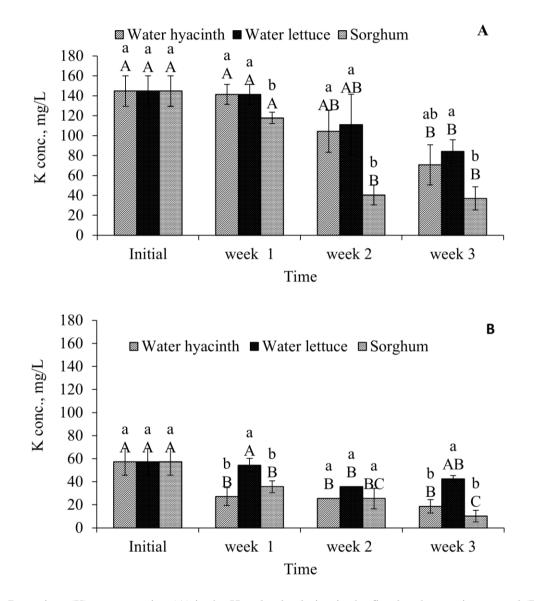


Figure 5 Potassium (K) concentration (A) in the Hoagland solution in the first batch experiment, and (B) in the runoff (undiluted) in the first batch experiment. The bars with the same capital letter and the same plant type are not significantly different at each sampling week over the experimental period. Similarly, the same small letter for the same sampling date and different plants are not significantly different from each other at p≤0.05.

#### 3.8 Ortho-phosphorus (OP)

Among different types of phosphate, OP is readily available and supplies the phosphate requirements for plants. From the feedlot runoff (diluted and undiluted), OP concentrations were considerably depleted by all the three plants within two weeks in the first batch of experiments (Figures 6A and 6B). In the second batch of experiment with the feedlot runoff, OP concentration decrease in water hyacinth, water lettuce, and sorghum were 69%, 59%, and 20% in the week 1, respectively, and 75%, 71%, and 60% in the week 2, as compared to initial concentration. The OP concentration increased 248% from week 2 to the end of the experiment, especially in water hyacinth and water lettuce seeded feedlot runoff (Figure 6B). However, as compared to the beginning of the experiment, at the end of the experiment, OP concentration values were reduced 57% by water lettuce and 68% by sorghum. The main reason for OP fluctuations in feedlot runoff was likely due to the release of OP from the TP. The TP was about six times greater in the second batch of feedlot runoff than the first batch of feedlot runoff. Additionally, the net biomass and the OP concentration in the solutions (in the feedlot runoff and Hoagland) indicated that higher OP could contribute to greater plant growth rate (Figures 2A and 2B). At the end of the first and second batch experiments, the OP concentration uptake by sorghum was significantly higher and followed by water hyacinth (Figure 6A). For the feedlot runoff, water lettuce and sorghum reduced significantly more OP than water hyacinth.

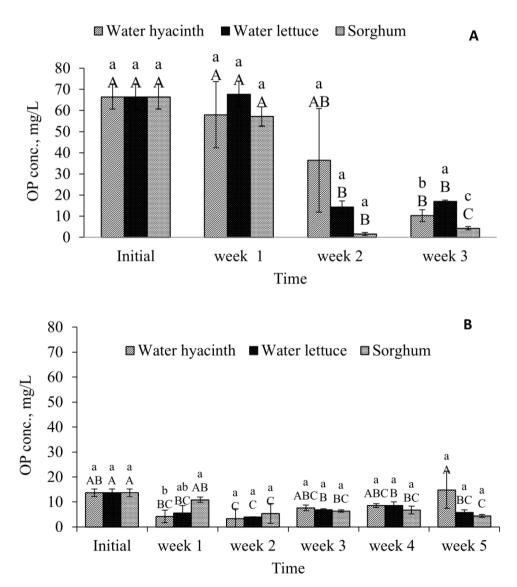


Figure 6 Orthophosphate (OP) concentration (A) in the Hoagland solution in the first batch experiment and (B) in the runoff (undiluted) in the second batch experiment. The bars with the same capital letter and the same plant type are not significantly different at each sampling week over the experimental period. Similarly, the same small letter for the same sampling date and different plants are not significantly different from each other at  $p \le 0.05$ .

#### 3.9 Total Phosphorus (TP)

Total phosphorus in feedlot runoff generally consisted of organically bounded P and OP. The organically bounded P is converted to the OP by bacteria, fungus, and other chemical reactions (Arcand and Schneider, 2006). Therefore, TP concentration plays a vital role in plant growth, though it is not readily available to plants as OP. In the first batch, TP concentration in the feedlot runoff was about ten times lower than that of the Hoagland solution (Figures 7A and 7B). However, TP concentrations in the second batch of feedlot runoff were nearly half (95.70 mg/L) of the Hoagland solution (174.83 mg/L).

