Long-term cultivation of grassland soils reduces soil organic C and N content and has been associated with a deterioration in the aggregate structure of the soil. This study examined the effects of bare fallow (moldboard plow), stubble mulch fallow (subtill), and no-till fallow management on aggregate size distribution and aggregate organic C and N contents compared with native grassland soil. Aggregate size fractions were separated by wet sieving and the proportion of soil was quantified for each aggregate size class. Mineral-associated (silt and clay) organic matter was isolated by dispersing aggregates in sodium hexametaphosphate and removing the sand and particulate organic matter (POM) by passing the dispersed aggregates through a 53-µm sieve. The POM fraction is composed primarily of partially decomposed root fragments and has an average C/N ratio of about 16. A large proportion of the total soil dry weight (50-60%) was isolated in the small macroaggregates (250-2000 µm) size class. The native grassland soil was more stable than the cultivated soils when slaked, and the no-till soil was more stable than the stubble mulch and bare fallow soil when slaked. Reduced tillage management is effective at increasing the proportion of macroaggregates and results in the accumulation of wheat (Triticum aestivum L.) derived POM within the aggregate structure compared with bare fallow soil. It has previously been shown that the POM fraction accounts for the majority of the soil organic matter (SOM) initially lost as a result of cultivation of grassland soils. The data reported in this study relates the loss of structural stability from cultivation to losses of organic C and N from the POM fraction.

Native grassland soils are generally highly structured and rich in SOM. Cultivation reduces SOM content (Jenny, 1941; Haas et al., 1957) and results in a deterioration of the aggregate structure (Chaney and Swift, 1984). The POM fraction, which is composed primarily of partially decomposed root fragments, accounts for the majority of the SOM, initially lost as a result of cultivation of grassland soils (Cambardella and Elliott, 1992). Reductions in aggregate stability after cultivation are most pronounced in soil macroaggregates, while the stability of soil microaggregates remains unchanged (Tisdall and Oades, 1982; Oades, 1984). As macroaggregates disintegrate with tillage, the proportion of microaggregates increases, since microaggregates are not destroyed by cultivation (Tisdall and Oades, 1980; Elliott, 1986). Microaggregates have a lower organic matter concentration than macroaggregates (Dormaar, 1983) and this organic matter is less labile than that associated with macroaggregates (Elliott, 1986; Gupta and Germida, 1988). The disintegration of macroaggregates with cultivation into nutrient-poor microaggregates and the subsequent release of plant-available nutrients may be one explanation for the observed pattern of lower organic matter contents and reduced nutrient-supplying efficiencies in cultivated soils when compared with grassland soils (Elliott, 1986).

Sustainable production has become a key issue in the management of cropping systems. Reduced tillage and no-till management have been initiated to mitigate some of the detrimental effects of intensive cultivation, such as reduction of soil organic matter and the concomitant increase in erosion and decrease in soil fertility (Fenster and Peterson, 1979). Little information exists in the literature on the effect of reduced tillage or no-till management on the nutrient contents of soil aggregates and their associated organic matter. This study examined the effects of bare fallow (moldboard plow), stubble mulch fallow (subtill), and no-till management on aggregate size distributions and aggregate organic C and N contents compared with native grassland.

### MATERIALS AND METHODS

**Field Sampling**

Soil was collected from an experimental site located at the High Plains Agricultural Research Laboratory near Sidney, NE. The soil type is a Duroc loam (a fine-silty, mixed, mesic Pachic Haplustoll) and its parent material is mixed loess and alluvium. The chemical properties of the soil are given in Table 1 (Fenster and Peterson, 1979). The site had never been cultivated prior to 1969, when it was plowed and sown to winter wheat. Plow depth at sodbreaking was 20 cm. Three management treatments were established in addition to the control native grass plots: bare fallow (plowing), stubble mulch (subtill), and no-till. One wheat crop was removed every other year and the alternate year the ground was fallow. Fertilizer was not applied to any of the samples. The bare fallow treatment was tilled to a depth of 10 to 15 cm using a moldboard plow in the spring of the fallow year followed by two to three cultivator or rotary rod weeder operations. Stubble mulch fallow was cultivated with 0.9- and 1.5-m sweeps two to four times a year at a depth of 10 cm, followed by a rotary rod weeder. Weed control during fallow in the no-till treatment was accomplished with herbicides (Fenster and Peterson, 1979).

