

# SOIL INCORPORATION OF COVERCROP BIOMASS: EFFECTS ON SOIL MICROORGANISMS AND NITROGEN LEVELS

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## Summary

**Biomass production, release of the usable nitrogen ions, ammonium and nitrate, and soil bacterial and fungal populations were measured for four covercrops after harvesting and soil incorporation. Two leguminous crops, Sunn hemp and pigeon pea, were studied as well as sorghum and marigold. As expected, the legumes provided the greatest amounts of soil nitrogen. Sorghum produced by far the greatest biomass and resulted in the largest soil microbial population increases. The results indicated that two to three weeks following the covercrop incorporation into soil is the most advantageous time to replant the next crop.**

## Introduction

Hawaii has a long history of plantation agriculture with year-round monocropping. These crops have sometimes been dependent on the use of soil fumigants and chemical fertilizers which resulted eventually in the reduction of natural soil biocontrol agents. In recent years, the large plantations have, in some areas, given way to many smaller farms and diverse crops and cropping systems. A project funded by the Sustainable Agriculture Research and Education program was installed to demonstrate to small farmers and to teach high school agriculture students the methods and advantages of using covercrops and alternate cropping to maintain soil fertility and healthy, natural soil microbial ecology without the use of expensive, toxic pesticides and chemical fertilizers.

Two farm sites were chosen for demonstration projects in Waialua, as well as the MOA (Mokichi Okada Association) farm located in Waimanalo. In order to learn more about the use of covercrops and to give additional information to the cooperators, data was collected from the Waimanalo farm site to determine the most advantageous time for replanting crops following incorporation of the covercrops included in the study. After harvesting and incorporation of the covercrops,

the plant material begins to be broken down by soil bacteria and fungi and nitrogen nutrient ions (ammonium and nitrates) are released. As the plant material becomes available for microbial colonization, the microbial populations increase and then slowly diminish again as the nutrients are consumed. Likewise, the released nitrogen compounds increase and then disappear from the soil as they are either taken up by weeds, another plant crop or immobilized into other compounds by microorganisms. To assist farmers in effectively making use of covercrops, the soil levels of bacteria, fungi and nitrogen compounds were measured for several covercrops over an eight-week period following covercrop harvest and incorporation. The optimum time for replanting the next crop would be when the nitrogen levels were at their highest.

## Materials and Methods

Field plots of two leguminous covercrops were observed: pigeon pea (*Cajanus cajan*) and Sunn hemp (*Crotalaria juncea*). The other two crops studied were marigold (*Tagetes erecta*) and a plot of alternating rows of sorghum (*Sorghum vulgare*) and velvet bean (*Mucuna pruriens*). Since it was not possible to carry out all four treatments at once, they proceeded sequentially from November, 2002 through September, 2003.

There were thus seasonal and temperature differences between the four treatments which could not be avoided. It is likely that microbial breakdown and nitrogen release

occurred more rapidly during warmer months, although no obvious difference was observed between treatments in this project.

Table 1. Activity Dates

<u>covercrop</u>	<u>harvest</u>	<u>incorporation</u>	<u>microbial counts</u>
pigeon pea	10/28/02	11/11/02	11/11 - 12/23/02
marigold	1/5/03	1/13/03	1/13 - 3/3/03
Sunn hemp	5/15/03	5/20/03	5/20 - 7/7/03
sorghum/velvet bean	7/15/03	7/28/03	7/28 - 9/23/03

Composite soil samples were taken in the test plots at harvest of each covercrop before and after incorporation. Additional samples were taken as close as possible to weekly intervals for four consecutive weeks after incorporation and again at six and eight weeks. In addition, samples were sent at the start of each test to the University of Hawaii Agricultural Diagnostic Service Center for analysis of soil pH, phosphorus, potassium, calcium, magnesium, ammonium, nitrate, and salinity.

