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Ap-42 #24

# The Influence of Plant Residues on Denitrification Rates in Conventional and Zero Tilled Soils[1]

M.S. AULAKH, D.A. RENNIE, AND E.A. PAUL[2]

## ABSTRACT

**A field study was conducted with treatments consisting of a factorial combination of N (0 or 100 kg N ha$^{-1}$ as $(NH_4)_2SO_4$, straw (0 or 3000 kg ha$^{-1}$), and two tillage treatments. Ground straw was mixed with the plow layer of soil in the conventional till (CT) plots and chopped straw was spread over the surface of the zero till (ZT) plots. Wheat (*Triticum aestivum* L.) was grown as the test crop. Gaseous losses of N were measured using the acetylene inhibition-soil core technique and compared with loss estimates obtained from the imbalance in the N budget of $^{15}$N-treated microplots located within the larger yield plots. When adequate inorganic N was present, the incorporation of straw in CT soil or the application of straw on the surface of ZT soil approximately doubled the accumulative gaseous N losses. The straw apparently increased the supply of energy material available to denitrifying organisms, and also increased surface soil moisture content (particularly during the month of June). This further stimulated denitrification in ZT soil. Unaccounted $^{15}$N on the fertilizer N balance studies agreed closely with cumulative N losses using the acetylene inhibition technique.**

***Additional Index Words:*** **wheat grain yield, protein content, N uptake, soil moisture, mineral N.**

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RECENT field studies using the acetylene inhibition technique have demonstrated that the moisture content of surface soil is a singlular environmental variable affecting gaseous losses of N; even rather small increases in volumetric soil moisture can be accompanied by very large increases in the rate of denitrification (3, 4). It was also found that gaseous N losses were 2 to 3 times higher from zero till (ZT) than conventional till (CT) soils (4). However, multiple regression analyses showed that such factors as moisture, temperature, inorganic $NO_3^- - N$ and $NH_4^+ - N$ accounted for only 37 to 66% of the variations in rate of denitrification. Soil bulk density also was shown to be of importance but no quantitative assessment of its significance was possible (1, 5).

Crop residue placement is a significant environmental difference between CT and ZT soils. In CT, crop residues are frequently worked uniformly into the surface soil, while in ZT they are left scattered on the surface or as standing stubble. There is reason to believe that the difference in crop residue placement may influence denitrification rates. For instance, the addition of a readily decomposable carbon material such as manure greatly enhanced denitrification (relatively high in N) in both laboratory and field studies (9, 15). Other studies have shown that the addition of wheat straw to undisturbed soil cores (7) or field plots (8) reduced N losses by denitrification due to increased immobilization of inorganic N. Aulakh et al. (1, 2, 4) have shown that rates of denitrification are only slightly or not at all influenced by rather wide variations in nitrate-N concentrations in the soil, and accordingly, immobilization may not be a significant factor influencing denitrification rates unless it is relatively complete.

The acetylene inhibition technique (17) developed as a means of assessing gaseous N losses has been used successfully in recent laboratory and field studies (1, 2, 3, 4, 14, 16). This method has been validated by $^{15}$N measurements in anaerobic soil systems (12) and by $^{13}$N measurements in a soil-water slurry (16) under laboratory conditions. In a recent study (2) we carried out its validation under field conditions by comparing the cumulative gaseous N losses with the amount of unaccounted fertilizer N in $^{15}$N balance studies; the two techniques agreed very closely, although the losses were relatively low (1–2 kg N ha$^{-1}$). The present field study further confirms the close agreement between these two techniques when losses were as high as 25 kg N ha$^{-1}$. This paper also documents the influence of decomposing crop residues on denitrification rates in the presence and absence of applied N under field conditions.

## MATERIALS AND METHODS

The experiment was established in the spring of 1981 on an Elstow clay loam, a Dark Brown Chernozemic soil (Typic Haploborolls). Some of the important soil characteristics and agronomic history of the CT and ZT fields (which have been under these tillage treatments since 1978) were reported earlier (1,4).

