Nitrogen Transfer in an Integrated System of Tilapia and Summer Bibb Lettuce

Celicina Azevedo, Mary W. Olsen and O. Eugene Maughan

Abstract

Nitrogen transfer from aquaculture to agriculture in integrated systems has been difficult to quantify, and the contribution of fish effluent to nitrogen utilization by crop plants is poorly documented. Therefore, labeled nitrogen, ^{15}N, was used in a small scale integrated system in a controlled environment greenhouse to determine nitrogen transfer from the fish system and its utilization by plants. Tilapia fish were grown in m^3 tanks and fed a complete diet that was amended with ^{15}N ammonium sulfate. Fish effluent amended with ingredients of a basic nutrient solution and 0%, 25%, 50% and 100% of the recommended mineral nitrogen was used to irrigate Summer Bibb lettuce in 5 gal lysimeters in two trials. In trial 1, percent ^{15}N recovered by lettuce was significantly higher in effluent amended with 50% mineral nitrogen as opposed to 0% or 100%. Dry weight of lettuce was significantly lower only when irrigated with effluent only (no mineral nitrogen added). In trial 2, percent ^{15}N recovered by lettuce was significantly higher in effluent amended with 25% mineral nitrogen as opposed to 0% or 50%. In treatments with the same amount of mineral nitrogen added, addition of effluent did not affect dry weight. Although ^{15}N recovery demonstrated efficient nitrogen transfer from the fish feed to the plants, results indicate that effluent alone does not supply sufficient nitrogen for lettuce production.

Introduction

Irrigation of agricultural crops with fish effluent increases the efficiency of water use by producing a second crop with the same water and virtually eliminates waste disposal problems (Olsen et al. 1993; D'Silva and Maughan, 1995). It also has been suggested that aquaculture effluents provide additional nutrients to crops when used for irrigation. Several studies have shown faster growth of plants when aquaculture effluent rather than water is used for irrigation (Olsen et al. 1993, Al-Jaloud et al. 1993, D'Silva and Maughan 1994, Khan 1996), but no studies have adequately quantified the amount of nitrogen transferred to plants from the effluent.

In recent years, labeled nitrogen, ^{15}N, has been used to characterize and quantify individual nitrogen fluxes in the soil-plant system and to evaluate the uptake of nitrogen fertilizer (Buresh et al. 1990, Bowen and Zapata 1991, Ledgard et al.. 1991, Mcgee et al.. 1995). The labeled N source can readily be identified and its uptake determined quantitatively. To quantify nutrient transfer in an integrated aquaculture and agriculture system, fish feed was labeled with ^{15}N and the effluent from an aquaculture facility was used to irrigate lettuce in a controlled environment greenhouse. The objective of this study was to quantify nitrogen uptake by lettuce from fish effluent as a determination of the nutrient contribution of the effluent in an integrated system.

Materials and Methods

Research was conducted in a controlled environment greenhouse at the University of Arizona Environmental Research Laboratory. Two trials were carried out using two independent one cubic meter fish tanks containing a 100 L biofilter filled with 20 L of beads. Each tank was filled with city water and five days later stocked with fish. In the first trial, the tank was stocked with large fish which were fed at 2% of their weight per day. In the second trial, tanks were stocked with small fish that were fed at 1.4% of fish weight per day.

Fish were regularly fed a commercial feed (32% protein) for 12 days before starting the experiment. All the fish were then weighed, and three fish were used for dry matter and nitrogen analysis. Fish were subsequently fed the ^{15}N labeled diet. The labeled diet was prepared by mixing ground commercial tilapia food (32% protein) with a water solution of ammonium ^{15}N sulfate at 10.3 atom percent of ^{15}N. Ammonium sulfate is used as a cattle feed supplement, and should not be hazardous to fish. Fish food and water containing ammonium ^{15}N sulfate were mixed, extruded dried and stored in the freezer. Effluent from the tanks was used to irrigate plants in certain treatments in both trials. Samples of fish effluent were collected before each irrigation and analyzed for total nitrogen and atom % of ^{15}N.

Summer Bibb lettuce (*Lactuca sativa* L.) seeds were sown in pre-formed peat pellets and transferred to lysimeters when they reached the three leaf stage. Each 20 L lysimeter was filled with 25.6 kg of washed river sand that had been previously dried in a glass house, and watered to field capacity with nutrient solution.