Water hyacinth and sorghum seeded in the feedlot runoff up took about 74% and 100% of TP in first batch experiment (Figure 7B). In the Hoagland solution, however, the average TP reduction was 33% and 92% at the end of water hyacinth and sorghum, respectively (Figure 7A). Higher TP reduction by plants in the runoff as compared to Hoagland solution in the first batch experiment was likely due to lower initial TP concentration in feedlot runoff. Similarly, TP reduced by water hyacinth and sorghum was 61% and 70% from the feedlot runoff and 9% and 52% from the Hoagland solution, respectively, in the second batch of experiment (data not shown). The lower percentage reduction of TP in the second batch of experiment as compared to the first batch of the experiment was due to the plant spacing (especially sorghum was planted 90 mm in the first batch and 45 mm spacing in the second batch). Thus, higher initial TP concentration in the feedlot runoff and low plant densities showed relatively low TP uptakes (18% and 30% lower by the water hyacinth and sorghum, respectively) by the plants in the second batch experiment (Figure 7 and Table 3). Similarly, in the first batch of experiment, diluted feedlot runoff, the final TP concentration was almost zero and TP uptake by plants was not significantly different. In the first and second batches of Hoagland solution, sorghum reduced the highest TP concentration (92% and 52%, respectively), whereas water lettuce reduced the least amount of TP concentration (13% and <1% of TP, respectively).

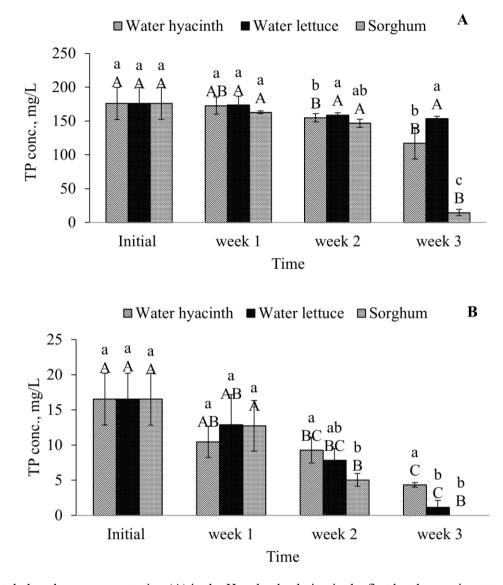


Figure 7 Total phosphorus concentration (A) in the Hoagland solution in the first batch experiment and (B) in the runoff (undiluted) in the first batch experiment. The bars with the same capital letter and the same plant type are not significantly different at each sampling week over the experimental period. Similarly, the same small letter for the same sampling date and different plants are not significantly different from each other at p≤0.05. Same scale in Y-axis was not used due to large concentration differences.

## 3.10 Overall nutrient percentage reduction by sorghum

The greatest nutrient reduction was observed by the sorghum plants, followed by water hyacinth and water lettuce (Figures 8 and 9). Nutrient reduction by other plants were similar or lower than the sorghum and they are not presented. TP reduction by sorghum in the first and second batch of the experiment from undiluted feedlot runoff was almost 100% and 70%, respectively (Figures 8 and 9). Similarly, OP reduction was

approximately 90% and 70% in the first and second batch of experiments, respectively. The differences in TP uptake were due to the differences in initial TP concentrations in the feedlot runoff, differences in initial plant densities, and differences in microclimate of the greenhouse. The NH<sub>4</sub>-N uptakes by sorghum were close to 95% in both batches using undiluted feedlot runoff. The percentage reductions of NO<sub>2</sub>-N+NO<sub>3</sub>-N, and K from the feedlot runoff (undiluted) in the first batch experiment were approximately 75%, and 82%, respectively. A similar nitrate removal efficiency (64%-83%) and K removal efficiency (64%) was observed by Ayyasamy et al. (2009) and Gamage and Yapa (2001), respectively, using hydroponic plants. Similarly, reduction of potassium was less than 40%, and NO<sub>2</sub>-N+NO<sub>3</sub>-N was almost 10%, in the second batch of feedlot runoff. In the first batch, reductions of TP and OP by sorghum were more than 90%, and NH<sub>4</sub>-N and NO<sub>2</sub>-N+NO<sub>3</sub>-N were close to 100% in the Hoagland solution (Figure 8).

