

# Evaluation of a Nitrogen-15 Microplot Design in a Furrow Irrigated Row Crop System

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## Abstract

Two field experiments were conducted in Arizona in at two locations, Maricopa in 1991 (Casa Grande sandy loam) and Marana (Pima clay loam) in 1995. The purposes of the experiments were to evaluate the dimensions of an  $^{15}\text{N}$  microplot design used in a furrow irrigated row crop system. The experiments each utilized ammonium sulfate fertilizer with 5 atom %  $^{15}\text{N}$  enrichment applied at a rate of 56 kg N/ha in simulated side-dress band application during the early bloom stage of development of Upland cotton (*Gossypium barbadense* L.). At each location, microplots were 4, 1.02 m rows wide and 1.00 m in length. Whole plant samples were collected at specific locations within and near the microplots. Uptake of  $^{15}\text{N}$  by plants was uniform within microplots but declined symmetrically in relation to microplot borders. Collection of plant materials within 25 cm of microplot borders provided uniform  $^{15}\text{N}$  enrichment levels for determining fertilizer N uptake and recovery. Use of microplots with the dimensions of those used in this study are sufficient for collecting plant materials from a 1 m<sup>2</sup> area; consisting of two, 50 cm segments from the interior two rows of the four row microplot. This also allows for sufficient distance from the perimeter of the microplot to account for border effects.

## Introduction

The management of nitrogen (N) nutrition is a very important aspect of any cotton (*Gossypium* spp.) production in Arizona or any of the irrigated regions of the desert Southwest. Agronomically, it is important for farmers to use fertilizer N efficiently to maintain optimum return in yield for the amount of fertilizer N provided. Also, from an environmental standpoint, it is important to manage fertilizer N so that downward movement of  $\text{NO}_3^-$ -N in the soil profile, can be minimized. Therefore, the ability to explicitly trace fertilizer N inputs to an irrigated cotton production system would be valuable in addressing many important questions.

Nitrogen is a rather ubiquitous element in a soil-plant system, occurring in many forms and being subject to a myriad of transformations. Therefore, it is very difficult to explicitly trace N in any soil-plant system that is derived from fertilizer sources. One technique available for tracing fertilizer N in a soil-plant system involves the use of N fertilizer materials containing enriched levels of  $^{15}\text{N}$ , a stable, naturally occurring isotope of N. In nature,  $^{14}\text{N}$  occurs at a rather constant ratio to  $^{15}\text{N}$  of 272:1 ( $^{14}\text{N}:$  $^{15}\text{N}$ ) or 0.3663 atom %  $^{15}\text{N}$  in naturally occurring N, such as atmospheric  $\text{N}_2$  gas. Utilization of N fertilizer sources containing enriched or depleted levels of  $^{15}\text{N}$  significantly different than 0.3663 atom %  $^{15}\text{N}$  provide a direct means of tracing N from those sources through a soil-plant system and explicitly differentiating it from indigenous soil N sources.

Although  $^{15}\text{N}$  labeled fertilizers provide potentially valuable tools for agricultural research, the costs associated with the use of this technique are high. The costs for the  $^{15}\text{N}$  labeled materials are very expensive. Techniques associated with field and laboratory aspects of  $^{15}\text{N}$  methodology are also very tedious and expensive in relation to conventional techniques using non-labeled fertilizer N sources. To sufficiently address some questions in agricultural research regarding the fate of N fertilizers in specific soil-plant systems, methods utilizing  $^{15}\text{N}$  labeled N sources are required. To efficiently utilize resources, researchers must simultaneously address two important objectives when dealing with  $^{15}\text{N}$  labeled materials in the field. These objectives are: 1) applying  $^{15}\text{N}$  labeled fertilizers to a sufficient area to accommodate appropriate amounts of plant and /or soil sampling and 2) to conserve on the amount of  $^{15}\text{N}$  labeled fertilizers applied and utilized in the study due to the high cost of the materials. Due to these two constraints,  $^{15}\text{N}$  labeled fertilizers are commonly applied to relatively small experimental units, commonly referred to as microplots.

One of the first steps necessary in implementing a program involving  $^{15}\text{N}$  fertilizers in the field is to address appropriate size and dimensions associated with the microplot areas receiving the labeled fertilizer materials.

