Changes in microbial biomass and community composition, and soil carbon and nitrogen pools after incorporation of rye into three California agricultural soils

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Abstract

The effects of long-term agricultural management on active soil organic matter (SOM) and short-term microbial C and N dynamics were investigated. Short-term changes in chemical and biological variables after incorporating fresh rye shoots were measured in intact soil cylinders from three contrasting agricultural systems. Two of the soils were from organic or conventional 4-yr rotations which had been in place for 6 yr as part of the University of California at Davis Sustainable Agriculture Farming Systems (SAFS) project and the third was from a double-cropped, intensive vegetable production system in the Salinas Valley of California. Microbial biomass (MB) and respiration, numbers of organisms in several trophic groups, soil inorganic N, dissolved organic C and recoverable rye were measured before and during the 6 weeks following rye incorporation. Active soil organic matter, expressed as the ratios of microbial biomass C or N to total soil C or N, respectively, appeared to be related to long-term management. These ratios increased in proportion to increased organic inputs and reduced tillage or periods of fallow. In all soils, MBC increased and decreased rapidly following rye incorporation, but MBN was fairly constant. Significant differences among the soils in MBC and MBN were maintained over the 6 week experiment. Following rye incorporation, fluorescein diacetate (FDA) active counts of bacteria and bacterial-feeding nematodes increased rapidly, whereas changes in FDA active fungal hyphal lengths and fungal-feeding nematodes were less pronounced. The rates of rye decomposition, respiration and net N mineralization were highest the first week after incorporation, coincident with increases in MBC and numbers of active bacteria in all three soils. There were significant differences among soils in numbers of organisms in the trophic groups on some sample dates, but changes in soil respiration and inorganic N and the rate of rye decomposition remained similar in all three soils. The SAFS organic soil had a somewhat lower ratio of bacterial to fungal biomass and lower ratio of respiration to MBC throughout the experiment than the SAFS conventional soil. Despite long-term differences in agricultural management and differences in active SOM contents among the three soils, the rates of rye decomposition and C and N mineralization were similar. Rye incorporation produced a short-term burst of microbial growth and activity of similar magnitude in all three soils although the initial MB contents in the three soils were different. Variations among the soils in FDA active counts of fungi and numbers of bacterial- and fungal-feeding nematodes indicated that microbial community composition was more responsive to rye incorporation than were changes in soil C and N pools. © 1998 Elsevier Science Ltd. All rights reserved.

1. Introduction

Agricultural benefits from increasing organic inputs to soil include greater soil N mineralization potential (Bonde et al., 1988; Fraser et al., 1988; Wander et al., 1994) as well as improved physical properties such as more rapid water infiltration and greater soil aggregation (Roberson et al., 1995). Such changes occur even with only small changes in total soil organic matter (SOM) due to enhancement of active SOM pools. Active SOM, which is comprised of soil microbial biomass (MB) and microbial metabolites and recently added, labile organic inputs to soil, has a complete turnover time on the order of a year, while the remainder of SOM has a complete turnover time in the order of 500 yr (Paul and Juma, 1981; Paul, 1984; Parton et al., 1987; Stevenson, 1994). Therefore it is not surprising that decreases or increases in response to manage-
ment changes are more rapid for active than total SOM (Janzen et al., 1992; Biederbeck et al., 1994). Because MB and recent organic additions are relatively labile nutrient pools and because MB is the agent of decomposition and N mineralization, active SOM supplies a majority of N mineralized although it represents a small fraction of total SOM (Paul and Juma, 1981). Soil microbes also play a large role in enhancement of physical structure because of the importance of bacterial extracellular polysaccharides and fungal hyphae in soil aggregate formation (Molope and Page, 1986; Eash et al., 1994; Roberson et al., 1995).

Microbial biomass has been proposed as an early indicator of changes in total SOM (Powlson et al., 1987). With its dual roles as a pool of labile nutrients and as the agent of decomposition of organic materials in soil, MB may be a sensitive indicator of changes in active SOM. However there are complications to the use of MB as an indicator of active SOM. Complete turnover time for MB is approximately 1 yr (Paul and Juma, 1981; Paul, 1984), however portions of MB turnover much more rapidly. MB contents can fluctuate sharply over days to weeks following tillage, wetting of dry soil or organic matter incorporation (Ocio et al., 1991; Wyland et al., 1995; Wyland et al., 1996; Gunapala and Scow, in press). The composition of the microbial community, in particular the proportions of bacteria and fungi, may also influence C and N turnover of active SOM. In studies where surface-placement or incorporation of crop residues have been examined, an increased proportion of fungal biomass associated with surface residues and slower decomposition of these residues have been found. Part of this decrease in the rate of decomposition is attributed to the greater C assimilation efficiency (where C assimilation efficiency is the ratio of microbial C to total C metabolized) and slower biomass turnover rates for fungi relative to bacteria (Holland and Coleman, 1987; Beare et al., 1992).