A similar nutrient reduction trend was also observed in Hoagland solution in both batches of experiments with sorghum. In some cases, nutrient reduction was more from the Hoagland solution than the feedlot runoff. Overall, the TP reduction from Hoagland in the first and second batches were 92% and 50%, respectively. Similarly, K reduction in the first Hoagland solution was 75%, whereas it was 85% in the second batch. The variation of percentage reduction of nutrient between first and second batches are likely due to density of plant, initial nutrient concentration. variation of and environmental conditions.

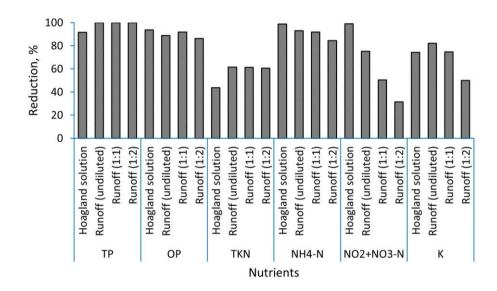


Figure 8 Overall nutrient percentage reduction by Sorghum plants in the first batch experiment

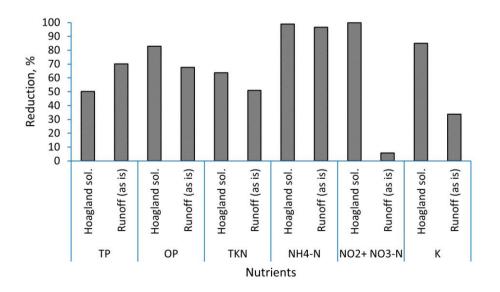


Figure 9 Overall nutrient percentage reduction by sorghum plants in the second batch experiment

### 3.11 Tissue mineral composition of the above ground plant part of plants grown in the feedlot runoff and Hoagland solution

Pooled mineral composition of above ground plant parts in the first and second batches of experiments are shown in Tables 5 and 6, respectively. Due to funding limitations of analysis, replicated samples were not analyzed for mineral composition, but to show the trends. Molybdenum (Mo) and zinc (Zn) concentrations were higher in water hyacinth, and sulfur (S) concentration was higher in sorghum plants grown in the diluted feedlot runoff (runoff 1:1 and runoff 1:2) than those grown in the feedlot runoff (undiluted). Most of the other elements were higher in plant tissue grown in the undiluted feedlot runoff than the diluted feedlot runoff (1:1 and 1:2). Sodium (Na<sup>+</sup>) concentration was higher in all plants grown in the feedlot runoff than the plants grown in the Hoagland solution, and this could have been due to the plant adjustment to the high salinity condition. It was noticed that the higher the dilution of feedlot runoff, the more Na concentration in plant tissues, as is shown in Table 6. The probable reason for higher Na accumulation in plant tissue is the ionic balance. When the feedlot runoff was diluted, the nutrient concentration, especially nitrogen concentration, decreased and was not sufficient for plant growth. Instead of NH<sub>4</sub><sup>+</sup> uptake in replacement of  $H^+$  ions, plants might have taken  $Na^+$  from the solution, thus increased Na concentration. The results show that the increasing order of  $Na^+$ concentration present in the plant tissue as a result of diluting feedlot runoff solution has a decreasing order of  $K^+$ concentration. Turhan and Eris (2005) also mention that  $Na^+$  is the competitive ion for  $K^+$ .

High Na<sup>+</sup> present in the solution hinders the uptake of  $Ca^{2+}$  in the plant tissues (Hu and Schmidhalter, 2005). which was shown true for all of plants grown in undiluted and diluted feedlot runoff (as-is shown in Table 6). From both batches and for all plants, concentration of P and Na was inversely proportional to each other. Choi and Lee (2012) reported that a higher level of P in soil reduced the iron (Fe), manganese (Mn), copper (Cu), and Zn concentrations in plant tissue because they are antagonistic in nature. In this study, an increasing P concentration in water hyacinth plant tissue showed a decreasing succession of Mo and Zn concentration and boron (B), calcium (Ca), Mg, S, Zn, and Fe concentrations in the first and second batches of the experiment, respectively. These conditions were also true for Mg, Mn, S, and Fe in the second batch of water lettuce tissues and for B, S, and Zn in the first batch and Mn and Fe in the second batch of sorghum tissues.

Table 5Mineral compositions (mean ±SD) of the above ground parts of three different plants grown in the
feedlot runoff and Hoagland solution in the first batch experiment. Mineral composition was determined by
the Inductively Coupled Plasma (ICP) system analyzer.