The treatments consisted of a factorial combination of two levels each of N [0 or 100 kg N ha$^{-1}$ as $(NH_4)_2SO_4$] and wheat straw (0 or 3000 kg ha$^{-1}$) replicated twice in individual 2 × 2 m macroplots in both the CT and ZT fields. Hard red spring wheat (*Triticum aestivum* L. var. Neepawa) was used as the test crop. After seeding the wheat in the ZT plots water with the required amount of fertilizer N was sprayed uniformly on the soil surface using a back-pack sprayer. To minimize $NH_3$ volatilization, some additional water was added to move the fertilizer N a few centimeters into the plow layer. The required amount of chopped wheat straw (3–5 cm long) was uniformly spread over the surface and the macroplots were covered with steel netting (chicken wire) to minimize straw losses by wind erosion. The netting was removed after four weeks when the crop was well established. To obtain uniform mixing and make the applied straw more effective, it was finely ground and mixed with the plow layer of CT plots before seeding wheat. Then fertilizer N was applied as in ZT plots using the same amount of water in all the plots.

Four microplots were established in both CT and ZT fields by inserting an open aluminum cylinder (20 cm i.d. by 45 cm long) in each N-fertilized macroplot, taking special care to prevent soil compaction. Each microplot treatment was prepared in a manner similar to the macroplot which sur-

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Table 1—The effect of various N and straw treatments on the wheat grain yield, protein and plant N uptake under conventional- and zero-till systems.

| Treatment | Grain yield, kg ha$^{-1}$ | Grain protein, % | Total N uptake kg ha$^{-1}$ |
|---|---|---|---|
| | Conventional-till | | |
| Control | 1681c§ | 12.6b | 73.4c |
| Straw† | 811a | 10.7a | 37.0a |
| $N_{100}$‡ | 1972d | 15.9de | 105.4e |
| | (1954)¶ | (16.5) | (117.2) |
| $N_{100}$ + straw | 1947d | 16.6e | 105.4e |
| | (1910) | (18.1) | (116.7) |
| | Zero-till | | |
| Control | 1468b | 12.0b | 62.1b |
| Straw | 1697c | 11.1a | 66.6b |
| $N_{100}$ | 2009d | 15.1cd | 96.4d |
| | (2098) | (16.2) | (108.4) |
| $N_{100}$ + straw | 2400e | 14.9c | 119.1f |
| | (2555) | (13.3) | (116.3) |

† Straw = 3000 kg ha$^{-1}$.
‡ $N_{100}$ = 100 kg ha$^{-1}$.
§ Values in the same column are significantly different at $P \leq 0.05$ (using Duncan's multiple range test) when not followed by the same letter.
¶ Figures in parenthesis represent values obtained from $^{15}$N-microplots.

rounded it except that $^{15}$N-labelled $(NH_4)_2SO_4$ (5.4% excess) was used as the source of fertilizer N.

Gaseous N losses were measured during the growing season (5 June–4 Sept. 1981) by removing three pairs of undisturbed soil cores from each treatment every week. The reader is referred to our earlier publication (1) for procedures used to measure: gaseous N losses with the acetylene inhibition technique, soil moisture, and inorganic soil N ($NO_3^-$ + $NO_2^-$, $NH_4^+$). The cores were incubated 24 h in 1100 cm$^3$ glass jars with airtight covers fitted with serum stoppers. Incubation under shaded conditions in soil outside the laboratory closely approximated the temperature within the canopy of grain crops in the field. Oxygen contents in the jars, monitored at various times with a gas chromatograph, remained above 19% during most of the incubations, but dropped as low as 15% on straw-amended samples with high soil moisture. Rainfall data were recorded at a meteorological station on the site.