In this study we compared soils from three agricultural systems which were likely to have differences in amounts of active SOM due to long-term differences in agricultural practices. We compared soils under organic (referred to as Y-O) or conventional (referred to as Y-C) management at the University of California at Davis Sustainable Agriculture Farming Systems (SAFS) project in Yolo County, California. The SAFS project compares differences in soil properties, pest occurrence, crop yields and economic viability of four farming systems and for this study the 4 yr crop rotations (tomato-, safflower-, corn-, wheat- or legume seed) for the organic and conventional systems had been followed for 6 yr. In the organic system green manure cover crops are used in place of winter fallow and animal manure is also used as an N source, while in the conventional system, inorganic N fertilizer is used. Because of the reduced fallow and increased organic inputs in the organic relative to the conventional soil, higher active SOM was expected in the organic soil (Collins et al., 1992; Wander et al., 1994). The third soil (referred to as S-C) was from a grower’s field in Monterey County in the Salinas Valley of California. This field was under intensive conventional vegetable crop production typical for the area. The crop rotation there consisted of two to three lettuce and cole crops per year which require frequent tillage and irrigation and which return very little plant material to the soil. In addition inorganic N fertilizer is used and soil is usually bare in winter. There is evidence for large declines in SOM in Salinas soils under similar management. Using soil survey information and recent SOM measurements, Wyland et al. (1996) found an approximate 50% decline in SOM between 1901 and 1993 in a Salinas Valley soil under intensive vegetable crop production. Active SOM was expected to be even more depleted in the Salinas than Yolo conventional soil because of the lower amounts of plant residue return and more frequent tillage in the Salinas soil.

Our objectives were to compare initial SOM pools in the three soils and to measure short-term changes in MB, microbial community composition, and soil C and N pools following incorporation of rye shoots in order to answer several questions. (1) Do these soils differ in MB and community composition? (2) Does a one-time input of a cover-crop override differences in MB due to management history or soil origin, and if so, for how long? (3) Do temporal patterns in MB and community composition correspond to changes in soil C and N pools?

2. Materials and methods

2.1. Field sampling

Soil was sampled from the SAFS project in the Sacramento Valley from one plot under organic and one under conventional tomatoes. The soil type at this site is Yolo loam, a fine-silty, mixed, nonacid, thermic Typic Xerorthents. In order to reduce the effect of decomposition of tomato roots during the subsequent experiment, tomato plants and their main roots were removed on 8 July, 1994 in the area where soil was to be sampled. The soil was irrigated twice before soil cores were sampled on 22 July, 1994. The average annual C inputs to the SAFS organic and conventional soils were approximately 9.7 and 7.1 t ha\(^{-1}\), respectively, based on 1989 to 1992 total dry matter inputs (Scow et al., 1994) and assuming 40% C content. For a complete description of the management and crop rotations for the SAFS project see Scow et al. (1994).
Soil was sampled from a grower’s field in the Salinas Valley on 29 July, 1994, from beds prepared for lettuce planting and following a recently harvested celery crop. The soil type was Salinas loam, a fine-loamy, mixed, thermic, Pachic Haploxeroll. Characteristics of the soils and treatment abbreviations are given in Table 1. Crop residue return to the Salinas soil assuming one lettuce and one cole crop per year was approximately 3.2 t ha⁻¹ yr⁻¹ (Louise Jackson, personal communication). In both locations intact soil cylinders 20.3 cm diameter × 30 cm deep were removed from the field by driving steel pipes with an inner diameter of 20.3 cm into the soil and digging out the pipes with the soil cylinders inside. At the same time soil bulk density was measured in the field with a metal cylinder of known volume (3.6 to 11.4 cm deep and 19 to 26.6 cm deep, three replications per soil).

2.2. Experimental design

In order to have a sufficient volume of soil for sampling over 6 weeks with minimal disturbance of the soil cores, three 20.3 cm diameter cylinders were used for one replication of each treatment. The three cylinders in one replication (experimental unit) were placed adjacent to each other, still in the metal pipes, in the 25°C constant temperature room where the experiment was conducted. There were three replications for each treatment giving a total of 9 cylinders per treatment and 27 cylinders total. The three experimental units of the three treatments were arranged in a completely randomized design in the constant temperature room.

Based on soil moisture samples taken in the field, the cylinders were brought to a constant moisture of approximately −0.03 MPa by applying water evenly over the cylinder surfaces at a rate which did not allow water to pool. The cylinders were stored for 2 to 3 weeks at 25°C, the temperature at which the experiment was carried out. There were three replications for each treatment giving a total of 9 cylinders per treatment and 27 cylinders total. The three experimental units of the three treatments were arranged in a completely randomized design in the constant temperature room.

Soil was sampled from a grower’s field in the Salinas Valley on 29 July, 1994, from beds prepared for lettuce planting and following a recently harvested celery crop. The soil type was Salinas loam, a fine-loamy, mixed, thermic, Pachic Haploxeroll. Characteristics of the soils and treatment abbreviations are given in Table 1. Crop residue return to the Salinas soil assuming one lettuce and one cole crop per year was approximately 3.2 t ha⁻¹ yr⁻¹ (Louise Jackson, personal communication). In both locations intact soil cylinders 20.3 cm diameter × 30 cm deep were removed from the field by driving steel pipes with an inner diameter of 20.3 cm into the soil and digging out the pipes with the soil cylinders inside. At the same time soil bulk density was measured in the field with a metal cylinder of known volume (3.6 to 11.4 cm deep and 19 to 26.6 cm deep, three replications per soil).

