Technical Report No. 275 MAJOR SOIL ARTHROPODS OF AN OKLAHOMA TALLGRASS PRAIRE

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#### GRASSLAND BIOME

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#### ABSTRACT

Soil arthropods were once thought to occur in small numbers compared to other groups, but new methods of extraction have resulted in increased attention to this group.

Arthropod data were obtained at the International Biological Program Osage Site, Foraker, Oklahoma. Six collections were taken between June 18, 1971, and November 21, 1972, in grazed and ungrazed grassland treatments. Each treatment consisted of two replicates with three quadrats sampled from each. Samples were obtained by a hydraulic-powered probe and a hand probe. They were processed by modified Berlese funnels.

Three Arthropod classes, 13 orders, and 46 families were identified. Majoi groups of arthropods from the grazed and ungrazed treatments were Acarina, Collembola, Diplura, Protura, Symphyla, and Thysanoptera. They were consistently obtained from the 20-50 cm depth, contradictory to the anticipated restriction to the upper 15 cm. The two 1971 collections revealed a probable seasonal distribution while the four 1972 collections were more consistent in numbers and biomass.

An analysis of covariance was run to determine possible differences in the two extraction techniques: a modified Berlese funnel and a high temperaturehumidity gradient cylinder. At best, the two techniques showed a significant difference at the 10% level.

An analysis of variance considered the effect of environmental factors on the numbers of mites. Soil water and root biomass demonstrated no effect on number of mites. There were, however, significant depth differences for springtails, mites, and total microarthropods. Other significant differences occurred in the Date x Treatment interaction as related to total number of mites collected. Although no significant difference was obtained comparing the grazed and ungrazed treatments, total average biomass  $(g/m^2)$  and numbers of arthropods  $(mean/m^2)$ were unexpectedly greater for the grazed treatment.

#### INTRODUCTION

At the time of Waksman's survey of the microfauna of the soil (1927), it had already been noted that certain animal types inhabited the soil in very large numbers. Soil arthropods were once thought to occur in rather small numbers compared to other groups. The development of new methods of extracting animals from the soil, however, has changed this idea and has resulted in increased attention to this group of animals (Salt, Hollick, Raw, and Brian 1948).

This study was conducted to consider methods of quantitative sampling of soil microarthropods of tallgrass prairie. The real economy of nature and the biological value of every species of animal can be appreciated only when the exact number of individuals inhabiting a given surface is known (Dogiel 1924).

For the sake of convenience, the soil fauna may be divided into "microfauna" (protozoa and other minute animals), "meiofauna" (microarthropods and nematodes), and "macrofauna" (larger arthropods, annelid worms, and others). Microarthropods belong to that group of soil animals sometimes designated as "meiofauna" (Murphy 1953).

This study involves an investigation of a grassland ecosystem located at the International Biological Program Grassland Biome Osage Site near Foraker, Oklahoma. The Analysis of Ecosystems program is designed to supply basic information through a comparative study of six major biomes: Grassland, Deciduous Forest, Coniferous Forest, Desert, Tundra, and Tropical Forest. The objectives for studying each biome are: (1) to elucidate productivity, nutrient cycling, energy flow, and other characteristics of ecosystems in a set of distinctive environments; (2) to determine the driving forces, the processes causing transfers of matter and energy among components, and the controlling variables in each biome; (3) to determine the ecosystem response to the natural and man-induced stress appropriate to each biome; (4) to understand the land-water interactions characteristic of each biome; and (5) to synthesize the results of these and previous studies into predictive models of temporal variation, effects of pollutants and of exploitation, stability, and other ecosystem characteristics necessary for resource management in each biome (IBP Newsletter 1971).

The objectives of this study were (1) to list the trophic level of each group of arthropods; (2) to provide information on the role of certain major groups of arthropods in the grasslands; (3) to obtain estimates of the numbers and biomass of below-ground arthropods, and (4) to check the accuracy of the extracting methods employed.

#### LITERATURE REVIEW

Accurate estimates of the numbers of organisms in terrestrial ecosystems are fundamental to the studies of productivity proposed for the International Biological Program. It is not feasible to count all the animals in a small area, so their numbers must be estimated from samples. The statistical problems involved in sampling soil animals of different sizes and distributions are discussed in several papers by Murphy (1962). The number of soil samples required depends on the extent of the area to be studied and on the size and amount of aggregation of the animals being studied.