At the end of the growing season, 1 m$^2$ area from each macroplot and all plants within each cylinder were harvested, dried at 60 ± 1°C, separated into grain and straw and weighed. The cylinders including the soil were removed, and representative soil samples were taken from the 45 to 60-cm depth. In the laboratory, the cylinders were cut open and the soil was sectioned into 0- to 15-, 15- to 30-, and 30- to 45 cm depths, each of which was weighed and air-dried. Roots plus crowns were collected, washed, dried, and weighed. Soil subsamples, ground to pass a 10-mesh (2-mm) sieve were extracted with 2*M* KCl for mineral-N analysis. Additional 100-mesh samples were digested for total Kjeldahl N analysis. The nonexchangeable $NH_4^+$ (commonly known as fixed $NH_4^+$) in the 0 to 15- and 15- to 30-cm soil samples was determined (10). Wheat grain, straw, and root + crown fractions were ground to pass a 100-mesh sieve, and assayed for total N. Kjeldahl digestion and distillation methods described by Rennie and Paul (13) were followed. Soil organic N was calculated by subtracting mineral N from total Kjeldahl N. Grain protein was estimated as 5.7 × percentage of N and expressed on a 13.5% moisture basis.

## RESULTS AND DISCUSSION

### Grain Yield, Protein Content, and Plant N Uptake

In the CT field, wheat grain yield was increased by 17% by the addition of fertilizer N, mixing 3000 kg wheat straw into the plow layer resulted in a 50% yield reduction, probably due to immobilization of mineral N (Table 1). When the N and straw were both applied, there was sufficient N for both immobilization and plant growth, and the grain yields were equal to those obtained with N alone.

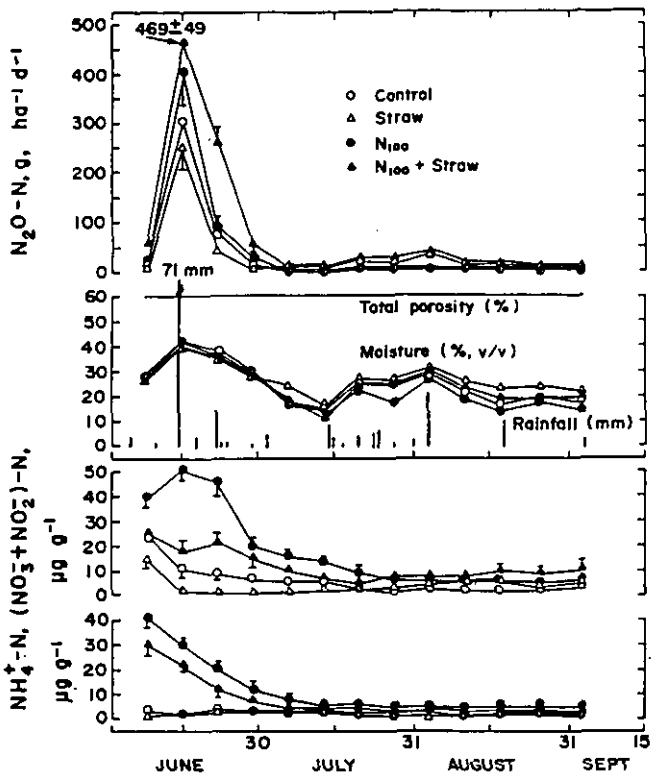


Fig. 1—Gaseous N losses ($N_2O$–N), soil moisture, rainfall, ($NO_3^-$ + $NO_2^-$)−N and $NH_4^+$−N of conventional-tilled Elstow clay loam seeded to wheat with various straw and N treatments. Φ indicates standard deviation within three replicates.

Grain yield from the ZT control was lower than from the CT control plot, but increased significantly with applied N. In contrast to the CT field, grain yield was increased 16% by the addition of straw, probably because surface placement of the straw provided a thick continuous layer of mulch that conserved a greater amount of rain water without immobilizing all available mineral N. The highest yield (63% above the control) was obtained from the ZT plots where N was applied in conjunction with the straw mulch surface. Nitrogen uptake by wheat (grain plus straw), followed the same pattern as grain yield.