Table 1. Soil properties (0–15 cm) of Yolo loam, organic farm management (Y-O), Yolo loam, conventional farm management (Y-C), and Salinas loam, conventional management (S-C)

<table>
<thead>
<tr>
<th>Soil</th>
<th>pH</th>
<th>Total SOM (% by weight)</th>
<th>C.E.C. (meq 100 g⁻¹ soil)</th>
<th>Sand %</th>
<th>Silt %</th>
<th>Clay %</th>
<th>P (μg P g⁻¹ soil)</th>
<th>Bulk density (g cm⁻³)</th>
<th>Soil moisture release curve (Y is g H₂O g⁻¹ soil, X is MPa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Y-O</td>
<td>7.27</td>
<td>1.75</td>
<td>23.5</td>
<td>37</td>
<td>45</td>
<td>18</td>
<td>32</td>
<td>1.39</td>
<td>Y = −0.0263 ln(X) + 0.1357</td>
</tr>
<tr>
<td>Y-C</td>
<td>6.88</td>
<td>1.57</td>
<td>23.0</td>
<td>38</td>
<td>44</td>
<td>18</td>
<td>23</td>
<td>1.37</td>
<td>Y = −0.0257 ln(X) + 0.1263</td>
</tr>
<tr>
<td>S-C</td>
<td>7.72</td>
<td>2.60</td>
<td>28.0</td>
<td>28</td>
<td>50</td>
<td>22</td>
<td>87</td>
<td>1.39</td>
<td>Y = −0.0191 ln(X) + 0.1615</td>
</tr>
</tbody>
</table>
we wanted to reduce variability of all measurements due to variations in the amount of rye residues present in the small samples used for analysis. Soil respiration with rye residues present and counts of active bacteria and measurements of fungal hyphae and soil gravimetric moisture were made immediately after sampling. Visible rye pieces were not included in the soil used for active counts. The remaining soil was weighed and sieved (4 mm), and all visible rye pieces were removed, rinsed and dried at 65°C to estimate the undecomposed plant material remaining. After sieving, soil was immediately stored at 4°C and all measurements and extractions were completed within 5 d. All analyses were performed in triplicate except for soil total C and N content, weight and C and N content of recovered plant material and nematode counts (one replication) and measurement of respiration without removal of rye residues (four replications).

2.3. Laboratory analyses

Background soil analyses for organic matter content (Nelson and Sommers, 1982), cation exchange capacity (Janitzky, 1986), particle size distribution (Gee and Bauder, 1979) and phosphorous (Olsen et al., 1954) were made on bulked air-dry soil samples from the pre-incorporation soil sample at the Division of Agriculture and Natural Resources Analytical Laboratory, University of California Cooperative Extension (Table 1). Soil pH was measured using a 1:1 soil to deionized water paste.

All other measurements were made on soils at the moisture content when sampled. Based on gravimetric water content, measurements were converted to a dry soil basis and based on field measurements of bulk density (Table 1), the measurements were converted to a volume basis. Values are reported this way in order to account for differences in field bulk density. Soil MB was measured using the fumigation extraction method (modified from Vance et al., 1987; Sparling and West, 1988; Tate et al., 1988). 15 g of moist soil were extracted immediately after sampling by shaking for 30 min with 60 ml of 0.5 m K₂SO₄ and 15 g were fumigated for 24 h with ethanol-free chloroform and then extracted as above. Microbial biomass C (MBC) was determined using a Shimadzu TOC-5050 analyzer (Shimadzu Columbia, MD) and MBN was determined by the ninhydrin method (Carter, 1991). Values reported for MBC and MBN are the flush due to fumigation (fumigated values — non-fumigated values) with no conversion.

Dissolved organic C (DOC) was determined according Burford and Bremner (1975) with modifications. Moist soil (7.5 g) was gently shaken with 15 ml deionized H₂O for 15 min. The soil was centrifuged for 5 min and the supernatant again for 30 min at 19,000g. The supernatant then was filtered through a 0.2 µm polycarbonate membrane filter (Poretics Corporation, Livermore, CA), and was analyzed for C using a Shimadzu TOC-5050.

To measure NO₃⁻N and NH₄⁺-N 5 g soil was shaken for 45 min in 25 ml 2 M KCl. The soil was centrifuged at 5,000 rpm for 5 min and the clear supernatant was analyzed using a Wescan ammonia analyzer (Alltech Assoc., Deerfield, IL) with a reduction column for NO₃⁻N (Carlson, 1978; Carlson, 1986).

Respiration was measured by incubating 5 g moist soil in a sealed 60 ml flask for 24 h. Then a 1 ml headspace sample was injected for CO₂ detection into a Horiba PIR-2000 (Horiba Instruments, Riverside, CA) which uses infrared detection. Respiration was measured without removing visible rye pieces in order to get an accurate measure of total CO₂ evolution. The high variability in CO₂ measured by this method was probably due to spatial variation in amount of rye present. Starting at 3 weeks, respiration was also measured on unsieved soil, with visible plant material removed (basal respiration).