The experimental design was a randomized complete block with three field replicates. The three tillage treatments and the native grassland control were randomly assigned plots within each of the three field replicates. The entire design was repeated in an adjacent area of the field so that the crop and fallow portions of the rotations were represented every year instead of every other year. Treatment plots within field replicates were 8.5 by 46.0 m (Fenster and Peterson, 1979). We sampled the west rotation of the experiment, which had been in fallow since the summer of 1989, in late July of 1990. Cores were taken with an 8-cm-diam. steel coring bit to a depth of 20 cm without removing surface residues. One core was pulled every 4.5 m along the length of each plot, for a total of 10 cores per plot. The exact horizontal location along the width of the plot for each core was randomly located. To minimize edge effects, a 0.5-m buffer zone was established along the edges of each plot. The soil was gently broken apart by hand in the laboratory and passed through a 2.8-mm sieve while still moist.
Table 1. Chemical properties of the Duroc loam (0–1% slope) in 1969 (data from Fenster and Peterson, 1979).

<table>
<thead>
<tr>
<th>Soil depth</th>
<th>pH</th>
<th>Ctl</th>
<th>Ntl</th>
<th>C/N</th>
</tr>
</thead>
<tbody>
<tr>
<td>cm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0-10</td>
<td>7.4</td>
<td>2.33</td>
<td>0.19</td>
<td>12.4</td>
</tr>
<tr>
<td>11-20</td>
<td>7.5</td>
<td>1.55</td>
<td>0.13</td>
<td>11.6</td>
</tr>
<tr>
<td>21-30</td>
<td>7.5</td>
<td>0.92</td>
<td>0.12</td>
<td>7.9</td>
</tr>
</tbody>
</table>

† Organic C.
‡ Total Kjeldahl N.

moist. The large pieces of stubble and root that had passed through the sieve were removed by hand. The sieved soil was dried overnight at 50 °C, after which the soil taken from each plot was composited and stored at 4 °C. Bulk density was estimated using the total weight and volume of all cores from each plot.

Laboratory Methods

A 100-g subsample of soil from each plot was wet sieved through a series of three sieves to obtain four aggregate size fractions: (i) >2000 μm (large macroaggregates), (ii) 250 to 2000 μm (small macroaggregates), (iii) 53 to 250 μm (microaggregates), and (iv) <53 μm (silt- plus clay-size particles). The initial moisture content of the soil prior to wet sieving can alter the aggregate size distribution and partitioning of the organic matter associated with the aggregate size fractions (Kemper and Rosenau, 1986). Soil was either capillary wetted overnight at 4 °C or left dry (slaked) prior to sieving in order to obtain information on the distribution and relative stability of the aggregates. The capillary-wetted soils were consistently brought up to field capacity plus 5%, the moisture content at which maximum aggregate stability is attained for these soils. Six sieving replicates were completed for each plot for both capillary-wetted and slaked pretreatments. The prewetted or dry soil samples were suspended in room temperature water on the largest sieve for 5 min before sieving. Aggregate disruption was accomplished by moving the sieve 3 cm vertically 50 times during a period of 2 min, being careful to break the surface of the water with each stroke. Material remaining on the sieve was backwashed into a round aluminum cake pan and dried at 50 °C overnight in a forced air oven. Soil plus water that passed through the sieve was poured onto the next finer sieve size and the process repeated (Elliott, 1986). The smallest fraction (silt plus clay) was centrifuged 10 min at 2500 x g and the pellet backwashed to an aluminum cake pan and dried overnight at 50 °C. The dried aggregate size fractions were weighed and stored in wide-mouth snap cap vials at room temperature.