A number of studies have been conducted with various crops and cropping systems to determine the appropriate sizes and dimensions of microplots for using  $^{15}\text{N}$  labeled fertilizers. Barriers have been used to eliminate lateral movement from microplot areas (Carter et al., 1967). However, these types of barriers create a number of artificial factors such as restricted root development, disturbed macropore arrangements, unnatural soil-water movement and drainage, and restriction of normal cultivation/tillage practices. Bufogle et al. (1997) compared open microplots with retainers (barriers) in a flooded rice (*Oryza sativa* L.) and concluded that 0.75 m<sup>2</sup> microplots with retainers best simulated large field plot in this type of a flood irrigation system.

The use of open microplot techniques have been more commonly recommended (Hauck et al., 1994) and developed. Johnson and Kurtz (1974), Olson (1980), Sanchez et al. (1987), Jokela and Randall (1987), Stumpe et al. (1989), and Blaylock et al. (1990) each worked with non-irrigated corn (*Zea mays* L.) row crop systems. Based on the earlier work in row cropping systems,  $^{15}\text{N}$  microplot areas with 2 X 2 m dimensions (usually with four rows of plants in the microplot area) would appear to be sufficient for uptake and recovery studies when plants are sampled from the interior rows. Maintaining a distance of 0.5 m (intrarow) from the border for plant samples would also be reasonable to conclude from this work. Follett et al. (1991) determined that minimum microplot areas of 1.5 by 1.5 m were adequate in winter wheat (*Triticum aestivum* L.) and that plants could be sampled 0.46 m from the microplot border without affecting  $^{15}\text{N}$  concentrations. In a buried drip irrigation system working with leaf lettuce (*Lactuca sativa* L.), McGee et al. (1995) found that microplots 1.02 by 2.00 m, with two planted rows of lettuce within, were adequate for plant and soil sampling needs. The uptake  $^{15}\text{N}$  was uniform within the microplots, even when plants were sampled 25 cm from the border.

Although a considerable amount of work has been done to address this issue, studies have not been conducted to explicitly address microplot dimensions needed for use in a furrow irrigated system. Drawing upon the information available from previous research, a project was initiated in Arizona at two locations. The objective of this project was to evaluate the dimensions of a  $^{15}\text{N}$  microplot design for use in a furrow irrigated row crop system with cotton.

## Materials and Methods

Field experiments were conducted in 1991 at the University of Arizona Maricopa Agricultural Center (MAC, 357 m elevation) and 1995 at the Marana Agricultural Center (MAR, 600 m elevation). Upland cotton (cv. DP 90) was planted on a Casa Grande sandy loam [fine-loamy mixed, hyperthermic Typic Natragid, reclaimed] on 19 April 1991 at MAC and on 24 April 1995 at MAR on a Pima clay loam [fine-silty, mixed (calcareous), thermic Typic Torrifluent]. At both locations, irrigation runs were 183 m in length. Row spacings were 1.02 m and planting was accomplished with a conventional planter. Microplots for  $^{15}\text{N}$  applications were placed approximately 30 m apart, in the center four rows of eight row mainplots that extended the full length of the irrigation run. Microplots were not placed within 30 m of the head (irrigation ditch) or tail of the field. Microplots were four, 1.02 m wide by 1.00 m in length. A total of six microplots were established at each location with two microplots/main plot and three mainplots. Following stand establishment, each row was thinned to an intrarow plant density of 12 plants/m.

Applications ( $^{15}\text{NH}_4$ )<sub>2</sub>SO<sub>4</sub> with 5 atom %  $^{15}\text{N}$  enrichment were made by simulated side-dress applications on the beds of each of the four rows in each microplot at a rate of 56 kg N/ha between first bloom and peak bloom stages of crop development at both locations. To simulate the side-dress applications, trenches were cut in the sides of the beds approximately 15 cm to the side and below the center of the row. Solutions containing appropriate amounts of ( $^{15}\text{NH}_4$ )<sub>2</sub>SO<sub>4</sub> dissolved in 500 ml distilled-deionized water were applied uniformly by hand in the each of the trenches cut by each row and immediately covered by dry soil. All cultivation, irrigation, and pest management practices were conducted in a uniform manner on an as-needed basis at both locations. Final irrigations were applied on 11 September 1995 at MAC and 1 August 1995 at MAR.