For organism counts 10⁻¹ to 10⁻⁶ soil dilutions were prepared in 50 mM phosphate buffer. To separate organisms from soil particles, the 10⁻¹ dilution was shaken gently (90 cm trajectory s⁻¹) for 5 min. For numbers of fluorescein diacetate (FDA) active bacteria and fungal hyphal length the method of Ingham and Klein (1984) was modified. To a 0.5 ml aliquot of the 10⁻¹ dilution, 1 ml of deionized H₂O and 1 ml of FDA solution were added. After 3 min, 2 ml molten agar (pH > 7.6) was added, the suspension was mixed and an aliquot was placed in a slide with a well 20 × 20 mm and 0.15 mm deep. Numbers of FDA active bacteria and fungal hyphal length were counted in six fields per slide within 1 h after slide preparation using epifluorescence microscopy.

Nematodes were extracted from 75 g soil using modified Cobb sieving and sugar centrifugation (Barker, 1985). Nematodes were separated into food preference groups; bacterial feeding, fungal feeding, plant parasitic or predatory based on morphology of the stoma and esophagus.

Both soil and plant material were ground to pass through a 420 µm screen. The C and N content of the plant material and total soil C and N content were measured by combustion and gas chromatography using a Carlo-Erba NA 1500 C and N elemental analysis system (Fisons Instruments, Beverly, MA).

2.4. Statistical analyses

SAS version 6.10 (SAS Institute, Cary, NC) was used for all data analyses. Data for individual sample dates were subjected to one-way analysis of variance
and where significant differences were found at $P < 0.05$, LSDs were calculated for mean separation. Data were also analyzed by combining all dates in a multivariate repeated measures analysis of variance. Contrasts between the initial pre-incorporation amounts of a variable and subsequent dates were generated to test whether post-incorporation amounts differed from the pre-incorporation amount (SAS Institute, 1988). All of the results from the repeated measures analysis of variance and contrasts are summarized in Table 2, but due to the large number of tests, Table 2 is not cited in the text. The rates of decomposition in the three soils were compared by fitting the recovered plant weight data to a first order decay model using the Marquardt algorithm (SAS Institute, 1988). Significant differences between the three sets of two curves were made by comparing the improvement in the fit of each curve fit separately to the pooled data of two curves according to Motulsky and Ransnas (1987).

3. Results

3.1. Pre-incorporation soil organic matter pools

Total soil C and N were about twice as high in the Salinas loam (S-C) as in the Yolo loam (Y-C, Y-O)
(Table 3). Total soil C and N were only slightly, and not significantly, higher in the Y-O than Y-C soil. Like total C and N, DOC was significantly higher in S-C followed by Y-O and Y-C. The pattern for MBC and MBN were different with the highest amounts in Y-O, followed by S-C and Y-C. Because MB is part of active SOM, comparing the ratio of MB to total SOM may indicate the relative amount of active SOM in the soils. The ratio of MBC to total soil C and MBN to total soil N were greatest in the Y-O soil, intermediate in the Y-C and lowest in the S-C soil and these ratios were significantly different among all three soils.

3.2. Microbial biomass and microbial community composition

Microbial biomass C increased substantially and rapidly following cover crop incorporation in all soils (Fig. 1a). It was 46–63% higher, 1 week after incorporation and then began to decline until it reached and leveled off at pre-incorporation quantities by week 4. MBC concentrations in the three soils remained significantly different throughout this rapid fluctuation in MBC with Y-O remaining highest followed by S-C and Y-C. Unlike MBC, MBN did not increase signifi-
cantly following incorporation, and was actually significantly lower from 3 to 6 weeks after incorporation (Fig. 1b). Like MBC the differences in MBN across soils remained significant throughout the experiment with MBN in Y-O remaining highest, followed by S-C and Y-C during the entire experiment. The C-to-N ratio of microbial biomass was elevated from 1 to 3 weeks post-incorporation after which it returned to pre-incorporation values (Table 4).

Numbers of FDA active bacteria increased sharply (24–52%) just 1 week after rye shoot incorporation (Fig. 2a). However unlike microbial biomass, numbers of FDA active bacteria did not decline until 3 weeks after incorporation and remained in significantly higher numbers thereafter than at pre-incorporation. In contrast to MBC and MBN, there were very few significant differences in the number of FDA-active bacteria among the three soils.

In the Yolo soils, bacterial-feeding nematodes were 6- to 7-fold more numerous at 2 weeks after incorporation and remained in numbers that were 4 to 6 times higher than pre-incorporation estimates (Fig. 2b). In the Salinas soil, numbers of bacterial-feeding nematodes increased more gradually than in the Yolo soils and reached a maximum at 4 weeks post-incorporation.

FDA-active fungal hyphae appeared to increase slightly during the 6 week experiment, however the effect of date was significant only at \( P = 0.10 \). The total length of FDA-active fungal hyphae was similar in the two Yolo soils and was about 50% lower in the Salinas soil than the Yolo soils in the samples before incorporation and for 3 weeks thereafter (Fig. 2c). On weeks 4 and 6 there were no significant differences among treatments in length of active hyphae. The ratio of FDA-active bacteria to FDA-active fungal hyphal length did not change over time in any of the treatments. This ratio was significantly higher in S-C than the Yolo soils for the first three dates and on all but week 2, followed the order S-C > Y-C > Y-O.

