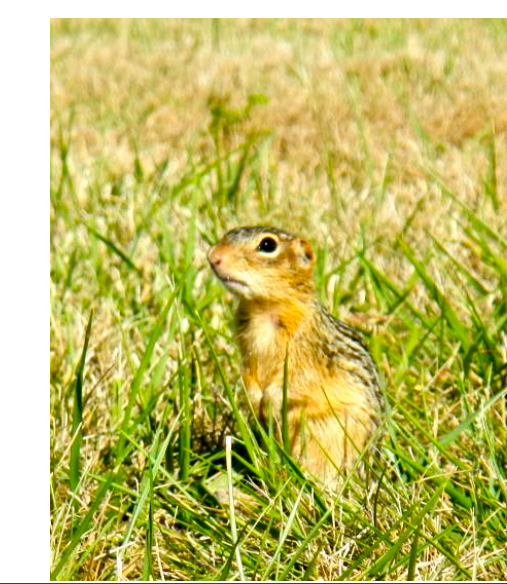


# Investigation of spatial isotope ratios in soil and the effects of fertilizer on plant isotope ratios



Jamie Kim and H. Thomas Goodwin

<sup>1</sup>Biology Department, Andrews University  
Jamiek@andrews.edu

## Abstract

A previous Honors study analyzed diet of thirteen-lined ground squirrels at the Andrews University Airpark by studying  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotope values of their fecal pellets (Chacko, 2013). Fecal samples collected within 25m of a cornfield had elevated isotope values.

High  $\delta^{13}\text{C}$  values indicate greater consumption of C4 plants, such as corn (Ehleringer et al., 1986); high  $\delta^{15}\text{N}$  values indicate increased consumption of animal matter (DeNiro and Epstein, 1981). However, the spatial pattern may be due to variation in the soil, reflecting long-term patterns in the vegetation.

Soil samples were taken throughout the airpark and analyzed for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ . These baseline values were removed from fecal values, but the patterns of the fecal samples still held.

Application of fertilizer to vegetation resulted in a significant increase in  $\delta^{15}\text{N}$ . Thus, the  $\delta^{15}\text{N}$  pattern observed in the fecal samples appears to be due to enrichment in corn.

## Methodology

Soil samples were collected throughout the airpark (Fig. 1) and analyzed for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotope values by the Stable Isotope Ratio Facility for Environmental Research (SIRFER) at the University of Utah. Baseline values of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  were calculated for each fecal sample by averaging the soil isotope values within 15m of the fecal sample. These baseline values were then subtracted from the fecal sample values in order to see if the spatial pattern persisted. T-tests were conducted between fecal sample isotope values within 25m of the cornfield and those further away.

The AU Department of Agriculture provided the cow manure and ammonium nitrate that was used to fertilize the cornfield. Eight plots were set up around the airpark, and each plot contained five separate 0.25 m<sup>2</sup> areas that were used as experimental groups (Fig. 2). The five groups were 1) nothing applied, 2) only water, 3) water and ammonium nitrate, 4) water and cow manure, and 5) water, ammonium nitrate, and cow manure. After 8 weeks, grass clippings were collected and measured for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ . The data were analyzed using a One-Way Analysis of Variance (ANOVA) then a post-hoc Tukey multiple comparisons test.

Figure 1 – Soil sample locations, along with  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of the samples