Plant samples were collected by removing the entire aboveground portions of individual plants from microplot areas on 26 September 1991 at MAC and 3 October 1995 at MAR according to the sampling scheme outlined in Figure 1. Plant samples associated with each plot and position designation were appropriately labeled, transported from the field, dried, and ground to pass through a 20-mesh screen. Subsamples were further ground to a very fine powder

and subjected to total N and isotope ratio analysis by use of a Carlo-Erba N/A 1500 Elemental Analyzer and a VG Isomass mass spectrometer. Relative fractions (RF) were determined in the following manner:

$$F = \text{fraction of labeled N in sample} = (A_s - A_r) / (A_f - A_r)$$

where:

$A_s$  = atom %  $^{15}\text{N}$  in sample

$A_r$  = atom %  $^{15}\text{N}$  in reference material (check)

$A_f$  = atom %  $^{15}\text{N}$  in fertilizer

$$\text{RF} = F_x (\text{sample } x) / F_1 (\text{center of microplot})$$

Results were analyzed statistically in accordance to procedures outlined by Steel and Torrie (1980) and the SAS Institute (SAS, 1988).

## Results

Atom %  $^{15}\text{N}$  concentrations in plant samples collected in the vertical (head to tail) dimension from at MAC in 1991 and MAR in 1992 are presented in Figures 2 and 3. Samples collected upslope to downslope are placed from left to right, with the microplot borders indicated with bold vertical lines and the atom %  $^{15}\text{N}$  in the plants from the check plots shown with the bold horizontal lines. In both cases, the atom %  $^{15}\text{N}$  concentrations were uniform from all samples collected within the microplot borders. At MAC the decline in atom %  $^{15}\text{N}$  was quite rapid outside the microplot boundary and returned very close to natural abundance (check plot levels) in samples collected 0.75 m outside the microplots. At MAR, slightly higher proportions of atom %  $^{15}\text{N}$  were detected in samples collected 0.25 m outside the microplot border than at MAC.

Distributions of atom %  $^{15}\text{N}$  in samples collected in the horizontal dimension across the microplot areas are presented in Figures 4 and 5 for MAC and MAR. Atom %  $^{15}\text{N}$  concentrations were quite uniform from all samples collected within microplot borders at both MAC and MAR. At MAC, atom %  $^{15}\text{N}$  levels were slightly lower in the center of the microplots compared to the samples taken at the lateral edges (outside rows), where they were quite similar at both locations. Atom %  $^{15}\text{N}$  concentrations were near natural abundance in samples taken from the first rows outside and immediately adjacent to the microplot areas at both locations (2.5 m from microplot centers).

One interesting way of evaluating these results is in terms of the symmetry associated with the  $^{15}\text{N}$  levels found in plant samples in relation to sample location within and beyond the microplot borders. Relative fractions of  $^{15}\text{N}$  are plotted in relation to distance from microplot border at MAC and MAR in Figures 6 and 7. Theoretically, RF levels should be about 0.5 at the microplot border assuming root-system sorption zone crop uptake of the fertilizer N and an equal opportunity of plants to utilize N derived from fertilizer and indigenous soil N (Sanchez et al., 1987). At both MAC and MAR, the RFs were symmetrical in terms of both up-slope and down-slope distributions and returned to near natural abundance levels approximately 0.5 m outside the microplots.

## Conclusions

The results of this study agree quite well with earlier work in row crop systems. Based on these results, a microplot design consisting of a minimum of 1m by 2 m dimension could be adequate for determining uptake and recovery of  $^{15}\text{N}$  labeled fertilizers. A suggested protocol for plant sample collection within a 1 m X 2 m microplot in a furrow irrigated crop system consisting of four, 1.02 m rows and 1 m in length, could include plant samples collected from a 1 m<sup>2</sup> area in the center of the microplot. The 1 m<sup>2</sup> area could consist of two, 0.5 m segments taken from the center two rows of the four row microplot. This would allow for 0.5 and 1.0 m distances from the microplot borders, vertically and horizontally, respectively.