Fungal-feeding nematodes increased in numbers after incorporation only in the Y-O soil, however they dropped to numbers similar to those in Y-C and S-C on week 4 (Fig. 2d). In Y-C and S-C, numbers of fun-

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Table 4. Selected measures of microbial community structure and activity without rye residues and rye C and N content in three soils before and for 6 weeks following incorporation of rye. Means followed by the same letter not significantly different at \( P < 0.05 \) for each sample date.

<table>
<thead>
<tr>
<th>Soil</th>
<th>Weeks after incorporation</th>
<th>0</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Y-O</td>
<td>MBC:MBN (µg C µg N⁻¹)</td>
<td>9.46</td>
<td>12.60</td>
<td>11.28</td>
<td>14.39</td>
<td>9.35</td>
<td>9.44</td>
</tr>
<tr>
<td>Y-C</td>
<td>9.43</td>
<td>15.30</td>
<td>13.53</td>
<td>15.94</td>
<td>10.22</td>
<td>10.65</td>
<td></td>
</tr>
<tr>
<td>Y-O</td>
<td>FDA bacteria:fungi (No. X 10⁻⁷: m)</td>
<td>1.63 b</td>
<td>1.94 b</td>
<td>1.97 b</td>
<td>1.76</td>
<td>1.86</td>
<td>1.30</td>
</tr>
<tr>
<td>Y-C</td>
<td>2.13 b</td>
<td>2.03 b</td>
<td>1.67 b</td>
<td>2.40</td>
<td>2.12</td>
<td>2.31</td>
<td></td>
</tr>
<tr>
<td>S-C</td>
<td>4.61 a</td>
<td>4.28 a</td>
<td>4.14 a</td>
<td>3.33</td>
<td>2.48</td>
<td>2.65</td>
<td></td>
</tr>
<tr>
<td>Y-O</td>
<td>Basal respiration (µg CO₂ cm⁻³ h⁻¹)</td>
<td>2.09 a</td>
<td>ND</td>
<td>ND</td>
<td>2.10</td>
<td>1.85 a</td>
<td>1.99</td>
</tr>
<tr>
<td>Y-C</td>
<td>1.35 b</td>
<td>ND</td>
<td>ND</td>
<td>1.52</td>
<td>1.51 b</td>
<td>1.70</td>
<td></td>
</tr>
<tr>
<td>S-C</td>
<td>1.11 b</td>
<td>ND</td>
<td>ND</td>
<td>1.61</td>
<td>1.36 b</td>
<td>1.73</td>
<td></td>
</tr>
<tr>
<td>Y-O</td>
<td>Basal resp.: MBC (µg CO₂ µg C⁻¹ h⁻¹)</td>
<td>0.0144</td>
<td>ND</td>
<td>ND</td>
<td>0.0115</td>
<td>0.0139 a</td>
<td>0.0147</td>
</tr>
<tr>
<td>Y-C</td>
<td>0.0148</td>
<td>ND</td>
<td>ND</td>
<td>0.0121</td>
<td>0.0163 a</td>
<td>0.0169</td>
<td></td>
</tr>
<tr>
<td>S-C</td>
<td>0.0097</td>
<td>ND</td>
<td>ND</td>
<td>0.0099</td>
<td>0.0111 b</td>
<td>0.0140</td>
<td></td>
</tr>
<tr>
<td>Y-O</td>
<td>Rye N content (% dry weight)</td>
<td>3.17</td>
<td>1.16</td>
<td>1.53 a</td>
<td>1.70</td>
<td>1.61</td>
<td>1.69</td>
</tr>
<tr>
<td>Y-C</td>
<td>3.17</td>
<td>1.24</td>
<td>1.49 a</td>
<td>1.54</td>
<td>1.75</td>
<td>1.63</td>
<td></td>
</tr>
<tr>
<td>S-C</td>
<td>3.17</td>
<td>1.22</td>
<td>1.22 b</td>
<td>1.47</td>
<td>1.60</td>
<td>1.73</td>
<td></td>
</tr>
<tr>
<td>Y-O</td>
<td>Rye C content (% dry weight)</td>
<td>43.1</td>
<td>39.7</td>
<td>41.8</td>
<td>43.4 a</td>
<td>41.9</td>
<td>41.9</td>
</tr>
<tr>
<td>Y-C</td>
<td>43.1</td>
<td>40.9</td>
<td>40.4</td>
<td>43.7 a</td>
<td>40.5</td>
<td>41.7</td>
<td></td>
</tr>
<tr>
<td>S-C</td>
<td>43.1</td>
<td>41.1</td>
<td>40.3</td>
<td>41.6 b</td>
<td>42.0</td>
<td>41.2</td>
<td></td>
</tr>
</tbody>
</table>

ND indicates not determined.
Fig. 2. Abundance of organisms in several trophic groups before and after rye incorporation, (a) FDA-active bacteria, (b) bacterial-feeding nematodes, (c) FDA-active fungal hyphae, (d) fungal-feeding nematodes in Y-O, Y-C, Q, and S-C. Means followed by the same letter not significantly different at $P < 0.05$ on each sample date. Error bars ± one standard error, $n = 3$. 
gal-feeding nematodes increased gradually throughout the experiment. Initial numbers of fungal-feeding nematodes were lower than bacterial-feeders, and they increased only by a factor of 2 to 3. Thus, numbers of bacterial-feeding nematodes remained 4 to 10 times higher than fungal-feeders in post-incorporation samples.