Once enough samples have been taken, the animals must be efficiently extracted from the soil. The efficiencies of the many extraction methods differ with different soil types and habitats.

#### Sampling Apparatus

Soil samples are usually taken with a golf-hole borer or a piece of metal tubing sharpened at one end. It has been suggested, however, that some animals may be killed by compression when the core is forced from such instruments. It is highly desirable to keep the core undisturbed. As a result, more elaborate corers have been developed. The most common design consists of a hardened outer cylinder containing one or more removable plastic or metal rings fitted with a handle. The cutting edge is often made slightly smaller in diameter than the rings which rest on a ledge some distance above the cutting edge. This results in the loss of the deepest part of the soil core, an unimportant difficulty in deep soils but a serious difficulty in shallow ones (Southwood 1966).

Coile (1936) devised two types of cutting cylinders for use in undisturbed forest soils to determine volume-weight, air space, water holding capacity, and wilting percentage.

Alexander and Jackson (1955) made an attempt to modify standard geological techniques for the impregnation and sectioning of friable rocks, so that translucent sections of relatively undisturbed soils could be obtained to observe micro-organisms in their natural environment.

Johnson, Southwood, and Entwistle (1957) used a vacuum sampler for ground and herb strata and surface litter. A fan unit collected samples by means of a small nozzle aided by a powerful stream of air into a flexible tube. The apparatus was initially developed to be correlated with vacuum samples from the air and above ground herbs and trees to attain an overview of complete life cycles.

It has been argued that serious losses can result from compression caused by the use of core samplers (Murphy 1958b). It does appear, on the other hand, that manipulation of the soil core by hand can lead to reduced yields (Macfadyen 1953). O'Connor (1957) used a split corer which eliminated the risk of compressing the sample by forcing it out the corer. The core was enclosed by two aluminum halves and tightened by a clamping ring band. When the band was loosened and the aluminum halves separated, the sample was exposed.

Murphy (1958a) developed the block method of sample removal. It was designed to provide undisturbed samples from compact stony soils where the conventional type of cylindrical sampler would not function efficiently. A core, 5 cm diameter, was cut to the required depth by using a metal cylinder inserted during the preparation of the block. The litter was first removed and then the core was cut with scalpels. It was cut transversely into subsamples, the depth depending upon the dimension of the soil horizons with the maximum depth for each being no greater than 2 cm.

Auerbach and Crossley (1960) developed a stainless steel cylinder beveled at one end to form a cutting edge. Sample holders were made of plastic or aluminum cylinders placed within the steel cylinder. A solid lucite cylinder was placed atop the holders to push the completed sample out of the tool.

Macfadyen (1961a) used a cylindrical sampling tool made of stainless steel containing inserts which held the samples. Each sample container was fitted to an aluminum cannister as soon as the sample was taken to prevent the escape of animals.

Undisturbed samples are difficult to take in soft humus rich situations such as manure. Torne (1962) used a sampler consisting of two concentric tubes each with cutting teeth. The inner tube was pushed down firmly and held stationary, thereby protecting the sample, while the outer one was rotated and

cut through the compost. Fallen leaves and other debris were usually sampled with quadrats such as a metal box with the top and bottom removed and the lower edge sharpened (Gabbutt 1959).

Belfield (1956) designed an apparatus for sampling hard tropical or frozen tundra. It included a removable cutting head and a heavy joining link, which was useful in enabling the sampling tube to be driven in despite iron concretions. Soil samples were removed from the tube by means of a piston arrangement operated by a hydraulic jack.

#### Sample Extraction (Berlese funnels)

Berlese (1905) is credited with being the first biologist to apply heat to animals in soil or litter suspended in a tray over a funnel. He used a water jacket, heated with a gas jet or spirit lamp, around the funnel and the sample was suspended near the top so that as it dried out the animals were forced to leave the medium. The first important change was Krause's (Murphy 1962) modification in which he substituted a heated water bath, placed over and around the sample container, for the hot water jacket of the original apparatus. Tullgren (Murphy 1962) was the first to represent the present-day modification. He used an electric bulb suspended above the tray so as to add the stimulus of light to that of heat to drive the animals downward into the funnel. Christenson (1936) had a similar arrangement, and, in addition, used a steeper sided funnel.