## Acknowledgements

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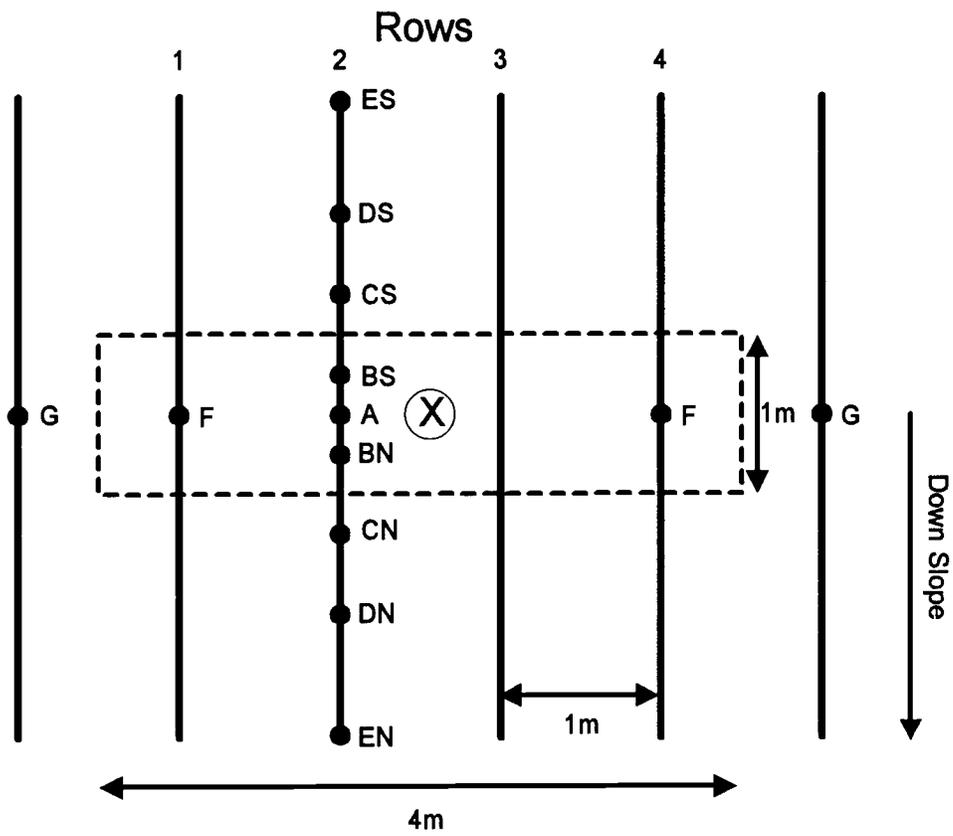


Figure 1 Microplot design and plant sampling locations for both 1991 and 1995.