3.3. Soil C and N pools

Respiration from soil including the rye residue increased from 1 to 3 times above pre-incorporation rates by 1 week after incorporation (Fig. 3). It then gradually declined, but did not return to pre-incorporation rates except in the Y-O soil. The lack of differences among soils may have been due to the variability in the amount of rye residue in the 5 g replicate subsamples of soil. Starting 3 weeks after incorporation, respiration was also measured in soil samples from which visible rye residues had been removed (basal respiration). Basal respiration rates remained somewhat elevated from pre-incorporation rates in Y-C and S-C at 3, 4 and 6 weeks after incorporation, but not in Y-O (Table 4). The basal respiration rate was significantly higher in Y-O than Y-C and S-C soils at pre-incorporation and at 4 weeks. The ratio of basal respiration to MBC did not differ substantially among the three soils, however, the ratio was always lowest in S-C followed by Y-O and Y-C.

Dissolved organic carbon (DOC) concentrations did not change significantly following rye shoot incorporation (Fig. 4). Throughout the experiment, DOC in S-C was significantly higher than Y-O which was significantly higher than Y-C.

Evidence of net N mineralization during the first week after incorporation was indicated by increases in NO$_3^-$-N concentrations of approximately 25 µg N cm$^{-3}$ in all three soils in the 0–15 cm layer (Fig. 5a). Substantial increases in nitrate below 15 cm in Y-O and S-C indicated net N mineralization or NO$_3^-$-N leaching, although no water was observed to leak from the bottom of the cylinders (Fig. 5b). However, the total change in inorganic N (0–25 cm) between any two sample dates was not significantly different among the three soils (data not shown). The soil NH$_4^+$-N levels remained low throughout the experiment (Fig. 5c and d).

The amount of rye residue recoverable by sieving declined rapidly in a first order decay pattern (Fig. 6). By 6 weeks after incorporation only 15% of the weight of added plant material was recoverable. The decomposition rates in the three soils were not significantly different. In all three soils the N content of the recovered rye dropped from the pre-incorporation value of 3.2% to 1.2% 1 week after incorporation after which it increased gradually to 1.7% by 6 weeks after incorporation (Table 4). Rye C content in the three soils remained between 40 and 43% for all dates.

Because the changes in weight or C and N content of recovered rye, respiration and inorganic N were not significantly different among the three soils (data not shown), we compared changes in these variables for the average of the three soils between each two sample dates (Fig. 7a and b). Because inorganic N increased

![Fig. 3. Soil respiration with rye residues in Y-O, Y-C, and S-C before and after rye incorporation. Means followed by the same letter not significantly different at $P < 0.05$ on each sample date. Errors bars ± one standard error, n = 3.](image-url)
at the 15–25 cm depth, indicating possible leaching from the 0–15 cm layer, we compared net N mineralized (the change in inorganic N) for the entire 0–25 cm to the decrease in rye N (Fig. 7a). The decrease in rye N occurred almost entirely during the first week when plant weight and N content both dropped substantially (Fig. 6 and Fig. 7a, Table 4). Not all of the decrease in rye N content during the first week could be accounted for as net N mineralization. Likewise most rye C was lost during the first week post-incorporation, and the increase in respiration above pre-incorporation rates did not account for rye C loss during the first week after incorporation (Fig. 7b).

4. Discussion

4.1. Total and active SOM

Because active SOM concentrations change more rapidly than total SOM concentrations (Janzen et al., 1992; Biederbeck et al., 1994), active SOM as a proportion of total SOM should increase when agricultural practices which increase total SOM have been initiated. Conversely, active SOM as a proportion of total SOM should be low in soils where organic matter contents have been declining according to the same reasoning. The ratios of MBC to total soil C and MBN to total soil N in this study followed the order Y-O > Y-C > S-C. This ordering corresponded to the relative intensity of agricultural management practices, e.g. organic inputs, tillage frequency and length of fallow, all of which are likely to affect soil organic matter content. Y-C received approximately 4 t ha\(^{-1}\) yr\(^{-1}\) more organic C than S-C, and S-C also experienced more frequent tillage due to double cropping. Y-O received approximately 3 t ha\(^{-1}\) yr\(^{-1}\) more organic C than Y-C and used winter cover crops in place of fallow in Y-C.

One complication to the use of MB as an indicator of active SOM is that being the most labile component of active SOM, MB may fluctuate over very short periods and therefore may not always be a stable indicator of active SOM. In our study MB increased 50–60%, but the increase was sustained for only 3 weeks.

The large short-term response of the microbial community to rye incorporation demonstrates that short-term fluctuations in MB may contribute substantially to annual turnover rates of MB. The concept of a constant turnover rate for MB and active SOM may need to be extended to include the contributions to turnover from disturbances such as residue addition or tillage.