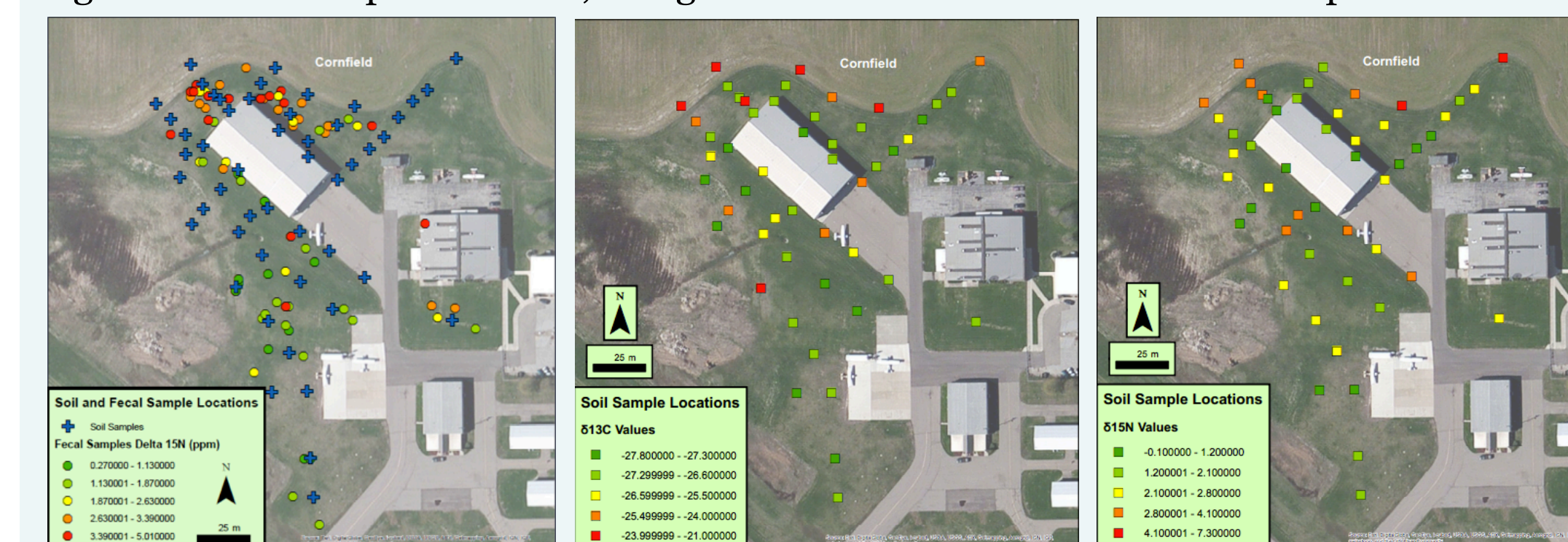
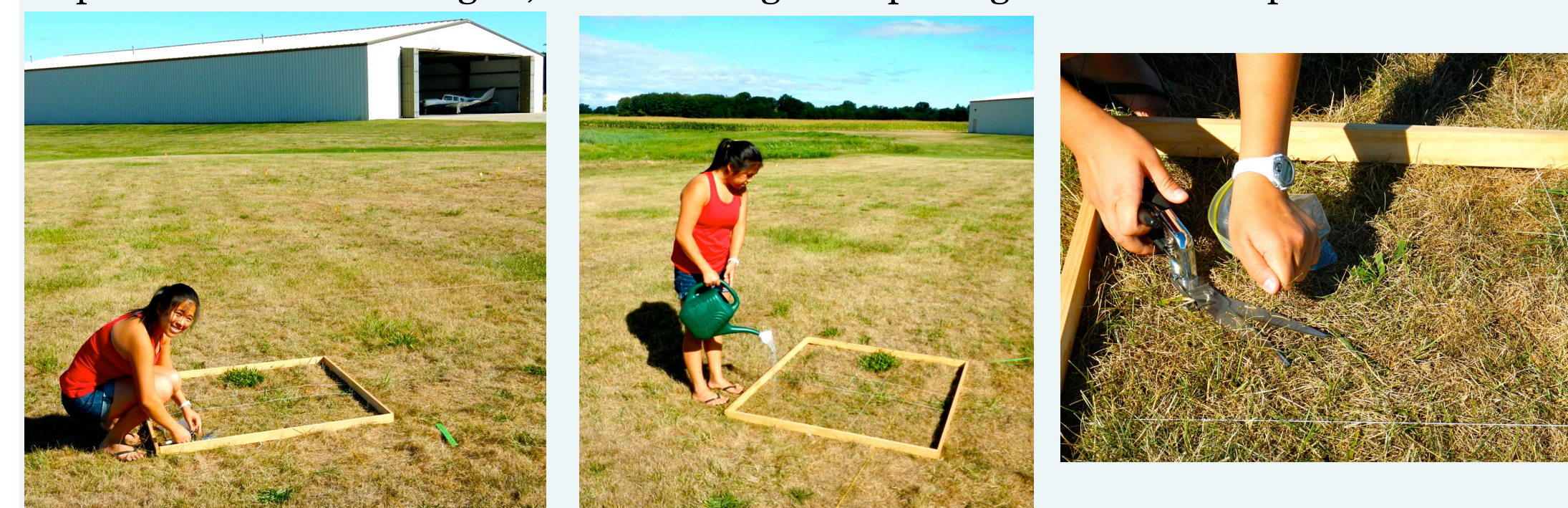


Figure 2 – The pictures below show the grid used for the fertilizer experiment, watering an experimental area in the grid, and collecting a sample of grass from an experimental area.



## Results

### Soil Study:

Soil  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values were not significantly higher near the cornfield. After the baseline soil  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values were removed from the fecal sample isotope values, the spatial pattern of elevated ratios near the cornfield remained (Fig. 3).