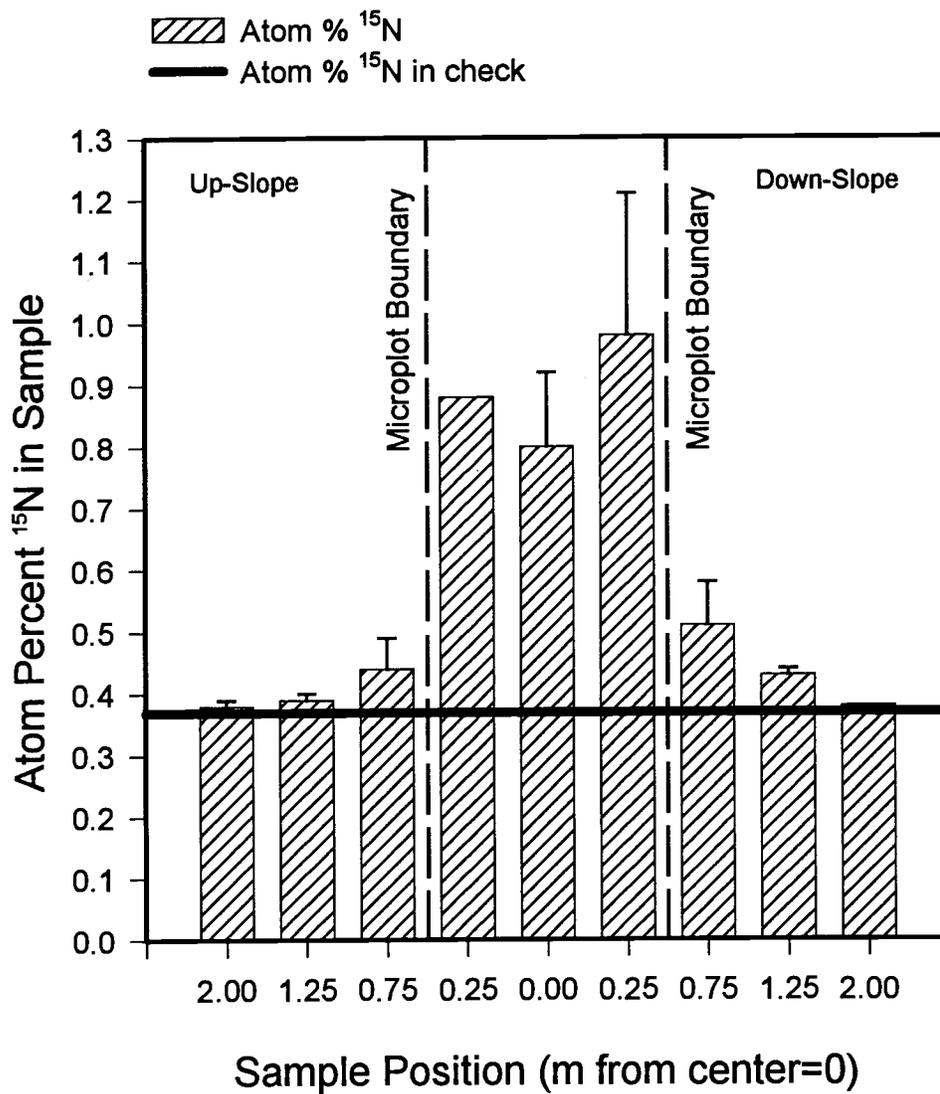


Figure 2. Distribution of  $^{15}\text{N}$  from microplot areas, Maricopa, AZ, 1991.