Tragardh (1933) was the first to analyze the funnel process. He considered that dessication (apart from a small negatively phototactic response) was the principle factor involved. He suggested that the fauna moved downwards through the sample ahead of a dessicating front and congregated near the base. When drying out of the latter reached a certain point, a rapid movement out of the sample occurred.

Ford (1937) was one of the first to use small funnels. He mounted a battery of 12 funnels, the heat being supplied by an electrically heated resistance wire placed in a cylinder or chimney resting on each funnel. As a result, he was able to obtain statistically valid estimates of the populations of the several species present in the material. Ford's heating arrangement, however, suggested that light emitted by the heat source was of little importance in the extraction process and later a variety of methods for drying the sample material was developed. Petrova (1940), for example, used an electric heater of hot sand with a metal plate interposed between the heat source and the funnel.

Haarlov (1947) used a Tullgren apparatus to obtain Collembola and Acarina from soil samples and found that greatest emigration occurred during the first hour. He realized the importance of condensed water, which in earlier funnels had collected on the sides of the funnel itself and entrapped the weaker animals. He modified the construction of the funnel accordingly allowing fresh air to circulate in the funnels. He also noted that the maximum temperature in the sample should not rise above that of the localities investigated. This was the first attempt to analyze temperature and humidity distribution in funnels during extraction. Kuhnelt (1950) also was troubled with condensed water and surrounded the outer wall with wood-wool or other insulating material to minimize the formation of water droplets on the inner surface of a glass funnel. Another advancement was Hammer's (1944) argument that soil should be inverted and placed in the funnel without being disturbed and teased apart on the grounds that handling killed the more delicate animals and obstructed the passages through which the remainder would move to the lower surface of the soil mass.

A readily transportable funnel was sometimes needed for use in the field or in places where electricity was not available. Jacot (1932) and Sauders (1959) described collapsible models. Each sample was dried by sunlight or in a heated room. Macfadyen (1953) developed an expedition funnel which used an independent and reliable source of heat especially for quantitative studies. A small paraffin stove heated a water-bath placed over the funnels. He also emphasized the prevention of water condensation on the funnel walls and the steepening of the temperature gradient within the sample. Perhaps the most unusual funnel arrangement was that described by Heydemann (1958) in which the funnel was placed in the field directly beneath the uppermost soil layer in a space excavated for that purpose. The funnel had a baffle arrangement to prevent soil debris from falling into the collecting vessel.

The split-funnel constructed by Murphy (1958a) was in part an adaptation of Ford's model. The apparatus consisted of a battery of 12 funnels and was particularly suited for handling large numbers of small samples. It incorporated a number of features including steep-sided funnels, thermostatic control of the temperature, and some attempt at providing different environmental conditions above and below the sample; the latter was accomplished through proper ventilation which insured a constant exchange of air preventing dessication of its undersurface in the early stages of extraction. Murphy also emphasized the advantages of increasing the temperature in the latter stages of extraction in order to drive out the more resistant organisms, as suggested by Macfadyen (1953). Paris and Pitelka (1962) noted that it was sometimes helpful to increase air circulation with wide plastic piping.

The elimination of detritus from the collecting vessel made counting and identification of the collected animals an easier process. To overcome this difficulty, Ulrich (Murphy 1962) constructed a subsidiary chamber attached to

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the funnel outlet giving access to the collecting vessel. A cone was placed over the opening forcing the animals to crawl beneath to escape the funnel. Valle (Murphy 1962) employed a multiple funnel apparatus consisting of one large funnel with two smaller ones, placed one within the other, attached beneath. Jacot (1936) first suggested the use of double-sieve device to reduce the loss of sample material followed by a similar method by Newell (1955). Murphy (1955), Dietrick et al. (1959), and Kempson et al. (1963) have all introduced devices to stop the fall of soil into the sample, thereby insuring a cleaner extraction.