The addition of straw without N lowered wheat grain protein content by 20% in both tillage systems. This can be attributed to immobilization of N in the CT plots and simple N dilution frequently associated with large yield increases due to more favorable moisture conditions in the ZT plots. The relatively high protein levels achieved with the $N_{100}$ and $N_{100}$ plus straw treatments for both cropping systems suggest that available N was more than adequate in these treatments.

Although the ZT control had a higher soil moisture regime than the CT control (Fig. 1 and 2), its grain yield, protein content, and N uptake were all lower. The high grain yield and N uptake obtained with the application of 100 kg N ha$^{-1}$ in conjunction with a

straw mulch emphasizes the need to ensure optimum fertility in ZT studies; otherwise, the greater moisture conserving characteristics of the surface straw mulch may not be reflected in greater production (6, 11). The yields obtained from $^{15}N$-microplots were, in general, comparable to those from corresponding macroplots (Table 1), thus confirming that realistic field conditions were achieved in the microplots.

### Recovery of Fertilizer N

As has already been suggested from the total N uptake data given in Table 1, plant uptake of fertilizer N was not significantly influenced by the incorporation of crop residues on the CT plots (Fig. 3); about 40% of the added fertilizer N was taken up by the crop. The greater fertilizer N uptake by the crop on the straw mulched ZT plots is a reflection of the more favorable growing conditions, especially the higher moisture level.

The amount of fertilizer N immobilized (organic N) or present in the inorganic form (extractable mineral N plus fixed $NH_4^+$) in the 0- to 60-cm profile varied rather widely between treatments (Fig. 3). A greater amount of fertilizer N was immobilized in the CT soil when straw was incorporated. This difference was partially offset by a lower content of inorganic N and the recovery of fertilizer N from these two CT treatments was quite similar. In contrast, while the amounts of fertilizer N immobilized in the no-straw treatments on CT and ZT soils were very similar, much less fertilizer N was immobilized on the ZT-straw treatment than on the CT-straw treatment. This could be attributed to higher plant uptake and denitrification which competed with immobilization for fertilizer N. In addition, washing the fertilizer N a few centimeters into the soil (see Materials and Methods) may have separated the fertilizer N from surface-placed straw enough to partially inhibit its immobilization. Only a very small portion (0.5–2%) of the fertilizer N was recovered as fixed ammonium.

Most of the fertilizer N recovered from the soil was found in the 0- to 30-cm depth (data not presented). However, management did influence the amount of fertilizer N found below the 30-cm depth; 1.5 and 1% of the applied fertilizer N was recovered in the 30- to 60-cm depth on the no straw and straw-CT treatment. In contrast, 6.6 and 3.6% were recovered in the no straw and straw-ZT treatments. Only trace amounts of the fertilizer N were found below 45 cm. It was therefore assumed that most of the unaccounted N was lost through denitrification. It is unlikely that significant amounts of the fertilizer were lost by the volatilization in light of the application procedure used. The $^{15}N$ data (Fig. 3) show that denitrification was stimulated by the addition of straw, whether fully incorporated or left on the surface. On ZT soil, surface placement of the crop residue resulted in a twofold increase in unaccounted N compared to the no-straw treatment.

### Gaseous Losses of N

Gaseous losses of N measured by the acetylene inhibition technique ($N_2O$-N g ha$^{-1}$ day$^{-1}$) followed similar patterns under both CT and ZT tillage systems (Fig. 1 and 2). The relatively high rates of $N_2O$ evolution that prevailed during June were related to heavy mid-June rainfall. During this period the unfertilized ZT-straw treatment lost N at a higher rate than the