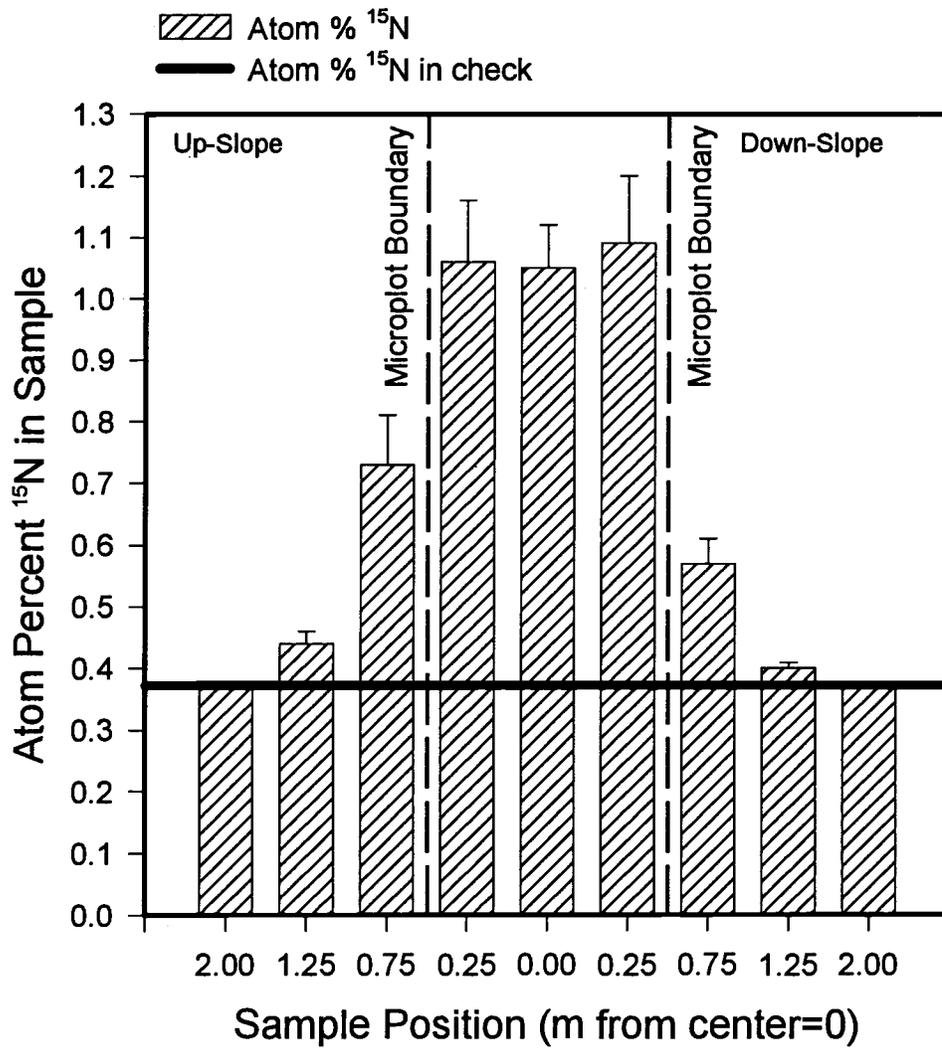


Figure 3. Distribution of <sup>15</sup>N from microplot areas, Marana, AZ, 1995.