Recently, there has been a major departure from the Berlese approach with more attention being given to larger arthropods. Reduction in funnel size has almost eliminated the quantitative sampling of these animal groups. The controlled-draught funnel designed by Macfadyen (1955) was such a modification and consisted of a large funnel for extracting larger animals where the humidity is derived from the sample and is controlled by manipulating the draught passing through the funnel. Different regimes of temperature and moisture were employed depending on the type of sample material, its water content, and the animals being extracted.

Another trend in the departure from the Berlese funnel was the use of a combination of repellent and attractant stimuli, the respective factors being a relatively high temperature and dessication on one hand together with the provision of a moist, cool environment as an attractant. Auerbach and Crossley (1960) developed such a high gradient apparatus, closely resembling Macfadyen's model, which had the advantage of compactness and greater control of the variables (temperature and humidity) which govern extraction efficiency. They did not, however, find significant differences between the high-gradient funnel and the conventional Tullgren extractor.

Kevan (1959) suggested a "hot rod" technique where a heating element was inserted vertically in the center of the sample, and the animals were driven outwards toward the periphery. Macfadyen (1961b) developed two extractors using the repellent-attractant combination. In the high gradient cylinder extractor, temperature and humidity were controlled by an electric heater placed above and a cold water-bath beneath the samples. The replacement of the funnel by a tube overcame the problem of animals being trapped on the sides of the funnel. The second model was an air-conditioned funnel for use with soils and litter where an undisturbed sample could not be easily obtained. Here, cool air was circulated around the funnels and the lower part of the sample containers while the upper part of the apparatus was heated in the usual way.

#### Sample Extraction (Flotation)

The separation of microarthropods by flotation involves the separation of arthropods, plant debris, and mineral soil particles. First, the mineral soil is separated from the organic matter (animals and plant debris) by using different specific gravities. Next, the microarthropods are separated from plant material by differential wetting, the plant material being wetted by water whereas the waxy arthropod cuticle is not.

Flotation methods were initially developed for the sampling of wireworm larvae and other insects. However, Salt and Hollick (1944) and others have since analyzed their microarthropod catch from the same method and have come to the conclusion that this method extracts at least as high a proportion of the microarthropods as funnels do.

Flotation was first introduced by Berlese in a simple apparatus that was described and tested by Balogh (Cragg 1962); it consisted of a boiling tube

with a constricted neck and plunger. Material from the funnel was mixed with a salt solution by agitation in the tube. After animals had floated to the surface, the plunger was raised and the surface solution poured off, leaving mineral matter behind.

The first of a series of much more elaborate techniques for soil arthropods was designed by Ladell (1936). The flotation vessel was a cylindrical container with a conical lid. The soil sample was supported on a sieve placed at the bottom of this container. The sieve was attached to a hollow metal shaft which was connected by a belt and pulley to an electric motor. In addition, compressed air was passed through this tube and entered the liquid in the vessel through horizontal tubes attached to the central shaft beneath the sample sieve. Soil was placed in position and the flotation vessel was filled with a solution of magnesium sulphate. Agitation of the sample sieve and aeration helped to release the animals, which rose to the surface of the liquid together with other organic material. When extraction was completed more solution was added, and eventually the floating material passed into a settling tank, also filled with a magnesium sulphate solution, to remove the smaller soil particles. From there, the catch passed on to a filter-paper in a Buchner funnel where the surplus liquid was removed. Edwards and Dennis (Murphy 1962) described a modified apparatus for extraction of Symphyla from the soil using a sodium chloride solution.

Strickland (1945) designed a somewhat modified apparatus but omitted the device for agitating the sample. Soil samples were placed in individual glass jars of a half saturated sodium chloride solution and allowed to deflocculate. After soaking 20 to 24 hours, the samples were placed in the extraction apparatus and covered with a salt solution. An air compressor percolated the soilsalt solution mixture which disentangled the arthropod fauna and vegetable debris from the soil and carried them to the surface of the solution. The sediment settled following compression. More salt solution was added, floating the vegetation and fauna on to a double thickness piece of muslin. The material was then washed into a beaker of water and allowed to boil, which caused the vegetation to sink to the bottom. Kerosene was added and the contents were well-stirred. All those animals with chitinous exoskeletons were taken up by the kerosene layer and easily removed from the muslin stretched taut over a photographic plate.