Soil samples for microbial analysis were heat dried overnight and reweighed to determine moisture content. For each field sample, three subsamples were run. One gram soil (dry weight) of each subsample was placed in 100 ml sterile, distilled water and shaken for 30 min. After settling, a dilution series was made in sterile, distilled water. From each dilution sample, 3 petri plates for bacteria and 3 for fungi were inoculated with 0.5 ml of the suspension. It was determined that dilutions of  $1 \times 10^{-4}$ ,  $1 \times 10^{-5}$ , and  $1 \times 10^{-6}$  were appropriate for bacterial counts. For fungi,  $1 \times 10^{-3}$  and  $1 \times 10^{-4}$  were used. This gave a total of 9 replicate plates for each dilution. The numbers of bacterial and fungal colony-forming units (cfu) per gram soil were

estimated from the final plate counts. The selective medium for detection of bacteria was peptone-yeast extract agar, pH 7.2 with 50 ppm mycostatin (nystatin) added. For fungi, 10% V8 juice agar, pH 5.5 with 500 ppm streptomycin was used. The plates were incubated for 3 days before counting.

## Results

During the project, which continued from November 2002 until October 2003, soil bacteria, fungi, and N levels were taken for each of the four test plots. The field soil samples moisture levels varied from 12% to about 17%. Nutrient levels of phosphorus, potassium, calcium, magnesium and soil pH values were sufficient to high (see Table 2). The biomass harvested and incorporated from each of the crops is shown in Table 3. Sorghum provided by far the greatest dry weight of material followed by pigeon pea. Sunn hemp and marigold produced lower amounts. Daily rainfall and temperature measurements were recorded for the duration of the project. Significant rain occurred in January, February, and March of 2003, but the rest of the months were quite dry. Temperature highs were around 84° F - 90° F from May through October, 2003 and 70° F to

83° F from January through March 2003.

Soil nitrate ( $\text{NO}_3$ ) levels following the four covercrops are shown in Fig. 7. They were lowest for sorghum (2.5 to 8.0 ppm), higher for marigold (5.0 to 10.0 ppm), and highest for Sunn hemp (13 to 23 ppm). Soil ammonium ( $\text{NH}_4$ ) levels varied considerably between crops and varied in different patterns. Following sorghum, ammonium levels remained low (0 to 2.3 ppm). They were higher following pigeon pea (7.1 to 10.8 ppm). Following marigold, ammonium was 6.8 ppm at harvest and rose over the eight-week period to 18 ppm. In contrast, with Sunn hemp ammonium at harvest was high (17 ppm), but fell steeply after three weeks to no measurable amounts at six weeks. To summarize, sorghum and marigold covercrops produced relatively low  $\text{NO}_3$  levels when tilled into soil. The  $\text{NO}_3$  levels from the leguminous crops Sunn hemp and pigeon pea were higher, although pigeon pea fell off quickly. Sorghum produced low soil  $\text{NH}_4$ , while pigeon pea was somewhat higher. Sunn hemp produced a high level of  $\text{NH}_4$  that fell off after three weeks while the  $\text{NO}_3$  levels remained high through eight weeks. Marigold produced high  $\text{NH}_4$  levels that remained high for the eight weeks while the  $\text{NO}_3$  levels remained low. As expected, the leguminous crops, Sunn hemp and pigeon pea provided higher levels of nitrogen than the non-leguminous crops, except for the high  $\text{NH}_4$  levels with marigold. Sunn hemp is reported to produce 120 lb N per acre in just nine to 12 weeks (SARE Handbook 3).

The results of the microbial counts for each crop are shown in Figs. 1 through 4. Total soil bacterial populations (Fig. 5) varied between crops and over the sampling periods of each. The higher counts did not appear to

correlate with warmer times of the year, but with the crop. Marigold and Sunn hemp had relatively low counts, whereas pigeon pea was higher and the sorghum/velvet bean plots had very high counts. The latter two peaked soon after harvest with a second peak at 4 to 6 weeks, decreasing after 6 to 8 weeks. Soil bacterial populations did not appear to correlate with the soil nitrogen levels released, but instead, correlated positively with the crop biomass.

Soil fungal populations (Fig. 6) were lowest in the pigeon pea plot from harvest through three weeks, but then showed an increase at about four weeks post harvest. The marigold plot fungal levels peaked at one week post harvest and then fell to low levels. Sunn hemp and sorghum fungal levels rose rapidly following harvest and fell gradually with a second peak occurring at about four weeks. Sorghum/velvet bean resulted in the highest soil bacterial and fungal populations overall of any of the four crops. Fungal levels correlated with crop biomass except in the case of pigeon pea. With pigeon pea, fungal levels were initially relatively low, but showed a large peak at four weeks. This may indicate that pigeon pea biomass contains relatively large proportions of compounds resistant to fungal degradation or that a fungal inhibitor is initially present.