4.2. Microbial biomass and C and N pool dynamics

The pattern of MB and C and N pool dynamics following rye incorporation in the three soils appeared to indicate C-limited microbial populations which responded very rapidly to C inputs. The rapid, but short-term increase in microbial populations may to correspond to zymogenous bacteria, which grow rapidly following the addition of high energy organic materials (Winogradsky, 1949). Some researchers have suggested that organisms which proliferate following disturbance and under high nutrient availability conditions are likely to be r-selected organisms character-
Fig. 5. Soil inorganic N before and after ryegrass incorporation (a) NO$_3$-N at 0–15 cm, (b) NO$_3$-N at 15–25 cm, (c) NH$_4$+ -N at 0–15 cm, (d) NH$_4$+ -N at 15–25 cm in Y-O, Y-C, Q, and S-C. Means followed by the same letter not significantly different at $P < 0.05$ on each sample date. Error bars are 1 standard error, n = 3.
Fig. 6. Weight of recoverable rye from 1 to 6 weeks after incorporation cm in Y-O, Y-C, and S-C. Weight at pre-incorporation is total weight added, on subsequent dates weight recovered is shown.

Fig. 7. Comparison of apparent N mineralized to rye N loss (a) and C mineralized above pre-incorporation values to rye C loss (b) between each two sample dates for the average of all three soils.
ized by high growth and reproductive rates and low yield of biomass per unit of substrate (Odum, 1969; Panikov, 1995).

Microbial activity as indicated by active counts of bacteria and fungi was still elevated over pre-incorporation in all three soils by week 6 and respiration was still elevated in Y-C and S-C at week 6. Both respiration with rye and basal respiration had returned to pre-incorporation rates in Y-O by 6 weeks after incorporation. Therefore the organic soil appeared to return more rapidly to a microbial community dominated by autochthonous or K-selected organisms which are more competitive under steady state, low nutrient supply conditions and have higher biomass yield efficiency (Winogradsky, 1949; Panikov, 1995).

Nitrogen did not appear to be limiting to microbial populations in these soils because MBN remained at the same concentration throughout the experiment. In contrast, Wyland et al. (1996) observed a rise and fall in MBN during 1 week following cover crop incorporation. A similar upsurgence and decline in MBN could have occurred before the 1 week sample in this study. With the rye’s low C-to-N ratio of 13-to-1, net N mineralization was expected (Janssø, 1958) and was observed in this study. Therefore an excess of inorganic N was available to soil microbes.

Not all of the rye N and C loss during the first week could be accounted for when decreases in recovered rye N and C were compared to apparent N and C mineralization. It is likely that some of the apparent decrease in rye C and N was due to incomplete recovery of the rye material. For the first week we compared the change from the total added to that recovered at week 1. For subsequent time periods we compared the change in recovered plant material between dates, and assuming constant recovery efficiency, no error would have been introduced. However we feel it is likely that recovery efficiency was high, on the order of 70 to 80% and this factor cannot account for all the discrepancies in C and N measured during the first week.

Denitrification was probably responsible for part of the discrepancy in N pools, since microbial immobilization of N appeared to be minimal. Because denitrification is often limited by C availability and is enhanced during periods of high microbial activity and consequent oxygen depletion (Paul and Clark, 1989), denitrification rates were likely higher during the first week after rye incorporation than in subsequent weeks. Microbial immobilization of C in the first week could have accounted for some of the discrepancy between rye C loss and increased respiration above the pre-incorporation rates. The average increase in chloroform-labile MBC of 67 μg C cm⁻³ soil corresponds to a total of 176 μg C cm⁻³ soil in MB assuming the calibration constant of 2.64 of Vance et al. (1987). The remaining unaccounted for C could have been due to high respiration rates soon after rye was incorporated stimulated by the presence of easily-decomposable compounds in the fresh rye, or by higher MBC contents following incorporation which had already declined by 1 week after incorporation.

The lack of change in DOC during the experiment may also have been due to rapid microbial utilization of readily available soluble compounds in the rye before the 1 week sample. DeLuca and Keeney (1993) found that anthrone-reactive C (a measure of soluble hexose sugars) declined to low and constant amounts in only 5 to 10 d after incorporation of plant residues. The lack of change in DOC content during the remainder of the experiment was not surprising as other researchers have found that DOC concentrations did not change with season (Boyer and Groffman, 1996). These authors also found that the potential degradation rate of DOC in a laboratory incubation was 8% d⁻¹ for DOC extracted from surface soil of a corn field. During a 210 d laboratory incubation of several soils, Cook and Allan (1992a,b) found that DOC increased, but the bioavailability of DOC decreased. Therefore it appears that a portion of DOC may turn-over rapidly, but that the resistant portion of DOC may increase over time when there have been no recent organic inputs to soil.

4.3. Microbial community composition and its relationship to C and N dynamics

When we compared active biomass measured by direct counts and total biomass as measured by fumigation–extraction, FDA active bacteria and fungi accounted for only 14% of microbial biomass C on average. This was based on a fungal hyphal dia of 3 μm, bacterial volume of 1 μm³, a biovolume to biomass of 1.09 g cm⁻³ for bacteria and fungi, dry matter contents of 30 and 20% for bacteria and fungi, respectively, (Bakken and Olsen, 1983), a C content of 45% for both bacteria and fungi, and converting chloroform-labile MBC to total according to Vance et al. (1987). Our FDA counts may have underestimated total active MB because not all bacterial isolates are permeable to FDA, and both bacteria and fungi vary in the degree of fluorescence of FDA between species and at different growth stages (Söderstrom, 1977; Lundgren, 1981; Chrzanowski et al., 1984). In addition a mild soil dispersal method was used in order to prevent damage to FDA active hyphae (Söderstrom, 1977) and it is likely that not all organisms were released from soil particles. However, we feel that the active counts reflected relative differences between the soils and over time.