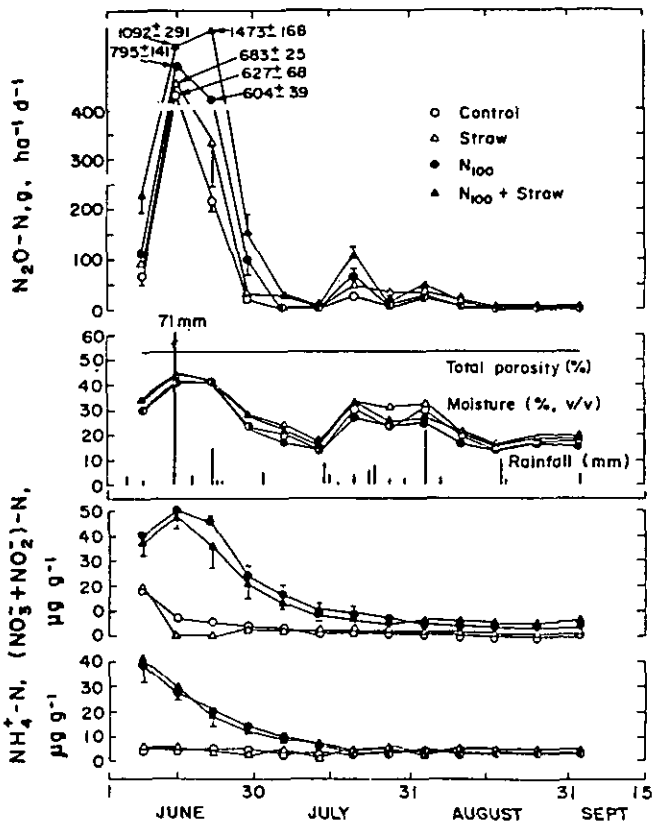


Fig. 2—Gaseous N losses ($N_2O-N$), soil moisture, rainfall, ($NO_3^- + NO_2^-$)−N and $NH_4^+$−N of zero-tilled Elstow clay loam seeded to wheat with various straw and N treatments. Φ indicates standard deviation within three replicates.

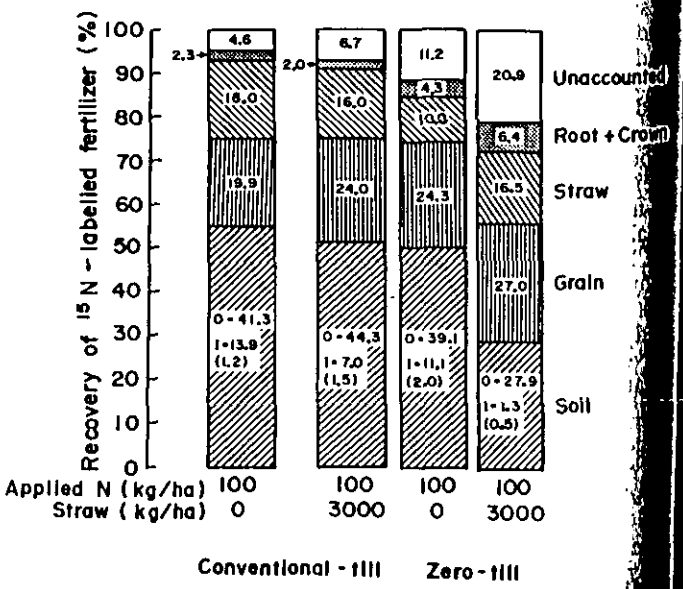


Fig. 3—Balance sheet of $^{15}N$-labelled fertilizer nitrogen. O = organic N, I = inorganic N including amount of fixed $NH_4^+$ (given in parentheses).

ZT control, but the relationship between these two treatments on CT soil was reversed. This is the only indication in these data that the addition of straw caused a decrease in denitrification rates compared to the controls.