A possible explanation for the existence of two microbial population peaks was suggested by Dr. Mituku Habte of the University of Hawaii. The first population increase may represent microorganisms that are consuming and multiplying on readily available organic compounds; carbohydrates and amino acids. The second peak could be composed of other microbial species that decompose more resistant compounds.

**Table 2. Field Plot Nutrient Analyses Preharvest**  
(+ sufficient, ++ high, +++ very high)

	Plot A6 Pigeon Pea	Plot A8 Marigold	Plot A1 Sunn hemp	Plot A4 Sorghum
pH	7.0 +	6.9 ++	6.8 ++	7.3 ++
ppm P	545 +++	222 +++	480 ++	66 ++
ppm K	496 ++	350 ++	480 ++	270 +
ppm Mg	----	1350 ++	1310 ++	1112 ++
salinity EC	----	0.27 +	0.41 +	0.36 +

**Table 3. Covercrop Harvested Weights (lb/acre)**

	Plot A6 Pigeon Pea	Plot A8 Marigold	Plot A1 Sunn hemp
fresh weight	25,694	19,788	11,943
dry weight	9,068	3,997 (est.)	4,942
	Plot A7 Sorghum	Plot A7 Velvet Bean	Plot A7 Total
fresh weight	100,858	7,984	108,842
dry weight	33,263	1,797	35,060

**Discussion**

Nitrogen in plant tissues exists in complex organic compounds, proteins and chlorophyll. However, it must be present as ammonium or nitrate ions in soil in order for plants to absorb it. Soil microorganisms are essential for the mineralization of organic compounds into the simpler N compounds.

Soil fungal counts did not correlate with the NH<sub>4</sub> or NO<sub>3</sub> levels, but appeared to correlate with the amount of biomass produced by each crop (see Table 3). Both

sorghum and Sunn hemp had high initial fungal counts. In all crops, the fungal populations peaked at one to two weeks and then declined (except marigold) with a second peak at four weeks.

Soil bacterial counts correlated negatively with NH<sub>4</sub> levels. Marigold and Sunn hemp, with high soil NH<sub>4</sub> levels had low bacterial populations following tillage. Sorghum bacterial counts were high, while pigeon pea was moderate. Pigeon pea contributes both organic matter and N to soil,

although the biomass production is much less than Sunn hemp and sorghum (University of Hawaii website). Despite its high biomass production, sorghum contributed low amounts of N to the soil (see Fig. 9).

With some variations, the soil microorganism populations following covercrop incorporation increased and then began to decrease after one to two weeks. Ammonium levels followed a similar trend except for marigold that remained high. Nitrate levels remained about the same for each crop over the eight-week period except for pigeon pea which peaked at one week and then fell. Upon biomass incorporation into soil, microorganisms begin to metabolize the available carbon and nitrogen compounds and populations increased. After the easily metabolized compounds are degraded, the populations begin to decline. As a predictor of the most favorable time to replant a crop following covercrop incorporation, the point at which microbial populations are just beginning to decline would be the time when the most  $\text{NO}_3$  and  $\text{NH}_4$  are available in the soil. In this project, the most favorable replant time appeared to be between two to three weeks after covercrop incorporation. Hue and Silva (2000) observed that ammonium released from a legume covercrop (*Leucaena*) soil amendment was highest between two to four weeks after application and remained low between six to ten weeks. After ten weeks, the  $\text{NH}_4$  in soil began to increase again for as long as 16 weeks. For chicken manure in the same trial, there was no such second release pattern and soil N remained low after four weeks. The authors attributed this second  $\text{NH}_4$  release from the covercrop to the degradation of nitrogen

compounds that were initially resistant to microbial action. Since our trials ended at eight weeks, we did not observe the later N release. This pattern of release from leguminous covercrops is especially beneficial for the subsequent crop since older, fruiting plants require much greater nitrogen inputs than the seedling plants.

Covercrops provide other advantages besides nitrogen nutrients. Marigold is known to reduce harmful nematodes. Soil organic matter is important for plant growth and soil texture. Some covercrops such as buckwheat, vetch and others, help to retain and scavenge soil phosphorus. Deep rooted covercrops help bring up leached calcium and potassium into the upper soil layers. In addition, covercrops control weeds and soil erosion.