Large changes in abundance of organisms in certain microbial groups coincided with rapid rye decomposition and C and N mineralization. Both active bac-
teria and bacterial-feeding nematodes increased substantially and rapidly following rye incorporation while fungi and fungal-feeding nematodes increased more slowly and to a lesser extent. Therefore it appears that the incorporation of a low C-to-N ratio cover crop stimulated a largely bacterial response in these soils, while fungal biomass increased when relatively resistant and higher C-to-N ratio plant material remained. Other researchers have found that decomposition of incorporated residues was dominated by bacteria (Beare et al., 1992) and have observed a similar succession of bacterial-followed by fungal-feeding nematodes after plant material incorporation (Sohlenius and Bostrom, 1984; Bohlen and Edwards, 1994).

Despite the overall similarities in organism number trends among the soils, there were significant differences in organism numbers on some dates. For example, there were lower FDA active fungal hyphae in S-C than the Yolo soils for 3 weeks after incorporation, lower bacterial-feeding nematodes in S-C than Y-O or Y-C 2 weeks after incorporation and higher fungal-feeding nematodes in Y-O than Y-C and S-C at 2 weeks post-incorporation. However these differences did not coincide with differences in dynamics of the C or N pools measured. This lack of correspondence may have been due to high experimental error in the organism counts, or it may have been that these organisms were not vital to the rye decomposition or C and N mineralization measured. Therefore in this study large temporal increases in microbial numbers generally coincided with large changes in soil C or N pools, but all trends in microbial community composition did not coincide with soil C and N pool dynamics.

Bacteria appear to dominate decomposition in these soils. FDA-active fungi comprised an average of only 25% of the biomass of FDA-active bacteria using the conversions listed above, even assuming a generous size estimate for fungal hyphal dia in agricultural soils of 3 μm (Schnürer et al., 1985; Holland and Coleman, 1987; Beare et al., 1992). In addition, FDA-active biomass may have been underestimated for bacteria relative to fungi because researchers have shown that only 80% of bacterial isolates from soil stain with FDA (Lundgren, 1981) compared to 100% of fungal isolates (Söderstrom, 1977). Numbers of FDA-active bacteria and length of fungal hyphae measured from field samples of SAFS organic and conventional soils in the months prior to this study were similar to those found in this study (Lundquist, unpublished data). Additional evidence for bacterial dominance of communities in these soils was found in the higher trophic levels. Fungal-feeding nematode numbers were always lower than bacterial-feeding nematodes during this experiment, as observed by Ferris et al. (1996) in the SAFS plots during the tomato growing season. In addition, fluctuations in MBC were more similar to the fluctuations in FDA-active bacteria than fungi. Finally, FDA-active hyphal lengths measured in this study (4 to 19 m g⁻¹ soil) were on the low end of those reported in the literature. In agricultural soils, hyphae range from 2 to 10 m g⁻¹ soil total hyphal length in a semi-arid climate (Holland and Coleman, 1987) up to 39 to 140 m g⁻¹ soil FDA active hyphal length, and 700–1800 m g⁻¹ soil total hyphal length in a cool, temperate climate (Hansson et al., 1990).

Practices which increase fungal abundance in soils may increase soil C storage because fungi generally have a higher C assimilation efficiency and lower growth rate than do bacteria (Holland and Coleman, 1987). Fungal biomass was higher in Y-O relative to Y-C as indicated by a lower ratio of active bacteria-to-fungi and this trend may help to explain the lower ratio of CO₂-to-MBC in Y-O than Y-C. Sakamoto and Oba (1994) also found a negative correlation between fungal biomass and the ratio of soil respiration to MBC. It is possible that the differences in the proportion of fungal biomass between Y-O and Y-C will increase in the SAFS project as the organic and conventional management practices are continued beyond 6 yr. However, factors such as climate or the use of tillage may limit the abundance of fungi in these soils and thus minimize potential differences in the proportion of fungal biomass that would be expected with higher organic matter inputs.

Counter to the trend between Y-C and Y-O, the Salinas soil had a higher active bacteria-to-fungi ratio than did both Yolo soils and a basal respiration to MBC ratio as low or lower than the two soils. The comparison between the two Yolo soils should be more reliable than between the Salinas and Yolo soils, because the Yolo soils differ only in their management history for the past 6 yr. The Yolo and Salinas soils, on the other hand, differ in parent material, soil-forming conditions and long-term farm management.

Anderson and Domsch (1990) found that the ratio of respiration to microbial biomass for soils under continuous crop rotations was lower than for soils under long-term monoculture. In our study, however, the two soils under crop rotation had higher ratios of basal respiration to MBC than the soil under an intensive double crop vegetable production system.

Although the three soils studied were under very different management strategies, rates of rye decomposition and C and N mineralization were similar in all three. Rye incorporation produced a similar short-term burst of microbial growth and activity in the three soils despite their initially different MB contents. Variability among the soils in FDA-active fungal hyphal length and numbers of bacterial- and fungal-feeding nematodes suggested that microbial community composition...
was more sensitive to rye incorporation than were changes in soil MB and C and N pools.

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