The inorganic N data given in Fig. 1 show that incorporation of 3000 kg ground wheat straw immobilized mineral N rapidly and reduced gaseous N losses early in the growing season. However, at later stages of plant growth when levels of mineral N were very low in both untreated and straw treated plots (Fig. 1), a higher rate of $N_2O$ emission was recorded from the straw treatment possibly due to a 2 to 4% higher soil moisture. The higher soil moisture may have resulted from lower water use by the poor crop on this treatment. Therefore, the cumulative losses of N were similar for both the untreated and straw-treated plots (Table 2). In a one-week incubation study, Craswell (7) noted rapid immobilization of labelled $NO_3^-$ and reduced denitrification losses when added straw was added to small soil cores. Our results confirm this observation, but also indicate that in the field the reduced denitrification losses are later offset by higher gaseous N losses as water accumulates due to poor plant growth. The much higher rates of $N_2O$ evolution from ZT than CT soil during the month of June produced a twofold difference in the total amount of N lost from the no-straw treatments and a threefold difference for the straw treatments (Table 2). The more favorable moisture conditions prevailing on the ZT-straw treatments is probably a major contributing factor to the higher $N_2O$ evolution rates and results in rather high accumulative N losses. Higher $NO_3^- - N$ levels in the soil on both $N_{100}$ treatments also encouraged higher rates of denitrification.

The data for cumulative gaseous N losses measured with acetylene inhibition techniques were in close agreement with N unaccounted for in the $^{15}N$ balance studies (Table 2). The $^{15}N$ technique measures only the fate of fertilizer N. The $C_2H_2$ inhibition method should reflect denitrification of both the fertilizer and the soil N. Denitrification activity was at a maximum in June shortly after fertilizer N was added and it is probably safe to assume that the majority of the $N_2O$ measured using the acetylene inhibition technique was derived from this source. Thus it has confirmed our earlier study (2) that these two different approaches of assessing gaseous N losses give quite similar results under field conditions.

## CONCLUSIONS

This study, using the acetylene inhibition technique to assess denitrification rates, has shown that the addition of wheat straw residues, either as a mulch on the surface or fully incorporated, resulted in a doubling of the accumulative gaseous N losses over the growing season. It is significant that fertilizer N balance studies provided an estimate of unaccountable $^{15}N$ which coincided very closely with that obtained using the acetylene reduction technique.

The $^{15}N$ balance approach permitted the measurement of the distribution of fertilizer N in the plant and soil at the end of the growing season. Under the conditions prevailing during this field study, immobilization of the added $^{15}N$ was significantly higher under conventional till than zero till. It can therefore be concluded that under conventional till where the straw facilitated the fertilizer N immobilization, it also increased the supply of energy material available to the denitrifying organisms and thus denitrification rates for the growing season increased sharply. Denitrification would appear to have been increased under zero till primarily because of the more favorable moisture conditions. Thus this study has clearly shown that the use of both the $^{15}N$ balance study and the acetylene reduction technique has materially added to our understanding of factors influencing denitrification under field conditions, and the influence of management on these rates.

**Table 2—Cumulative gaseous N losses (kg N ha$^{-1}$) measured with acetylene inhibition technique (AIT) and $^{15}N$-balance approach.**

| | Conventional-till | | Zero-till | |
|---|---|---|---|---|
| Treatment | AIT | $^{15}N$ | AIT | $^{15}N$ |
| Control | 3.20 ± 0.42a§ | – | 7.25 ± 0.48b | – |
| Straw† | 3.41 ± 0.44a | – | 9.85 ± 0.23c | – |
| $N_{100}$‡ | 4.28 ± 0.51a | 4.62 ± 1.08a | 11.88 ± 0.43d | 11.20 ± 3.78d |
| $N_{100}$ + straw | 7.25 ± 0.43b | 6.65 ± 1.41b | 22.51 ± 2.24e | 20.87 ± 4.32e |

† Straw = 3000 kg ha$^{-1}$.
‡ $N_{100}$ = 100 kg N ha$^{-1}$.
§ Values are significantly different at $P \leq 0.05$ (using Duncan's multiple range test) when not followed by the same letter.