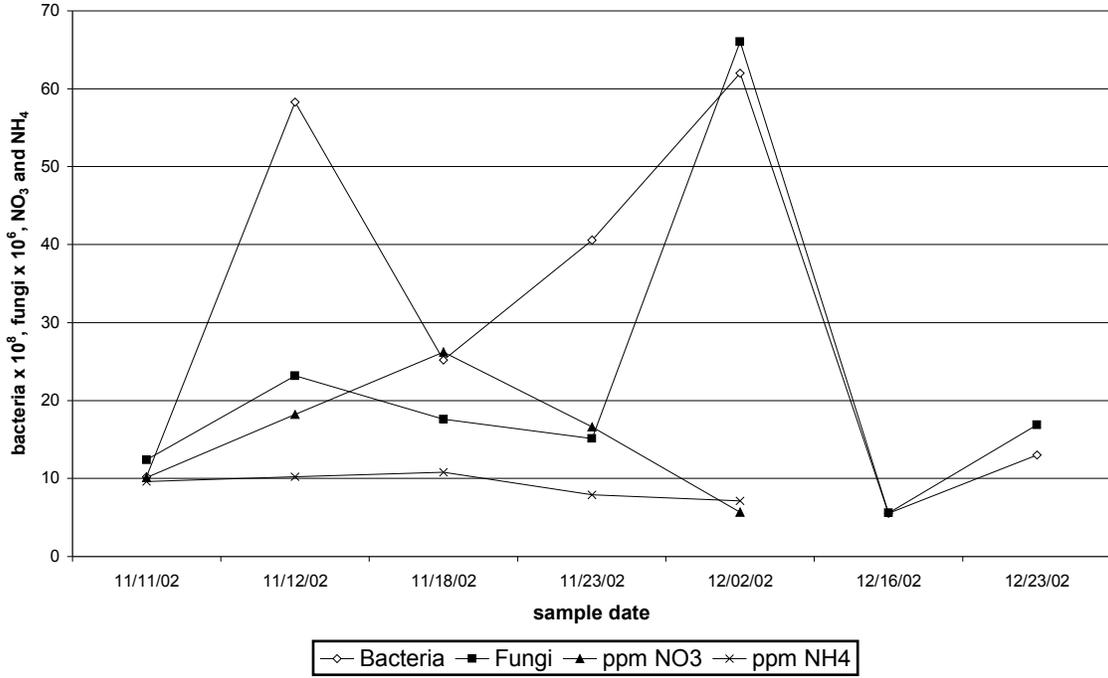
### References

Hue, N.V. and J.A. Silva. 2000. Organic soil amendments for sustainable agriculture: organic sources of nitrogen, phosphorus, and potassium. pp 133-144 *in* Plant Nutrient Management in Hawaii's Soils. J.A. Silva and R.S. Uchida, Eds. CTAHR, University of Hawaii.