Treatment		Element	Element (mg/kg dry weight)											
Plant	Solution	Macronutrients				Micron	Micronutrients							
	$\mathrm{HS}^{*}$	Ca	Mg	Р	S	Fe	Mn	Zn	Cu	Na	В	Mo		
Water hyacinth	FRU	20131	5341	12110	4487	454	227.6	163.9	25.33	3571	41.85	12.64		
	FR1:1	17620	10188	5031	5587	2095	479.1	30.3	22.64	5229	28.84	0.68		
	FR1:2	14102	10506	4219	5455	1366	218.2	31.7	28.54	9829	22.59	0.88		
	HS	12896	10097	3697	5773	666	190	47.5	16.26	11087	21.57	1.16		
Water lettuce	FRU	15914	3678	8828	5172	200	150.8	248.9	25.79	3918	45.21	6.24		
	FR1:1	20465	9488	3727	3448	1546	623.1	55.2	13.69	10736	44.25	0.92		
	FR1:2	15914	7724	2983	3268	1145	329.4	37.9	13.01	11446	43.92	0.72		
	HS	15876	7326	2722	2707	724	211.2	34.7	12.35	14437	40.54	0.6		
Sorghum	FRU	7710	4045	5219	2430	367	78.4	41.6	7.09	802	59.29	1.8		
	FR1:1	20567	9816	3809	3380	5917	377.1	40.9	16.11	3450	34.32	<mdi< td=""></mdi<>		
	FR1:2	9631	6772	3765	5157	1611	179.4	27.3	9.98	3514	34.39	0.68		
	HS*	10896	7477	3094	5396	2081	188.6	41.7	14.78	3522	36.54	<mdl< td=""></mdl<>		

Note: Nutrient solutions: HS- Hoagland solution; FRU- Feedlot runoff undiluted; FR1:1 - Feedlot runoff 1:1 diluted; FR1:2 - Feedlot runoff 1:2 diluted;

Table 6Tissue mineral compositions (mean ±SD) of the above ground plant parts of the three differentplants grown in feedlot runoff and Hoagland solution in the second batch experiment. Mineral compositionwas determined by the Inductively Coupled Plasma (ICP) system analyzer.

Treatment		Element	Element (mg/kg dry weight)										
	Macron	Macronutrients				Micronutrients							
Plant	Solution	Ca	Mg	Р	S	Fe	Mn	Zn	Cu	Na	В	Мо	
Water hyacinth	Initial	14291	5685	9025	5012	126	32.2	16.23	15.7	11336	21.7	1.72	
	Feedlot runoff (undiluted)	12867	10119	5936	8135	554	420.3	109.97	12.67	7931	32.72	1.28	
	Hoagland solution	10508	6116	13279	4591	300	223.4	165.85	32.27	3192	27.64	25.15	
	Initial	23271	6586	6330	6847	306	47.8	24.51	14.88	15365	40.76	4.43	
Water lettuce	Feedlot runoff (undiluted)	12510	10553	6069	6780	2979	701.4	89.82	12.26	9984	37.04	2.04	
	Hoagland solution	24006	5828	8282	5614	749	359.5	1567.65	91.18	3289	46.74	41.16	
Sorghum	Initial	18251	9631	2716	4152	4287	237.1	45.8	17.85	7972	16.8	0.91	
	Feedlot runoff (undiluted)	6881	5494	5123	3635	1497	192.8	53.84	9.68	2394	23.82	0.44	
	Hoagland solution	9822	7088	8674	4476	388	124.9	64.95	13.32	1776	33.57	5.19	

#### 4 Conclusions

Water hyacinth, water lettuce, and sorghum plants uptook nutrients from both feedlot runoff and Hoagland solution, but sorghum reduced most of the  $NH_4$ -N concentration. Similarly, sorghum outperformed water lettuce and water hyacinth in removing TP from undiluted feedlot runoff in both batch experiments. On an average, sorghum also reduced more K (190%) than the other two plants in the Hoagland solution. Overall, sorghum outperformed water lettuce and water hyacinth in uptaking OP and K from feedlot runoff and Hoagland solution in both batch experiments.

Compared to water lettuce, water hyacinth and sorghum produced higher plant biomass in both batches of experiments with feedlot runoff and Hoagland solution. The overall net plant biomass average of water hyacinth, water lettuce, and sorghum in the Hoagland solution was 134%, 304%, and 344% more than the feedlot runoff, respectively. Therefore, there is a huge potential to remove nutrient runoff from feedlots using hydroponic plants. Sorghum might be the best candidate in treating nutrient runoff from feedlot.

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