In the Salt and Hollick (1944) apparatus, the soil sample was broken by a water jet and washed through sieves. The animals and plant debris were then floated to the surface of a magnesium sulphate solution, skimmed off, shaken with xylene and water, and hand-picked from the upper xylene layer. Its principal advantage over the original flotation technique was that it facilitated the removal of the arthropods from the organic debris by introducing a separation stage using xylene or benzene.

Raw (1956a) refined the Salt and Hollick Apparatus to extract the minutest members of the arthropod soil fauna. This involved a pre-treatment under reduced pressure of the gently crumbled soil sample with a solution containing 50 gm sodium hexametaphosphate and 20 gm sodium bicarbonate per liter, and freezing the benzene containing the arthropods. The former process caused good dispersal of the soil so that few, if any, arthropods were left within small particles, and the latter insured easier handling, since one could extract the arthropods simply by placing the frozen benzene plug in a sintered glass filtration crucible.

Edwards and Heath (1964) developed a mechanized Salt and Hollick apparatus. Polyethylene cylinders, similar to those in the Salt and Hollick method, contained mesh baskets that held the samples. Each cylinder rotated while a

high pressure flat fan jet of water broke up the soil. A second flat jet prevented silt from blocking the sieve at the base of the cylinder. After 20 to 30 minutes, only the larger soil particles and organic matter remained, and these were separated as in the Salt and Hollick method.

Literature on the role of major soil arthropods in tallgrass prairie soil is found in Appendix I.

#### MATERIALS AND METHODS

#### The Study Area

The study site, the K. S. Adams Ranch, was located in Osage County in the northeast corner of Oklahoma. It is a beef ranch operated by Mr. Dick Whetsell, Pawhuska, Oklahoma. Elevation is 1250 feet with mostly rolling topography. Average temperature is 2.4 C in January and 27 C in July. Average annual precipitation is 36.6 inches with 25.0 inches occurring during April to September. The growing season is about 205 days in length.

The soil is a Brunizem of the Labette-Summit-Sogn association. These are dark colored soils mostly with clay subsoils developed on shales, sandstones, and limestones under tallgrass. Specifically, the control area is on a Labette soil with a dark silty clay A Horizon 0-16". The B<sub>1</sub> is dark-brown 16-23", B<sub>2</sub> is reddish-brown 23-32", B<sub>3</sub> is a brown silty clay 32-42", and most of the bedrock is limestone at three to six feet (Risser 1970).

The experimental design included two treatments. The ungrazed control was a 12.6 acre (150 by 330 meters) rectangle which had been ungrazed for approximately 15 years. The adjacent grazed area was lightly to moderately winter-grazed from mid-October to mid-May, and no grazing occurred during the invertebrate collecting season. Neither the grazed nor ungrazed treatments had been burned or subjected to other major disturbance for a number of years. Each treatment had a 15 to 20 acre pond within 1200 to 1300 feet of the collecting areas.

The major grass species in both treatments were <u>Andropogon scoparius</u> (little bluestem) and <u>Panicum virgatum</u> (switchgrass) followed in importance by <u>Sporobolus asper</u> (tall dropseed), <u>Sorghastrum nutans</u> (Indiangrass), <u>Andropogon</u> <u>gerardi</u> (big bluestem), <u>Bromus Japonicus</u> (brome grass) and <u>Poa pratensis</u> (Kentucky bluegrass). The plant species and frequency data for 1971 are listed in Table 1.

#### Collections

Collections were taken bimonthly from May 16, 1972 through November 21, 1972. Only two collections were taken in 1971, however, as compared to four collections in 1972. Collecting methods for 1972 corresponded closely to the methods in IBP Technical Report No. 85 (French 1971). An additional method ~ was used during 1972 as an efficiency measure and for the purpose of having a more complete soil faunal review. A total of six cores per treatment for each sampling method was taken on all collection dates (Table 2).

Each treatment (grazed and ungrazed) was placed on a grid system which allowed a predestined position for each quadrat to be sampled. Each had two replicates with five quadrats per replicate. The grid was approximately 150 by 330 meters. The quadrats were approximately 15 meters apart. Samples were taken in an undisturbed region within the quadrats. Collections for soil arthropods were only taken from the first three quadrats of each replicate. Litter and crowns had been previously removed.