Figure 3 – Fecal sample  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotope values plotted by location on a map of the AU Airpark, before and after soil baseline removal.

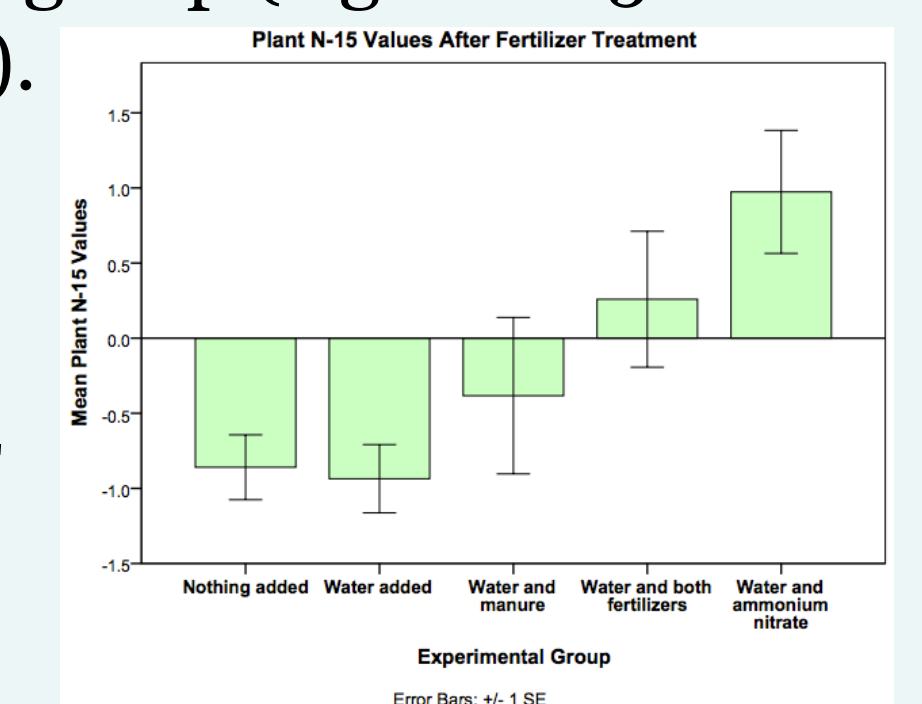
The bar graphs indicate the isotope value means of the fecal samples obtained near the cornfield (within 25m) and further away. Error bars: +/- 1 SE



### Fertilizer Experiment:

The One-Way ANOVA indicated a significant variance across the groups ( $p < 0.010$ ;  $F = 4.207$ ,  $df = 4, 34$ ). The post hoc test (Tukey HSD) identified the water and ammonium nitrate group as the significantly different group (sig.=0.015 when compared to the water only group).

Figure 4 – Average plant  $\delta^{15}\text{N}$  values after 8 weeks of different types of fertilizer treatment. Error Bars: +/- 1 SE



## Conclusion

The spatial pattern of the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotopes in the fecal samples did persist even after the baseline soil isotope averages were removed. Thus, the  $\delta^{13}\text{C}$  pattern does seem to be due to greater consumption of corn, a C4 plant. However, there are two possible explanations for the  $\delta^{15}\text{N}$  pattern: the squirrels are consuming more insects near the cornfield, or the corn itself is enriched in N-15 due to fertilizer application.

Animals preferentially excrete N-14, which results in an accumulation of N-15 as one moves up the trophic levels (DeNiro and Epstein, 1981). The spatial variation of  $\delta^{15}\text{N}$  values may thus be indicative of greater consumption of insects and other animal matter. Due to the effect that the ammonium nitrate fertilizer has on plant material, however, it seems more likely that the spatial pattern in the fecal  $\delta^{15}\text{N}$  values is due to N-15 enrichment in the corn due to fertilizer.

## Bibliography

Chacko, A. 2013. Spatial and seasonal variation in the stable isotopic composition of thirteen-lined ground squirrel fecal pellets as an estimate of variation in diet. Honors thesis, Andrews University, Berrien Springs.

DeNiro, M. J. and S. E Epstein. 1981. Influence of diet on the distribution of nitrogen isotopes in animals. *Geochimica et Cosmochimica Acta* 45:341-351.

Ehleringer, J. R., P. W. Rundel, and K. A. Nagy. 1986. Stable Isotopes in Physiological Ecology and Food Web Research. *Trends in Ecology & Evolution* 1:42-4