Prior to chemical analysis, plant residue and roots that were larger than ~1 mm in length were removed by hand with forceps from subsamples of the aggregate size fractions. The aggregates were ground in a mortar and pestle and subsampled to determine total organic C (Snyder and Trofymow, 1984) and total Kjeldahl N (Nelson and Sommers, 1980) by wet oxidation. Total N was quantified using a Lachat flow-injection system (Lachat Instruments, Milwaukee, WI).

The mineral-associated organic matter contained in each aggregate size fraction was separated by dispersing the aggregate soil with hexametaphosphate and passing the dispersed samples through a 53-μm sieve (Cambardella and Elliott, 1992). The material remaining on the sieve consisted of sand and POM; the soil slurry that passed through the sieve contained the mineral-associated organic matter. Water in the slurry was evaporated in a forced air oven at 50 °C and the dried sample was ground with a mortar and pestle and analyzed for total organic C and total Kjeldahl N as described above. The sand content of each aggregate size fraction was determined by weighing the material that was retained on the sieve following dispersion. Since there was a different amount of sand present in each of the aggregate size fractions, some of which was not originally associated with the aggregates, it was necessary to correct the intact aggregate C and N data for sand in order to make comparisons across aggregate size classes (Elliott et al., 1991).

Total interaggregate POM-C and POM-N were estimated by suspending 1-g subsamples of whole soil taken from each of the tillage treatments in 15 mL of sodium polytungstate adjusted to a density of 1.85 g/cm³ (Elliott et al., 1991). The suspended soil was evacuated for 10 min at ~186 kPa to remove entrapped air from the soil pore space. The POM that was not protected within the aggregate structure of the soil (interaggregate POM) floated to the top of the heavy liquid after sitting overnight at room temperature. The interaggregate POM was removed by aspiration, washed several times with deionized water, trapped on a glass fiber filter, and analyzed for organic C and N as described above. Interaggregate POM-C and N was expressed as the total amount of C or N in the interaggregate POM per gram of original soil. Total POM C and N were estimated as described by Cambardella and Elliott (1992). We calculated intraaggregate POM-C and N as the difference between total and interaggregate POM C and N.

The data were analyzed as a split-plot design using the SAS statistical package for analysis of variance (ANOVA-GLM, SAS Institute, 1990). Tillage management was the main plot treatment and aggregate size class and pretreatment (capillary wetted or slaked) the split-plot treatments. Main and interactive effect means were compared using Tukey's honestly significant difference mean separation test with a 0.05 significance level (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Aggregate Size Distributions

The small macroaggregates (250–2000 μm) comprised the highest percentage of the total soil for all management treatments for capillary-wetted soils (Fig. 1a). No-till soil had significantly more material in the 250- to 2000-μm size class relative to the other treatments although native grassland soil had more large macroaggregates (>2000 μm). This suggests that no-till management is effective at improving or maintaining the macroaggregate structure of these grassland soils compared with bare fallow and stubble mulch management.

Aggregate size distributions for slaked soils were different than those obtained for capillary-wetted soils for all treatments (Fig. 1b). Slaking reduced the amount of soil in the large macroaggregate size class to near zero in the cultivated treatments, and significantly reduced the amount of material in the small macroaggregate size class compared with capillary-wetted soil for all the cultivated treatments (Fig. 1a and 1b). Slaking causes considerable disruption of the soil structure as a result of internal forces that arise as water rapidly enters the soil pore volume. Capillary wetting slowly brings the soil to field capacity, which allows trapped air to escape, with minimal disruption of the soil structure. Macroaggregates that survive the disruptive forces of slaking have relatively higher structural stability when compared with macroaggregates that survive capillary wetting (Kemper and Rosenau, 1986). The amount of soil in the small macroaggregate size class from the slaked no-till soil was more than that for the slaked stubble mulch or bare fallow treatments (Fig. 1b). This suggests that no-till soil has greater aggregate stability than stubble mulch or bare fallow soil. The small macroaggregate size fraction in
the native grassland soil was not affected by slaking (Fig. 1a and 1b), suggesting that native grassland soils are more stable than cultivated soils when slaked. These data are in agreement with results reported by Elliott (1986). The microaggregate fraction increased significantly for all treatments with slaking, while only stubble mulch soil showed an increase in the smallest size fraction (Fig. 1a and 1b).