One of the sampling methods involved the use of power probe mounted in an upright position on the back of a pickup (Fig. 1). The services and

| Species                                  | Grazed | Ungrazed |
|------------------------------------------|--------|----------|
| 0,00,00                                  |        | j        |
|                                          |        |          |
| Andropogon scoparius Michx.              | 74     | 100      |
| Panicum scribnerianum Nash.              | 70     | 100      |
| Sorghastrum nutans (L.) Nash.            | 30     | 40       |
| Sporobolus asper (Michx.) Kunth          | 76     | 38       |
| Panicum virgatum L.                      | 62     | 36       |
| Carex spp. L.                            | 2<br>4 | 32       |
| Psoralea tenuiflora Pursh.               | 4      | 32       |
| Bouteloua curtipendula (Michx.) Torr.    | 4      | 28       |
| Bromus japonicus Thunb.                  | 90     | 26       |
| Leptoloma cognatum (Schult.) Chase       | 44     | 24       |
| Agrostis hiemalis (Walt.) B.S.P.         | 10     | 22       |
| Vernonia baldwini Torr.                  | 4      | 22       |
| Ruellia humilis Nutt.                    | 30     | 22       |
| Andropogon gerardi Vitman                | 20     | 20       |
| Aster ericoides L.                       | 0      | 18       |
| Salvia azurea Lam.                       | 0      | 14       |
| Oxalis stricta L.                        | 16     | 14       |
| Croton capitatus Michx.                  | 18     | 12       |
| Strophostyles leiosperma (T. & G.) Piper | 10     | 8        |
| Amorpha canescens Pursh.                 | 2      | 8        |
| Schrankia nuttallii (DC.) Standley       | 0      | 4        |
| Poa pratensis L.                         | 14     | 4        |
| Ambrosia psilostachya DC.                | 50     | 4        |
| Coreopsis grandiflora Hogg.              | 0      | 4        |
| Achillea lanulosa Nutt.                  | 18     | 4        |
| Solidago missouriensis Nutt.             | 0      | 2        |
| Euphorbia supina Raf.                    | 20     | 2        |
| Gallum texense Grav                      | 0      | 2<br>2   |
| Physalis pumila Nutt.                    | 0      | 2        |
| Bouteloua gracilis (H.B.K.) Lag.         | 0      | 2        |
| Nemastylis geminiflora Nutt.             | 0      | ō        |
| Baptisia leucophaea Nutt.                | 0      | õ        |
| Lespedeza stipulacea Maxim.              | 26     | Ō        |
| Aristida oligantha Michx.                | 24     | 0        |
| Setaria viridis (L.) Beauv.              | 12     | Ō        |
| Desmodium illinoiense Gray               | 6      | 0        |
| Medicago lupulina L.                     | 2      | ō        |
| Euphorbia corollata L.                   | 4      | õ        |
| Andropogon virginicus L.                 | 2      | õ        |
| Andropogon saccharoides Swartz           | 2      | õ        |
|                                          | —      | 5        |

Table 1. Species list<sup>\*</sup> and frequency data of vegetation for 1971, Osage Site.<sup>\*\*</sup>

Table 1 (concluded).

| Species                                | Grazed | Ungrazed |  |
|----------------------------------------|--------|----------|--|
|                                        |        |          |  |
| Lactuca ludoviciana (Nutt.) DC.        | 2      | 0        |  |
| Elymus canadensis L.                   | 2      | 0        |  |
| Gutierrezia dracunculoides (DC.) Blake | 4      | 0        |  |
| Tridens flavus (L.) Hitchc.            | 6      | 0        |  |
| Muhlenbergia sobolifera (Muhl.) Trin.  | 2      | Ō        |  |
| Petalostemum purpureum (Vent.) Rydb.   | 2      | Ō        |  |

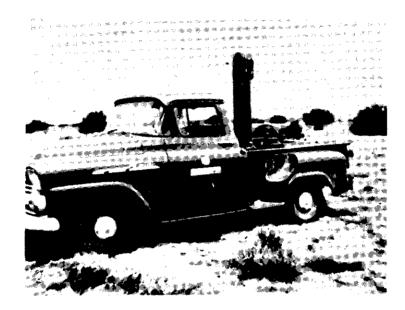
\*Represents 90-95% of the species present.