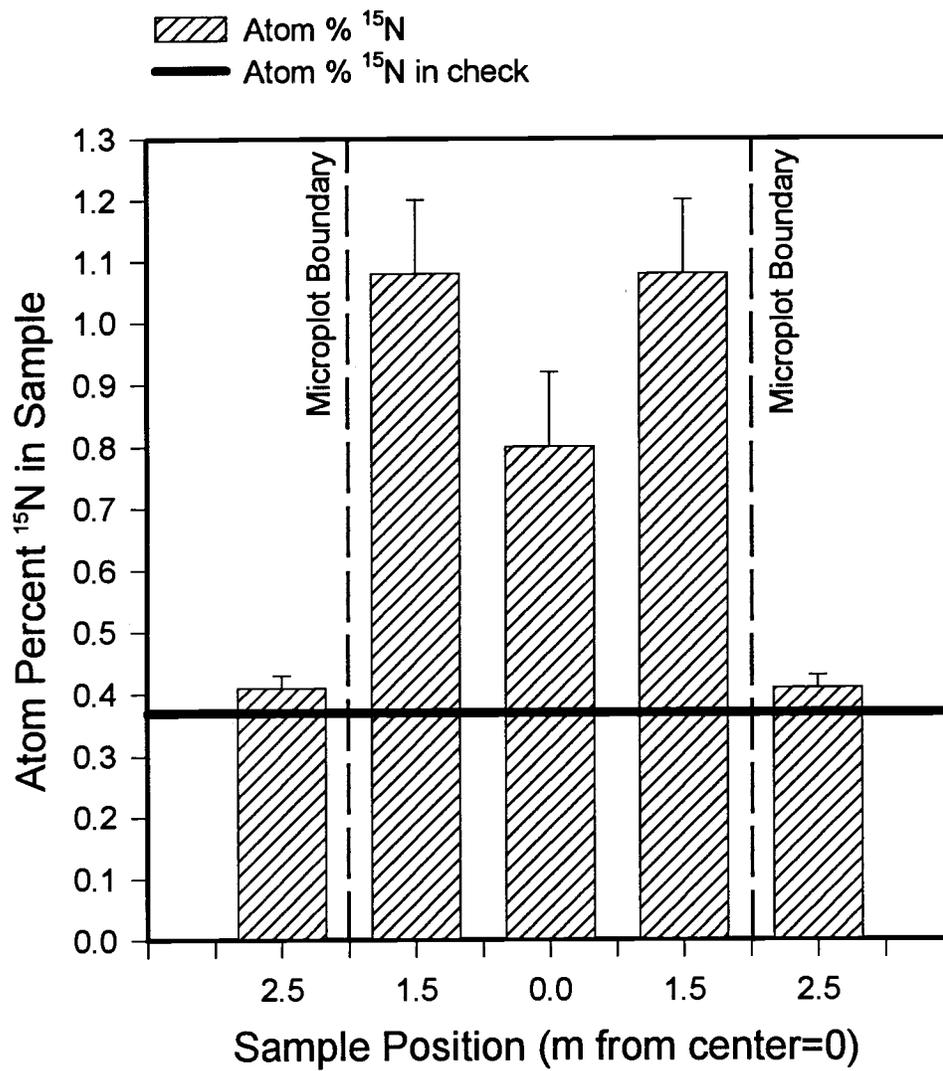


Figure 4. Lateral distribution of <sup>15</sup>N across microplot areas, Maricopa, AZ, 1991.

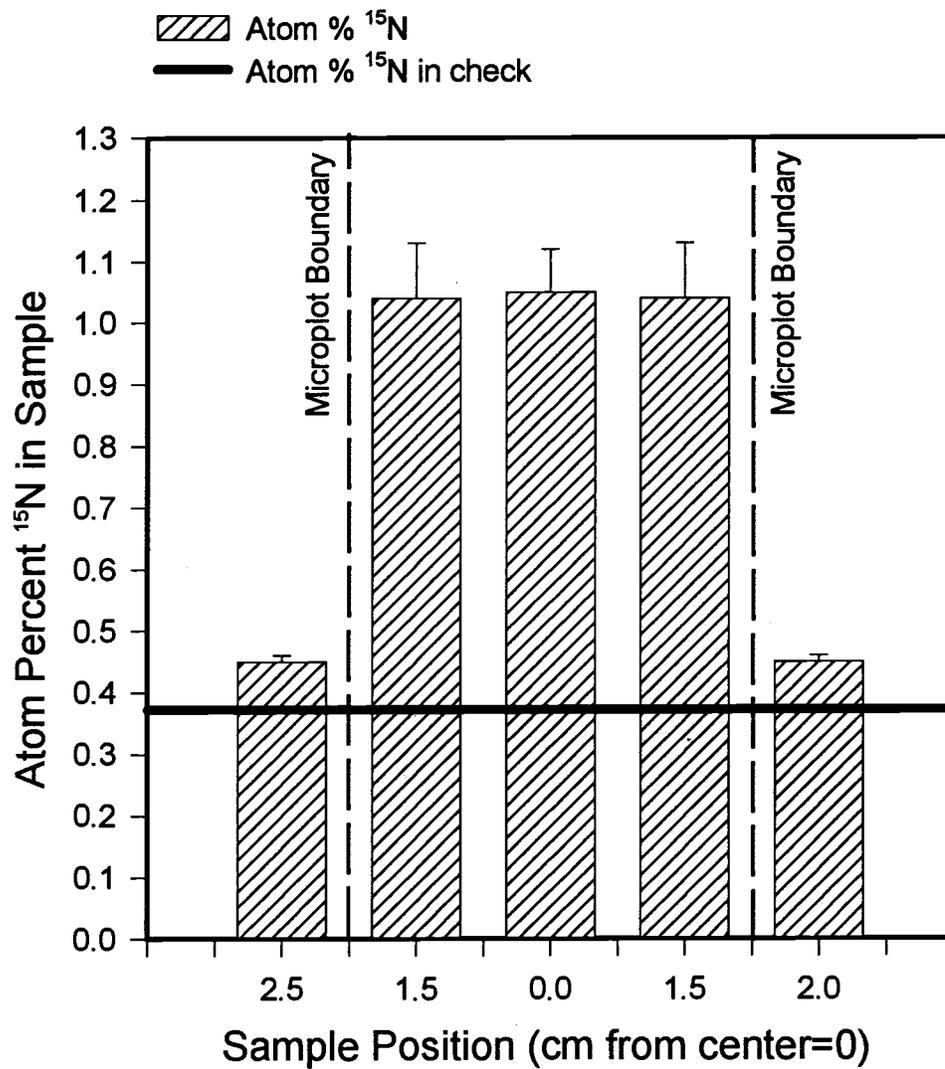


Figure 5. Lateral distribution of  $^{15}\text{N}$  across microplot areas, Marana, AZ, 1995.