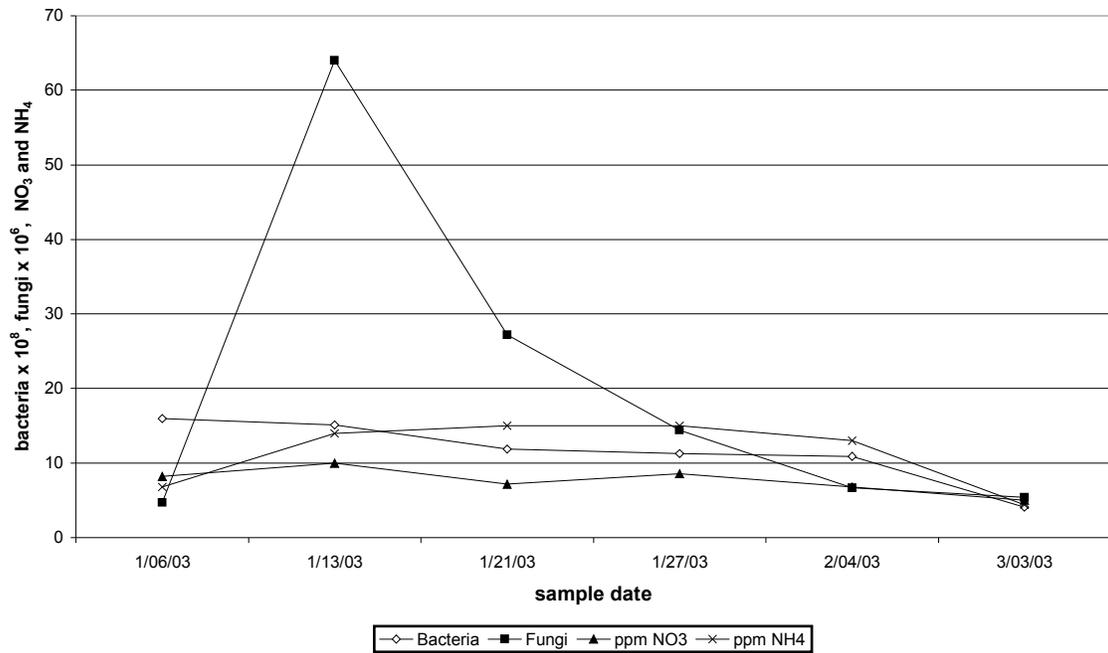
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<http://www2.ctahr.hawaii.edu/sustainag/pigeonpea.htm>.

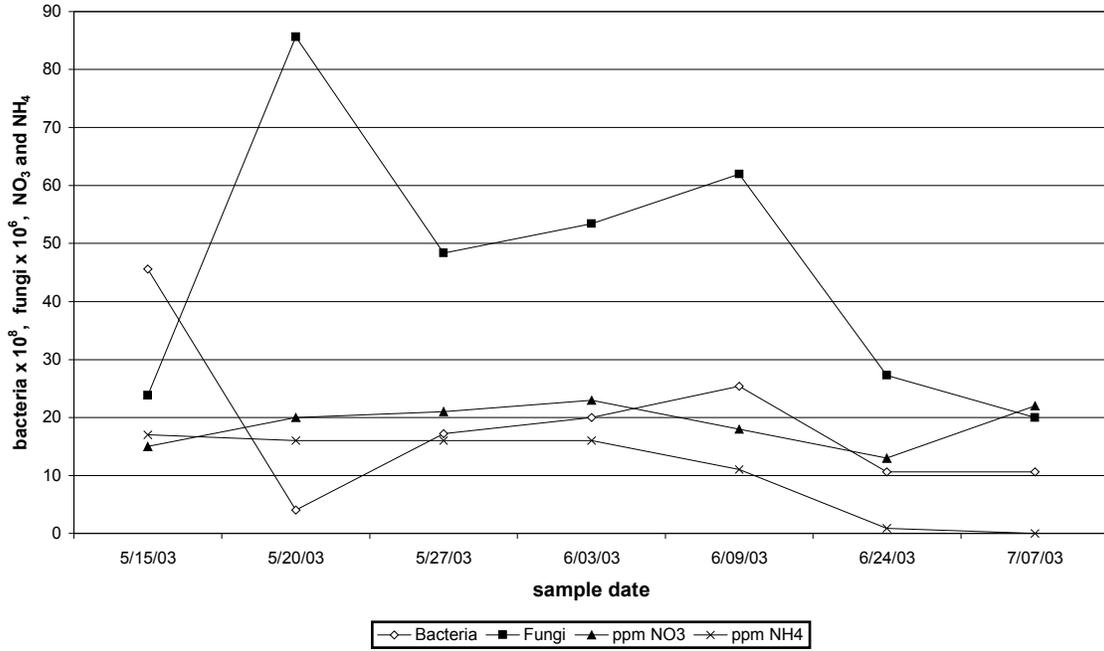
**Fig. 1. Pigeon Pea Bacteria and Fungi (per g Soil) and Soil N Levels (ppm)**