\*\* Data supplied by Dr. Paul G. Risser, University of Oklahoma

| 1971    | 1972 |          |    |
|---------|------|----------|----|
| June    | 18   | Мау      | 16 |
| October | 10   | July     | 5  |
|         |      | August   | 8  |
|         |      | November | 21 |

Table 2. Soil arthropod samples taken at the Osage Site, 1971 and 1972.\*

\*Treatments were ungrazed and grazed with 2 replicates each, 5 quadrats per replicate.

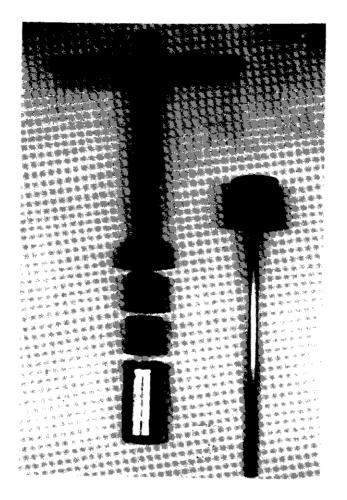


### EXPLANATION OF FIG. 1

Pickup truck employed by the Soil Conservation Service of Oklahoma with encased double roller chain and 60 cm steel power probe attached laterally. equipment were made available by Mr. Bob G. Bourlier, Soil Scientist, and the Soil Conservation Service of Oklahoma. The probing machine consisted of a hydraulic cylinder approximately 60 cm long with the lower end attached to a double roller chain extending over a double sprocket on the upper end, down the front, and under a double sprocket on the lower end of the machine. The hydraulic cylinder was powered by an oil pressure pump. A probe adapter was mounted to the chain on the front near the upper part. When the probe was connected to the adapter, it extended downward in a vertical position and was driven into the soil as the chain moved down the face of the machine. The tubular steel probe was 120 cm long and 5 cm in diameter with a 3 cm wide slot cut lengthwise from the front of the probe for view and removal of the soil core. The tip of the probe, or bit, was tempered steel with a 4.5 cm opening and was threaded on one end for attachment to the probe. The upper end of the probe was solid steel 2.5 cm in diameter with protruding ears for the attachment to the adapter.

The soil samples taken with the power probe extended to a depth of 50 cm. The core was then subdivided into depths of 0-5 cm, 5-10 cm, 10-20 cm, and 20-50 cm. Each was placed into one-quart freezer bags. Identification labels giving treatment, replicate, and quadrat were placed with each sample. Bags were tied and placed in a large Coleman ice chest which was lined with industrial plastic bags to prevent samples from becoming damp as the ice melted. A 25-pound block of ice adequately cooled the ice chest, prevented drying of the samples, and reduced invertebrate activity.

The second sampling method, as designated by IBP, involved the use of a hand probe (Fig. 2). It consisted of a steel bar 38 cm long and 2.5 cm in diameter. This was enlarged and threaded at the distal end to provide for the attachment of a steel probe 11 cm long and 5.5 cm in diameter. The probe was



#### EXPLANATION OF FIG. 2

Hand probe prescribed by IBP; Metal "T" handle; two 5 cm aluminum cylinders, 11 cm steel probe, and metal rod plunger. adapted for the incorporation of two aluminum sleeves, 5 cm long and 5 cm in diameter. The proximal end consisted of a steel cylinder with an attached rod that glided within the steel bar and a "T" handle to aid in driving the apparatus into the soil.

The aluminum sleeves were removed following a sampling. Each sleeve, representing depths of 0-5 cm and 5-10 cm, was placed in one-quart freezer bags with identification labels. The samples were handled and stored as previously mentioned.