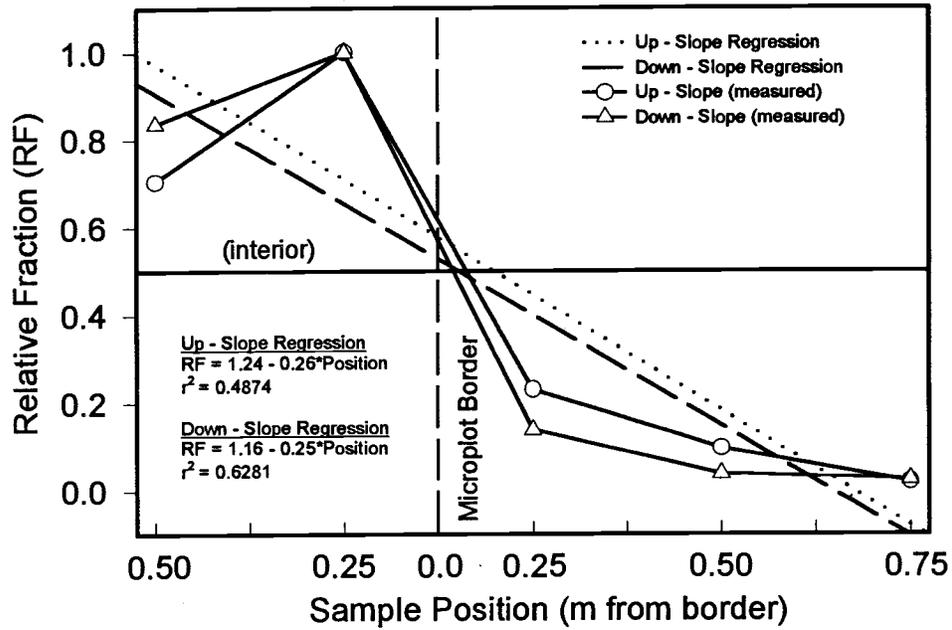


Figure 6. Symmetry of relative fraction of  $^{15}\text{N}$  across microplot areas, Maricopa, AZ, 1991.

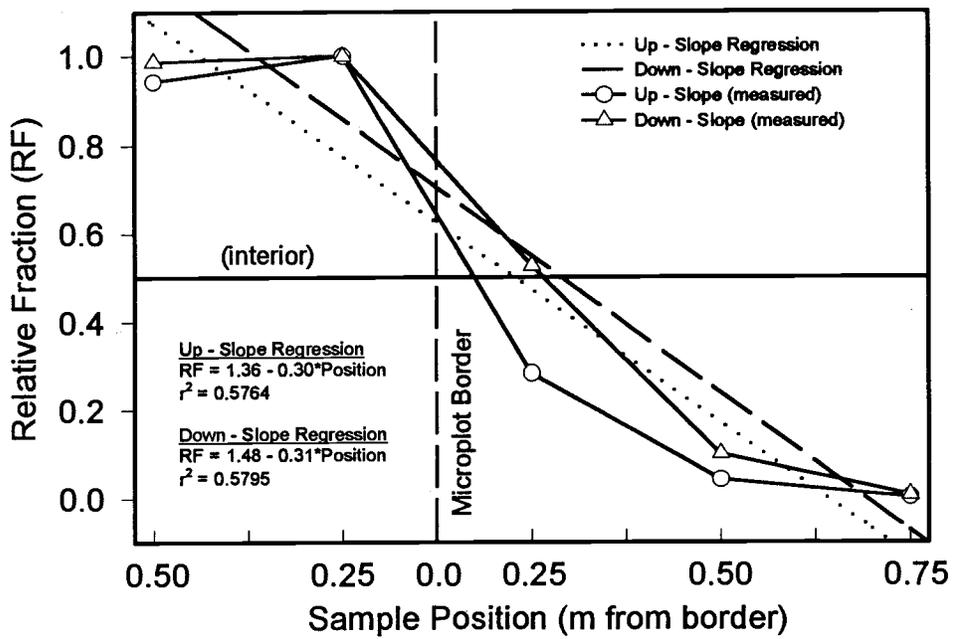


Figure 7. Symmetry of relative fraction of  $^{15}\text{N}$  across microplot areas, Marana, AZ, 1995.