Trial 1 was a randomized complete block design (RCBD) with four treatments and four replicates. The treatments were: basic nutrient solution with 100% nitrogen combined with tap water (BN); basic nutrient solution with 100% nitrogen combined with fish effluent (BNE); basic nutrient solution with 50% of nitrogen combined with fish effluent (BN/2 E); basic nutrient solution with 0% nitrogen combined with fish effluent in which effluent was the only source of nitrogen (BE). All other nutrients in the basic nutrient solution remained in their original concentrations in the four treatments.

All data were analyzed by parametric methods using ANOVA. The percentage of ^{15}N recovered was analyzed only for treatments BNE, BN/2 E and BE because treatment BN did not receive fish effluent. When differences were significant, treatment means were examined with the linear combination of treatment means procedure (Kuehl, 1994).

Trial 2 was a factorial experiment with water source and nitrogen level as factors. Sources of water were tap water and fish effluent and levels of nitrogen were 50% , 25% and 0 % of nitrogen in basic nutrient solution. The six treatments were 50% of nitrogen in basic nutrient solution with tap water (BN/2), 50% of nitrogen in basic nutrient solution with fish effluent (BN/2 E), 25% of nitrogen in basic nutrient solution with tap water (BN/4), 25% of nitrogen in basic nutrient solution with fish effluent (BN/4 E), 0% of nitrogen in basic nutrient solution with tap water (B), 0% of nitrogen in basic nutrient solution with fish effluent (BE). Four replicate lettuce plants were used for each of the six treatments.

Data with heterogeneous variances were transformed, and percentage of ^{15}N in excess of background was transformed using logit transformation. Data were analyzed by parametric methods using a factorial analysis of variance with type of water, nitrogen level, blocks and type of water x nitrogen level (interaction effect) as factors. The percentage of ^{15}N recovered was analyzed only for BN/2 E, BN/4 E and BE because treatments BN/2, BN/4, and B did not receive the labeled fish effluent. Therefore the analysis was conducted as a two way analysis of variance, with treatments and blocks as effects. When differences in these analyses were significant, treatment means were examined with the linear combination of treatment means procedure (contrast analyses).

Total nitrogen and atoms % of ^{15}N were measured in irrigation water before each irrigation. A pooled sample (prepared according to the amount of water in each irrigation) was also analyzed. Lettuce plants were harvested at marketable size. Plants were oven dried at 60° C for dry weight determination. Total nitrogen content in water and plants was determined by the micro-Kjeldahl method modified to recover NO_3^- (Bremmer and Mulvaney 1982). Distillates from nitrogen analyses were analyzed for ^{15}N using a VG 602c light gas isotope mass spectrometer. Total nitrogen uptake by lettuce was calculated by multiplying the total nitrogen content in lettuce by the dry weight.

The amount of total nitrogen (N) retained in lysimeters was calculated by multiplying the water used by the concentration of nitrogen in irrigation water. To calculate the percentage of ^{15}N recovered by plants, ^{15}N background levels were subtracted from all measured levels of ^{15}N. Therefore, ^{15}N levels reported represent the difference between normal background and measured values. The amount of ^{15}N in the lysimeter was calculated by:

^{15}N in lysimeter (μg) = Total N (μg)$_{\text{in lysimeter}}$ * Atoms % of ^{15}N in irrigation water / 100.

After the amount of ^{15}N was calculated for each irrigation, the total amount was summed for each treatment and block.

The amount of ^{15}N in harvested lettuce was calculated by:

^{15}N in lettuce (μg) = Total N taken up (μg)$_{\text{in lettuce}}$ * Atoms % of ^{15}N in lettuce/ 100.

The percentage of recovery of labeled nitrogen from the fish effluent by the lettuce plants was estimated by dividing the amount of ^{15}N in harvested lettuce by the amount of ^{15}N retained in the lysimeter (all irrigations summed).

Results and Discussion

In both Trial 1 and Trial 2 , the ratio of inorganic to organic nitrogen varied among treatments, increasing with the amount of inorganic nitrogen in nutrient solution (Figures 1 and 2). The ^{15}N atoms % increased as the amount of inorganic nitrogen decreased in both trials.

In both trials, the mean percentage of ^{15}N recovered by lettuce differed among treatments. In Trial 1, the percentage recovered was 15.11% for BNE, 39.91% for BN/2E, and 23.67% for BE (Figure 3). Recovery was significantly different in treatment BNE than in BN/2E (p=0.003) and in treatment BN/2E than in BE (p=0.004). In Trial 2, the mean percentage of ^{15}N recovered by lettuce was 57.00% for BN/2E, 71.35% for BN/4E, and 53.76% for BE (Figure 4). Recovery in treatment BN/2E was significantly different than in BN/4E (p=0.03) and in treatment BN/4E than in BE (p=0.02).