**Fig. 2. Marigold Bacteria and Fungi (per g Soil) and Soil N Levels (ppm)**



**Fig. 3. Sunn Hemp Bacteria and Fungi (per g Soil) and Soil N Levels (ppm)**



**Fig. 4. Sorghum / Velvet Bean Bacteria and Fungi (per g Soil) and Soil N Levels (ppm)**

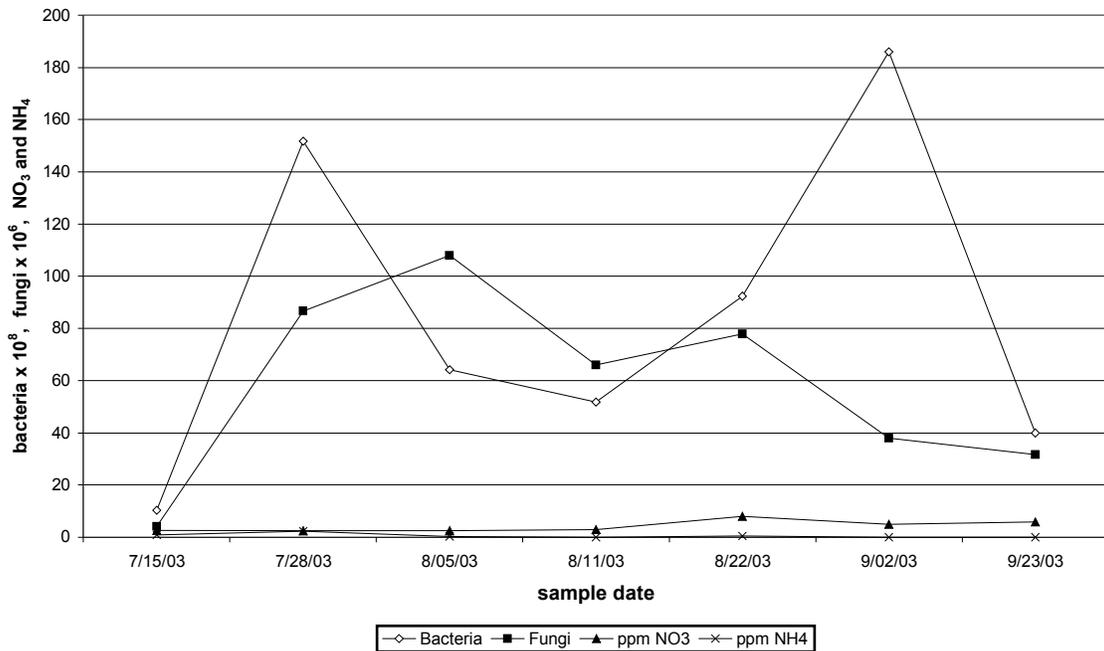


Fig. 5. Soil Bacteria Following Covercrop Incorporation

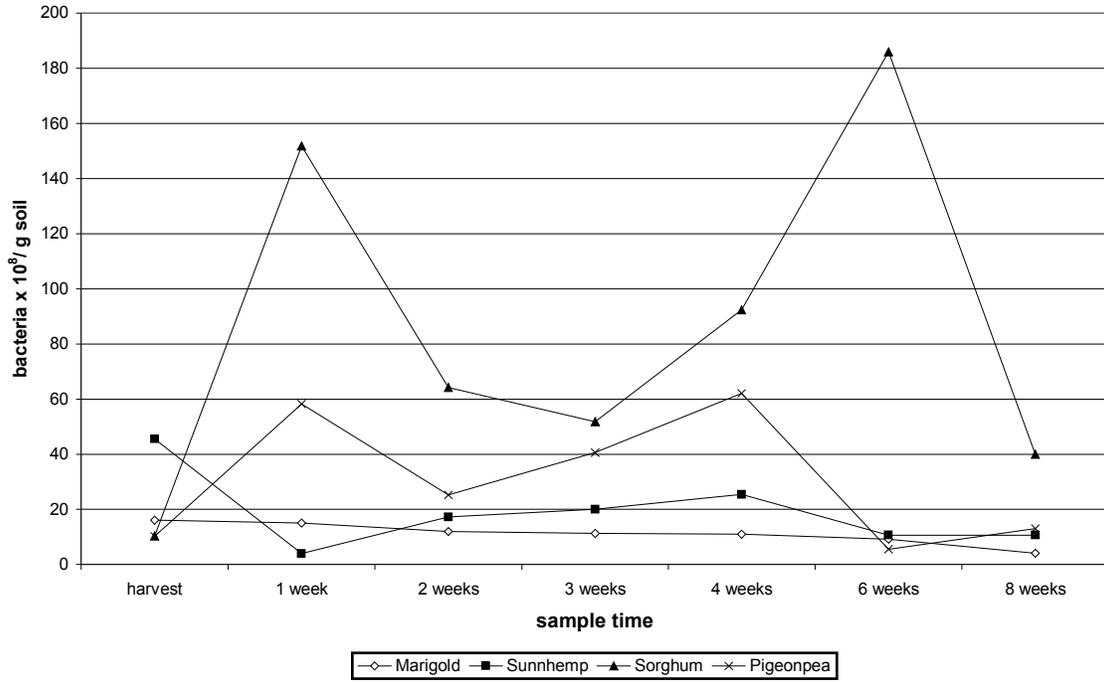


Fig. 6. Soil Fungi Following Covercrop Incorporation

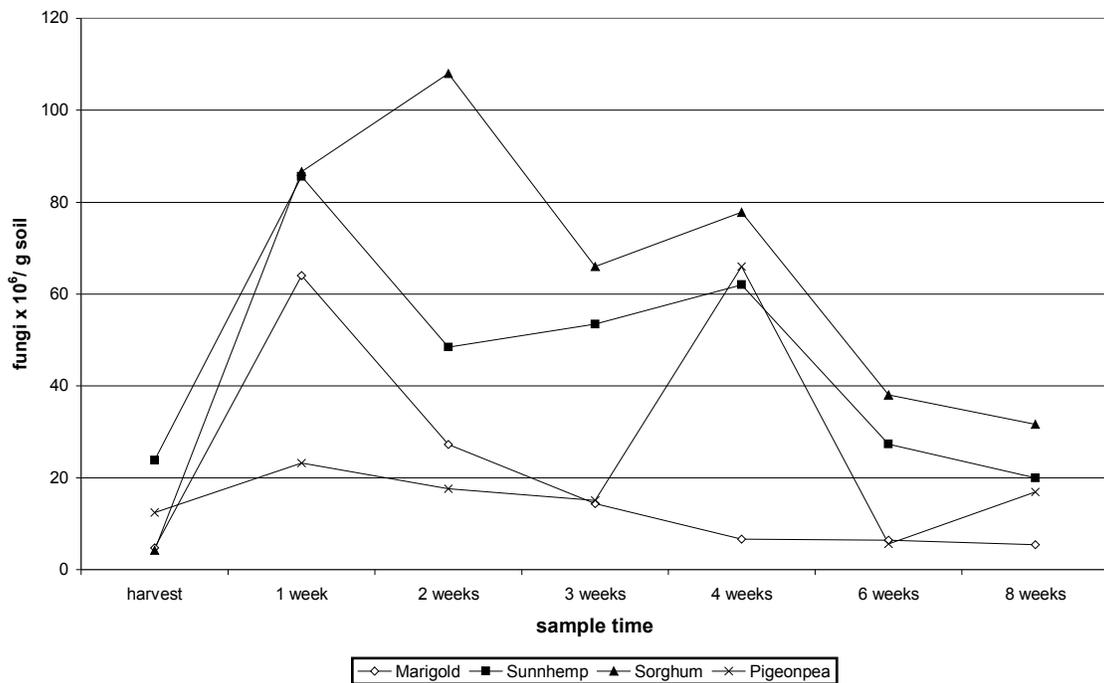