Most of the material in all treatments remained in the microaggregate fraction with slaking and was not reduced to particles <53 μm, suggesting that microaggregates are more stable than macroaggregates. This observation was also reported by Elliott (1986) and supports the idea that macroaggregates are composed of microaggregates (Tisdall and Oades, 1982) that are capable of resisting the disruptive forces of cultivation and slaking.

The macroaggregate size fraction had a smaller percentage of sand than the microaggregate fraction for capillary-wetted soil and were not differences in the sand proportion within the aggregate size fractions across treatments (Table 2). Slaking increased the sand content of the cultivated soil macroaggregates and decreased the proportion of sand in the microaggregates (Table 2). These data support the observation that slaking disrupts the macroaggregate structure in the cultivated soils, leaving the macroaggregates depleted in soil and enriched in sand. The soil was redistributed into the microaggregate fraction, resulting in a smaller proportion of the microaggregate fraction being composed of sand after slaking.

### Organic Carbon and Nitrogen Concentrations

Sand-free organic C and N concentrations for capillary-wetted soil were equal to one another in the three larger aggregate size classes and were always higher than the organic C and N concentration of the <53-μm fraction for all the treatments (Fig. 2a and 3a). There were no significant differences in the sand-free organic C or N concentrations among the three cultivated treatments for any of the aggregate size classes (Fig. 2a and 3a). Native grassland soil generally had significantly higher organic C and N concentrations in the 250- to 2000-μm size class compared with the cultivated soils (Fig. 2a and 3a). In slaked soils, the amount of organic C and N in the 250- to 2000-μm size class was equal for all the treatments, although native grassland had more organic C and N in the >2000-μm size than the cultivated soils (Fig. 2b and 2b). The amount of organic C and N in an aggregate size fraction is a function of how much soil is in that aggregate fraction and the concentration of C and N of that soil. The distribution of organic C (Fig. 4) and N in the aggregates appears to be primarily controlled by the amount of soil present in the aggregate size fraction (Fig. 1) and not by the concentration of organic C and N in the aggregates (Fig. 2 and 3). This effect was especially pronounced in the slaked soils, suggesting that the organic matter content is related to aggregate stability in grassland soils.

Slaking increased the concentration of sand-free organic C in the small macroaggregate size fraction for the cultivated soils (Fig. 2a and 2b). The sand-free organic N concentration in the small macroaggregate size fraction increased with slaking in the no-till and stubble mulch treatments but not in the bare fallow treatment (Fig. 3a and 3b). Recall that slaking reduced the amount of soil in the small macroaggregate size fraction for the culti-

### Table 2. The effect of slaking on the percentage of sand in the aggregate size fractions.

<table>
<thead>
<tr>
<th>Management Treatment</th>
<th>&gt;2000 μm</th>
<th>250-2000 μm</th>
<th>53-250 μm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bare fallow capillary-wetted</td>
<td>38b†</td>
<td>42b</td>
<td>50a</td>
</tr>
<tr>
<td>slaked</td>
<td>70a</td>
<td>62a</td>
<td>41b</td>
</tr>
<tr>
<td>Stubble mulch capillary-wetted</td>
<td>32b</td>
<td>37b</td>
<td>45a</td>
</tr>
<tr>
<td>slaked</td>
<td>74a</td>
<td>49a</td>
<td>37b</td>
</tr>
<tr>
<td>No-till capillary-wetted</td>
<td>28b</td>
<td>33a</td>
<td>45a</td>
</tr>
<tr>
<td>slaked</td>
<td>43a</td>
<td>37a</td>
<td>34b</td>
</tr>
<tr>
<td>Native grassland capillary wetted</td>
<td>30a</td>
<td>34a</td>
<td>44a</td>
</tr>
<tr>
<td>slaked</td>
<td>31a</td>
<td>34a</td>
<td>38a</td>
</tr>
</tbody>
</table>