Lettuce production, based on dry weight, varied by treatment in both trials. In Trial 1, dry weight was significantly different only for treatments BN, BNE and BN/2E combined versus treatment BE (p<0.0001). Dry weight production was eight times higher in treatments BN, BNE and BN/2E than in treatment BE (Figure 5). In Trial 2, treatment effects were significantly different for B versus BE (p<0.0001) and for BN/2 and BN/2E combined versus BN/4 and BN/4E combined (p=0.004). Mean dry weights of lettuce in treatments B and BE were only about 20% of that in treatments BN/2 and BN/2E (Figure 6).

Plants can utilize only inorganic nitrogen, and in both trials, plants used virtually all of the inorganic nitrogen in the effluent when effluent was the only source of nitrogen. In Trial 1, lettuce irrigated with BE, the basic nutrient solution with only fish effluent added, recovered 24% of the ^{15}N applied (Figure 3) and only 24% of the nitrogen in the effluent was inorganic (Figure 1). In Trial 2, the percent inorganic N in BE was higher as was the per cent recovery.

Despite the fact that the efficiency of recovery of inorganic nitrogen from the effluent was high in both trials, the effluent did not provide enough inorganic nitrogen for suitable plant growth. When inorganic nitrogen is added to the basic nutrient solution with effluent either as 100%, 50% or 25% of the nitrogen given for the complete nutrient solution, there were no differences in dry weights. Only increasing the inorganic nitrogen increased dry weights.

If fish effluent alone does not provide sufficient nitrogen for lettuce production, it seems reasonable to use a combination of effluent with commercial chemical fertilizer as suggested by Al-Jaloud et al. (1993). However, the per cent recovery of ^{15}N decreased significantly as the amount of inorganic nitrogen was increased from 50% to 100% in Trial 1 and from 25% to 50% in Trial 2. Therefore, some of the nitrogen in the effluent was lost when the two nitrogen sources were combined. It is unknown if this relationship was unique to this experiment or is consistent in other systems.

References

Al-Jaloud, A.A., G. Hussain; A.A. Alsadon; A.Q. Siddiqui and A.Al-Najada. 1993. Use of aquaculture effluent as a supplemental source of nitrogen fertilizer to wheat crop. *Arid Soil Research and Rehabilitation* 7 **:**223-241.

Bowen, G.D. and F. Zapata. 1991. Efficiency in uptake and use of nitrogen by plants. Pages 349-362.*In Stable Isotopes in Plant Nutrition, Soil Fertility and Environmental Studies.*

Bremmer, J.M. and C. S. Mulvaney. 1982. Nitrogen-total. Pages 595-624. *In*: A .L. Page, R.H. Miller, D.R. Keeney (eds.) *Methods of Soil Analysis. Part 2, Chemical and Microbiological Properties*. American Society of Agronomy, Soil Science Society of America, Madison, WI.

Buresh, R.J., P.L.G. Vlek and K. Harmsen. 1991. Rate of fertilizer nitrogen applied to wheat under simulated Mediterranean environmental conditions. *Fertilizer Research* 23(1) 25-36.

D'Silva, A.M. and O.E. Maughan. 1994. Multiple use of water: integration of fishculture and tree growing. *Agroforestry Systems* 26:1- 7.

D′Silva, A.M. and O.E. Maughan. 1995. Effect of density and water quality on red tilapia in pulsed flow culture systems. *Journal of Applied Aquaculture* 5:69-75.

Kuehl, R.O. 1994. *Statistical Principles of Research Design and Analysis.* ITP International Thompson Publishing.

Ledgard, S.F., G.S. Smith; and J.A. Douglas. 1991. Utilization of fertilizer and stored nitrogen by asparagus and kiwi fruit estimated using ^{15}N. Pages 371-377. *In: Stable Isotopes in Plant Nutrition, Soil Fertility and Environmental Studies. Proceedings of an International Symposium on the Use of Stable Isotopes in Plant Nutrition, Soil Fertility and Environmental Studies.* International Atomic Energy Agency, Vienna.

Mcgee, E.A., T.L. Tompson, and T.W. McCreary, 1995. Apparatus for application of ^{15}N fertilizers through buried OMP tubing. *Soil Science Society of America Journal* 59(5):1495-1498.