Fig. 7. Soil Nitrate Levels Following Covercrop Incorporation

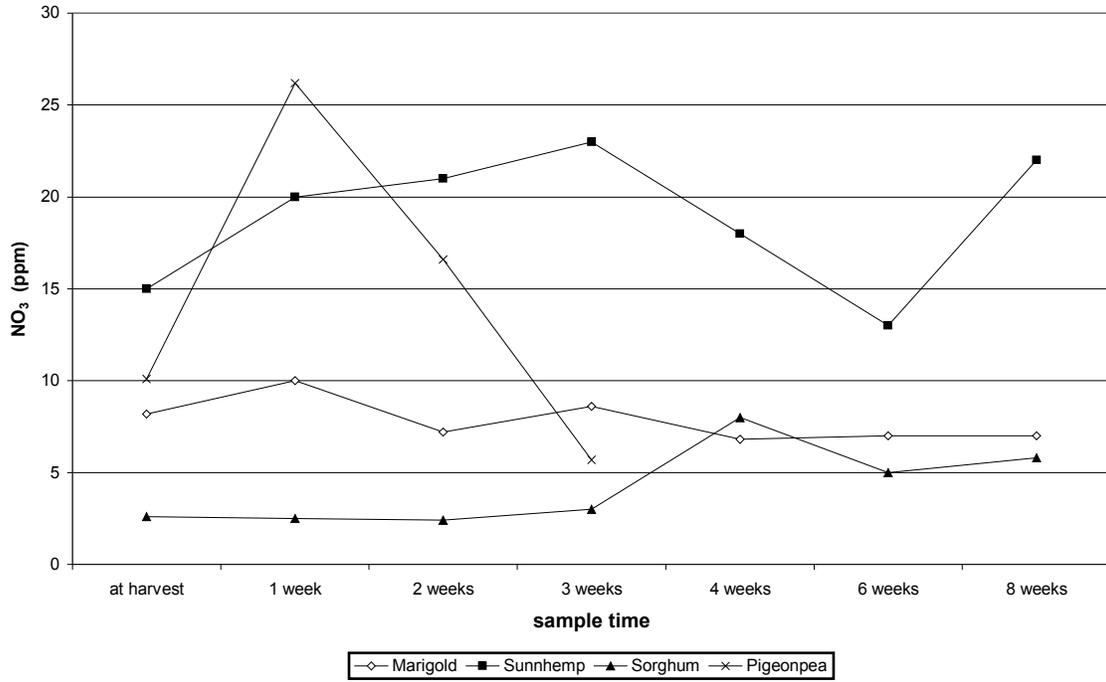


Fig. 8. Soil Ammonium Levels Following Covercrop Incorporation

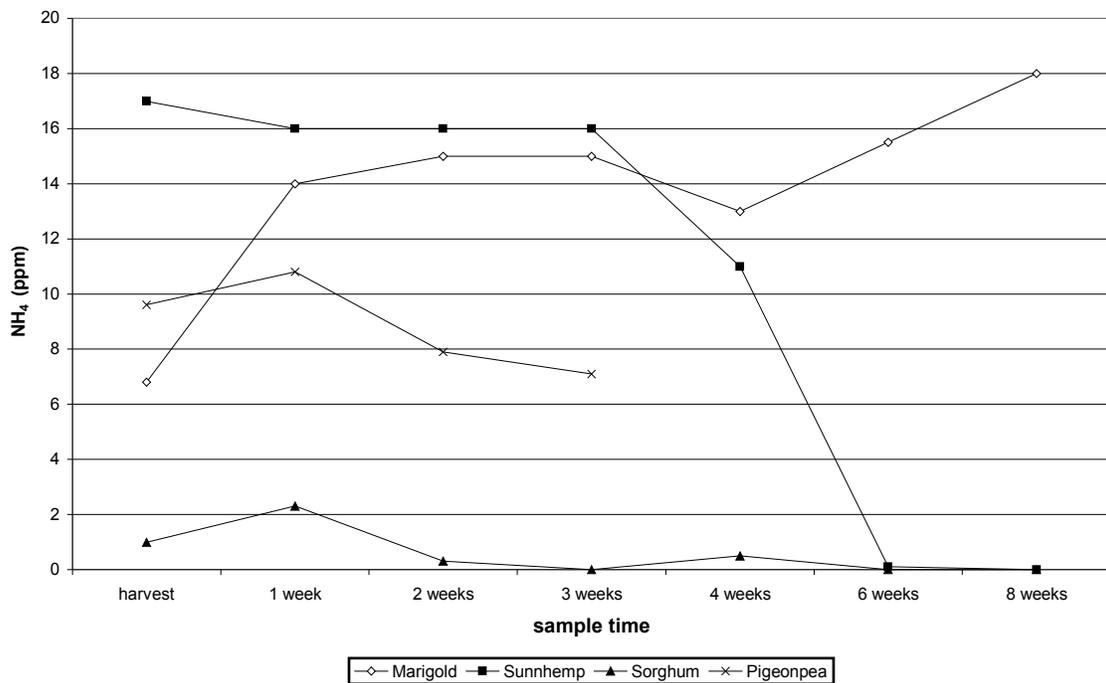


Fig. 9. Total Soil N Levels Following Covercrop Incorporation