Absolute validation of either the acetylene inhibition methods or the $^{15}N$ technique is difficult under field conditions. The acetylene method requires addition of an external gas and enclosure in some type of chamber. Sampling to ensure that all denitrification events are recorded could involve an even more intense program than that carried out in this study. The $^{15}N$ technique involves problems with uniform distribution of tracer, possible alterations of the size or distribution of the potentially denitrifiable N pool, and the uncertainties associated with possible leaching or volatilization. These losses were minimized in this study as the major denitrification events occurred in June before there was opportunity for substantial alteration of the soil N pool by mineralization.

The fact that the two methods gave very similar results in a number of different treatments is encouraging but must be considered somewhat coincidental. However, it is of interest that the decrease measured in $^{15}N$ immobilization coincided with a doubling of gaseous N losses as measured by both the $^{15}N$ balance and the summation of $C_2H_2$ inhibition studies conducted weekly. The acetylene inhibition method makes it possible to relate the dynamics of N loss to short-term environmental changes such as rainfall events, soil type management such as ZT or straw incorporation and crop development. The $^{15}N$ technique involves a balance sheet approach that does not distinguish the source of loss but provides an integrated measurement of the fate of added N.

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# Aerobic and Anaerobic Microbial Populations in No-till and Plowed Soils[1]

D. M. LINN AND J. W. DORAN[2]

## ABSTRACT

**Surface soils from long-term tillage comparison experiments at six U.S. locations were characterized for aerobic and anaerobic microbial populations and denitrification potential using an in situ acetylene blockage technique. Measurements of soil water content, bulk density, and relative differences in pH, $NO_3^-$-N, water-soluble C, and total C and N contents between tillage treatments were also determined at the time of sampling. Numbers of aerobic and anaerobic microorganisms in surface (0–75 mm) no-till soils averaged 1.35 to 1.41 and 1.27 to 1.31 times greater, respectively, than in surface-plowed soils. Bulk density, volumetric water content, water-filled pore space, and water-soluble C and organic C and N values were similarly greater for surface no-till soils compared to conventionally tilled soils. Deeper in the soil (75–300 mm), however, aerobic microbial populations were significantly greater in conventionally tilled soils. In contrast, below 150 mm, the numbers of anaerobic microorganisms differed little between tillage treatments. In no-till soils, however, these organisms were found to comprise a greater proportion of the total bacterial population than in conventionally tilled soils. Measurements of the denitrification potential from soils at three locations generally followed the observed differences in anaerobic microbial populations. Denitrifying activity, after irrigation with 15 mm of water, was substantially greater in surface 0- to 75-mm no-till soils than in conventionally tilled soils at all locations. At the 75- to 150-mm soil depth, however, the denitrification potential in conventionally tilled soils was the same or higher than that of no-till soils. In surface no-till soils, increased numbers of anaerobic microorganisms and a substantially greater denitrification potential, following irrigation, indicate the presence of less-aerobic conditions in comparison to conventionally tilled soils. This condition appears to result from greater soil bulk densities and/or water contents of no-till soils, which act to increase water-filled porosity and the potential for water to act as a barrier to the diffusion of oxygen through the soil profile.**

***Additional Index Words:*** **Soil aeration, denitrification, conservation tillage, microbial ecology.**

Linn, D. M., and J. W. Doran. 1984. Aerobic and anaerobic microbial populations in no-till and plowed soils. Soil Sci. Soc. Am. J. 48:794–799.

THE NUMBERS, types, and activities of soil microorganisms are important to the productivity of soil through their regulatory effect on soil carbon (C) and nitrogen (N) levels. Several early studies comparing the microbial populations of reduced-tillage and plowed soils demonstrated that greater numbers of aerobic microorganisms were often associated with the surface 0 to 150 mm of reduced-tillage soils (Dawson et al., 1948; Gamble et al., 1952; Schaller and Evans, 1954). More recently, Barber and Standell (1977) and Doran (1980b) found that microbial numbers, dehydrogenase activities, and respiration rates in surface no-till soils were significantly greater than those in plowed soils. However, at a soil depth below 50 to 75 mm, these indexes of microbial activity were often

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