Olsen, M.W., K..M. Fitzsimmons and D.H. Moore, 1993. Surface irrigation of cotton using aquaculture effluent. Pages 159-165. *In: Techniques for Modern Aquaculture* (J.K. Wang, Ed.) St. Joseph, MI:ASAE pub. 02-93.

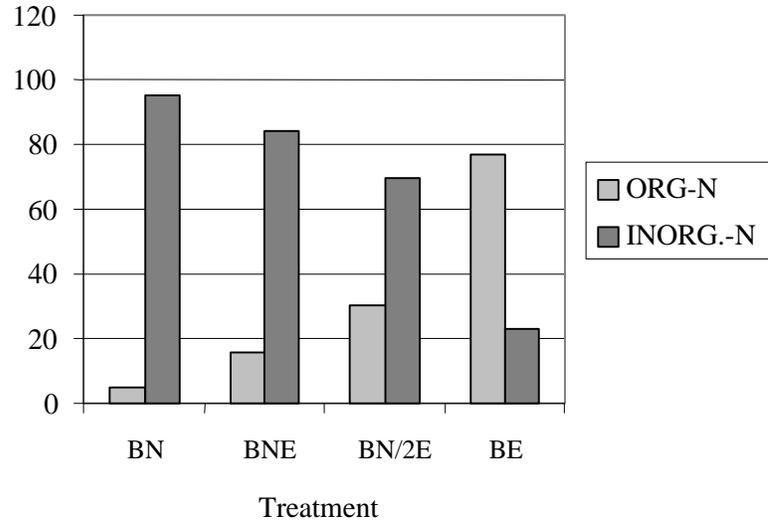Figure 1. Percentage of organic and inorganic nitrogen in irrigation water in Trial 1.

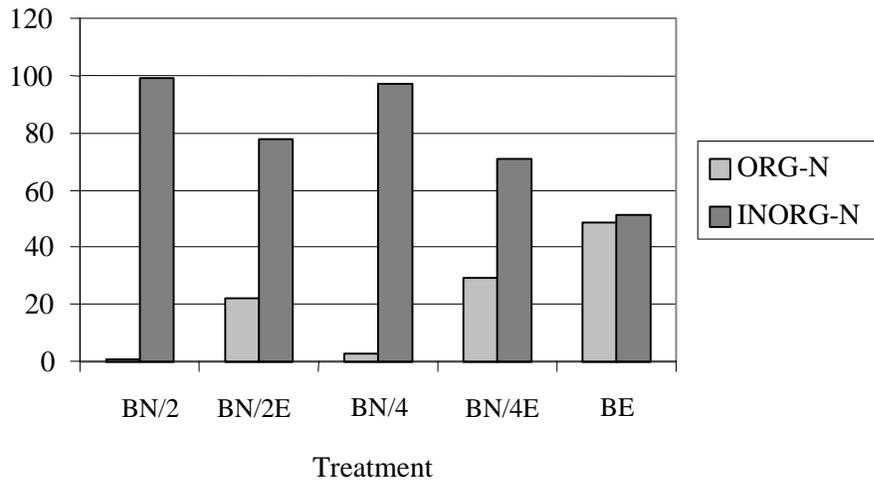Figure 2. Percentage of organic and inorganic nitrogen in irrigation water in Trial 2.

Figure 3. Mean percentage of ^{15}N recovered by lettuce in Trial 1.
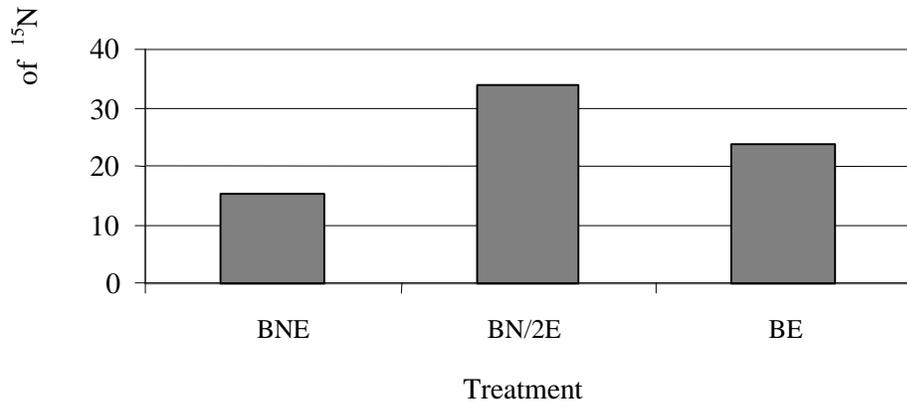
of ^{15}N

40

30

20

10

0

BNE   BN/2E   BE

Treatment

Figure 4. Mean percentage of ^{15}N recovered by lettuce Trial 2.