† Values within an aggregate size and within a management treatment followed by the same letter are not significantly different at P > 0.05 according to Tukey's honestly significant difference mean separation test.
Fig. 2. The effect of management on sand-free aggregate C concentrations for (a) capillary-wetted soil and (b) slaked soil. Values followed by the same uppercase letter within a management treatment and between aggregate size classes are not significantly different at $P > 0.05$ according to Tukey's HSD mean separation test. Values followed by the same lowercase letter within an aggregate size and between management treatments are not significantly different at $P > 0.05$ according to Tukey's HSD mean separation test.

Fig. 3. The effect of management on sand-free aggregate N concentrations for (a) capillary-wetted soil and (b) slaked soil. Values followed by the same uppercase letter within a management treatment and between aggregate size classes are not significantly different at $P > 0.05$ according to Tukey's HSD mean separation test. Values followed by the same lowercase letter within an aggregate size and between management treatments are not significantly different at $P > 0.05$ according to Tukey's HSD mean separation test.

Fig. 4. The effect of management on the amount of organic C contained in the aggregate size classes for capillary-wetted soil. Values followed by the same uppercase letter within a management treatment and between aggregate size classes are not significantly different at $P > 0.05$ according to Tukey's HSD mean separation test. Values followed by the same lowercase letter within an aggregate size and between management treatments are not significantly different at $P > 0.05$ according to Tukey's HSD mean separation test.

The C/N ratios of bare fallow, stubble mulch, no-till, and native grassland intraaggregate POM are shown in Table 5 along with values previously reported for total POM for the four management treatments (Cambardella and Elliott, 1992). For the native grassland soil, the C/N ratio of total POM did not differ from that of intraaggregate POM. For the cultivated soils, however, the C/N ratio was higher for the intraaggregate POM than the total POM, with a trend of higher C/N ratios as the tillage treatments (Fig. 1a and 1b). This suggests that slaking destroys less-stable small macroaggregates in the cultivated soils, leaving behind more-stable small macroaggregates enriched in organic C and N, similar to observations made by Elliott (1986).

However, when the amount of C and N contained in the POM that was trapped inside the small macroaggregates was subtracted from the sand-free C and N content of the small macroaggregates, there were no significant differences in the organic C and N concentrations between slaked and capillary-wetted small macroaggregates (Table 3 and 4). This suggests that the loss of structural stability in the small macroaggregates with slaking is related, either directly or indirectly, to the loss of POM in these soils.
Table 3. The effect of slaking on the mineral-associated organic C concentration of aggregates derived from cultivated and native grassland soil.

<table>
<thead>
<tr>
<th>Management treatment</th>
<th>Total C (mg C g⁻¹ soil)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&gt;2000 µm</td>
</tr>
<tr>
<td>Bare fallow</td>
<td>Bare fallow capillary-wetted slaked</td>
</tr>
<tr>
<td></td>
<td>Bare fallow capillary-wetted slaked</td>
</tr>
<tr>
<td>Stubble mulch</td>
<td>Stubble mulch capillary-wetted slaked</td>
</tr>
<tr>
<td>No-till</td>
<td>No-till capillary-wetted slaked</td>
</tr>
<tr>
<td>Native grassland</td>
<td>Native grassland capillary-wetted slaked</td>
</tr>
</tbody>
</table>

† Values within an aggregate size and within a management treatment followed by the same letter are not significantly different at P > 0.05 according to Tukey's honestly significant difference mean separation test.
‡ Not enough soil for analysis.