^{15}N recovered

80

70

60

50

40

30

20
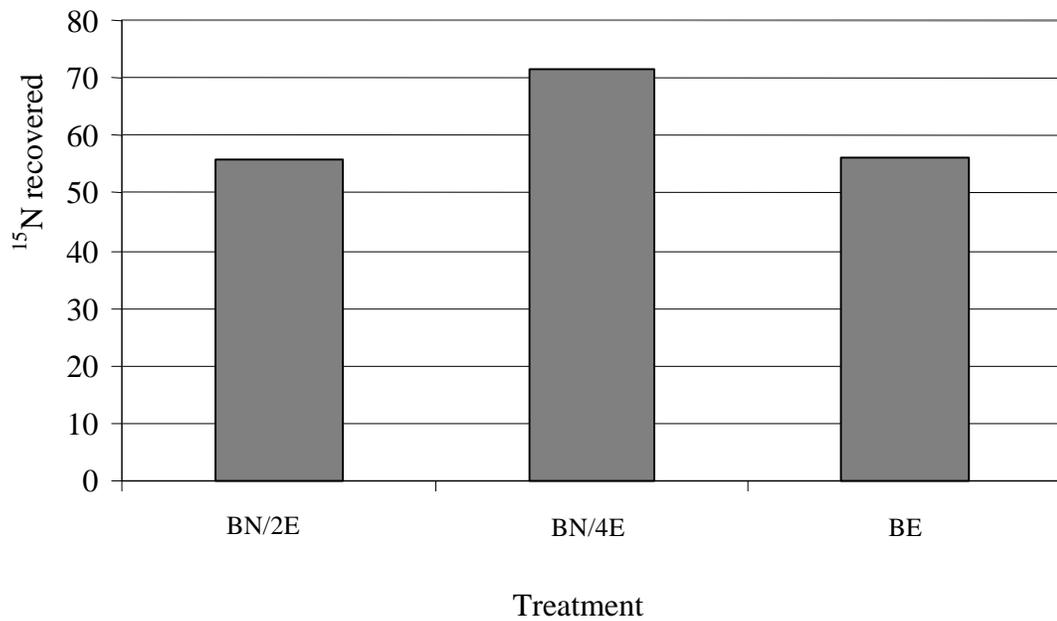
10

0

BN/2E   BN/4E   BE

Treatment

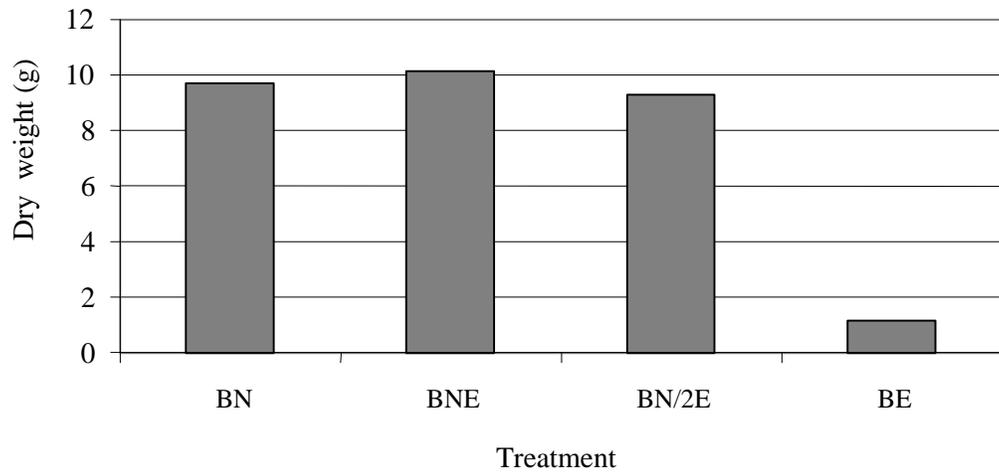Figure 5. Mean dry weight (g) of lettuce produced in Trial 1.

Figure 6. Mean dry weight (g) of lettuce produced in Trial 2.