intensity decreased (Table 5). Wheat residue has a C/N ratio of 80 (Buyanovsky and Wagner, 1987) and blue grama grass [Bouteloua gracilis (Willd. ex Kunth) Lagasca ex Griffiths], one of the dominant grass species in the native grassland community at our research site, has a C/N ratio of 40 (Clark, 1977). These patterns suggest that no-till and stubble mulch management are accumulating wheat-derived POM within the aggregate structure of the soil, where it may be protected from decomposition. Plowing disrupts the aggregate structure and increases the availability of the wheat-derived POM that was protected within the structure of the soil (Rovira and Greacen, 1957). Tillage also changes the local soil microclimate, which may also lead to increased decomposition of the wheat-derived POM. Stable C isotope data indicated that only 13% of the total POM C in bare fallow soil was wheat derived, while the other management treatments contained a larger proportion of wheat-derived POM (Cambardella and Elliott, 1992). Therefore, bare fallow management appears to stimulate the decomposition of wheat-derived POM, whereas wheat-derived POM may be protected inside macroaggregates in stubble mulch and no-till soil.

Mineral-associated organic C and N concentrations for capillary-wetted bare fallow small macroaggregates were generally lower than for capillary-wetted no-till and stubble mulch small macroaggregates (Fig. 5a and 5b). We hypothesize that plowing during 20 yr of bare fallow management not only destroyed the less-stable macroaggregates that contain the intraaggregate POM, but also disrupted the structure of the more-stable macroaggregates. This hypothesis is supported by the observation that total soil mineral-associated organic C and N was

Table 5. The effect of cultivation on the C/N ratios of total and intraaggregate particulate organic matter (POM).

<table>
<thead>
<tr>
<th>Management treatment</th>
<th>Total POM†</th>
<th>Intraaggregate POM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bare fallow</td>
<td>12±3a</td>
<td>20±9</td>
</tr>
<tr>
<td>Stubble mulch</td>
<td>17±0.5</td>
<td>26±7</td>
</tr>
<tr>
<td>No-till</td>
<td>21±4</td>
<td>60±13</td>
</tr>
<tr>
<td>Native sod</td>
<td>17±2</td>
<td>17±0.4</td>
</tr>
</tbody>
</table>

† Data from Cambardella and Elliott (1992) from same site.
‡ Mean ± one standard deviation of replicate plots.

Fig. 5. The effect of management on aggregate mineral-associated (a) organic C concentrations and (b) organic N concentrations for capillary-wetted soil. Values followed by the same uppercase letter within a management treatment and between aggregate size classes are not significantly different at P > 0.05 according to Tukey's HSD mean separation test. Values followed by the same lowercase letter within an aggregate size and between management treatments are not significantly different at P > 0.05 according to Tukey's HSD mean separation test.
reduced after 20 yr of bare fallow management of these grassland soils (Cambardella and Elliott, 1992).

CONCLUSIONS

Cultivation destroys the macroaggregate structure of grassland soils with a concomitant reduction in soil organic C and N. The reduction in total soil organic matter content has been related to losses of organic C and N from the POM fraction (Tiessen and Stewart, 1983; Cambardella and Elliott, 1992). Therefore, the loss of structural stability in grassland soils that is associated with cultivation may be related, either directly or indirectly, to losses of organic C and N from the POM fraction. Organic C and N losses resulting from cultivation also appear to occur in the more mineral-associated fractions from bare fallow macroaggregates.

Research presented here suggests that reduced tillage management may ameliorate the detrimental effects of intensive agricultural land use. No-till management appears to be especially effective at maintaining or increasing aggregate stability and slowing the decomposition loss of newly incorporated wheat-derived POM C and N. Wood et al. (1991) reported that imposing no-till management on soils previously cultivated for >50 yr rapidly altered the depth distribution of organic C and N and reduced losses of \( \text{NO}_3^- \) to leaching. They also reported a more rapid accumulation of soil organic C compared with soil organic N under no-till management, reflecting inputs of high C/N ratio plant residues. Wheat-derived POM may be an important fraction in these tilled grassland soils for providing nutrients for plant growth. Adoption of sustainable management practices, such as no-till, may promote the maintenance of this important soil organic matter fraction and enhance the fertility of intensively managed soils.

ACKNOWLEDGMENTS

Funding for this research was provided by NSF Grant no. BSR-860591. The authors wish to thank Indy Burke, Steve Corak, and Tim Parkin for their critical review of this manuscript.

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