#### Processing of the Samples

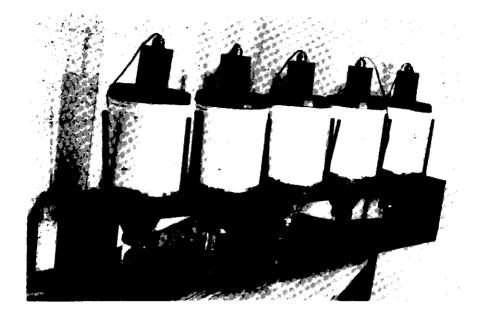
Samples were returned to the laboratory and placed in two types of Berlese funnels: one for power probe samples and the other for hand probe samples. The cores remained intact and were kept in the funnels for one week. Invertebrates were collected in 70 per cent isopropyl alcohol, and the core was then removed and saved for further analysis. Soil cores not immediately placed in the funnels were stored temporarily in a refrigerator held at 5°C.

Adult insects were generally identified to family whereas immatures and non-insects were identified only to order. Families were kept separate in four dram vials so they could be weighed by treatment; weighing per quadrat or replicate was not feasible due to the small amount of biomass. Numbers of families were recorded by quadrat, however.

Specimens were oven-dried at 60°C for 24 hours and weighed on a Mettler Balance to ten-thousandths accuracy.

#### <u>Construction of the Berlese Funnels (Power Probe Samples)</u>

Modified funnels (Fig. 3) were constructed from galvanized cans 30 cm in height with the bottom removed. A tin reducing cone was soldered to the



# EXPLANATION OF FIG. 3

Modified Berlese extractors showing general construction and method of suspension; four such units were used, thus twenty samples could be processed at once. bottom of the cylinder. A pint Mason jar 11d was soldered to the apex of the reducing cone so that the jars could be attached and removed quickly. The inside diameter of the cylinder was 26 cm.

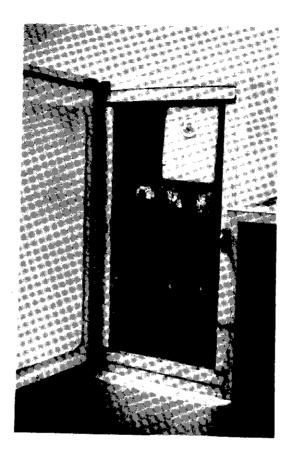
The lid was converted into the heat source by cutting a circle in the center and soldering an oil can to the top of the lid. A circle was then cut in the top of the oil can, and a light fixture was soldered in the opening. A 60 watt bulb was used to supply the heat.

Two wire screens of .7 cm and .1 cm mesh hardware cloth were used in the cylinder to trap large soil particles. The top screen could be removed to clean the second screen or for removing the samples. The sides of the bottom screen were soldered to the bottom of the cylinder, and a small opening was cut in the center to allow invertebrates to escape. Twenty funnels were used while the other samples were temporarily stored. Funnels were set in wooden frames that held five funnels each (Fig. 3) (Reed 1972).

## Construction of the Berlese Funnels (Hand Probe Samples)

The second set of funnels (Fig. 4) employed a temperature and humidity gradient within an undisturbed soil sample (Merchant and Crossley 1970).

Two pieces of plywood 2 cm thick were cut to proper size to fit on refrigerator shelves. Two pieces of .4 cm mesh hardware cloth were cut in the same dimensions as the plywood baffle. In order to accomodate the soil cores within their aluminum retaining sleeves, 9 holes were cut 5.5 cm in diameter in the plywood baffle. Holes were 5 cm apart and arranged in three rows. The funnels were constructed of 345 gm beverage cans 12.5 cm in height with both ends removed. A string of lights was attached to the beverage cans so that each had a light centered over a hole in the baffle. The cans were kept in place by angle braces and .5 cm wood and metal screws. A piece of aluminum foil was



# EXPLANATION OF FIG. 4

Merchant and Crossley inexpensive, high efficiency Tullgren extractor prescribed by IBP; nine funnels on each plywood baffle to allow for the processing of eighteen samples. placed over the light end of each can to serve as a reflector. Strips of 1.5 cm plywood were then attached to the lower side of the baffle to act as shims to allow the soil core to protrude below the baffle.

The extractor was prepared by placing the hardware cloth on the refrigerator shelf, and the plywood baffle, with cans, was placed on top of the hardware cloth. Reducing cones, 6.5 cm powder funnels, were attached with rubber bands to the underside of the refrigerator shelf and aligned with the holes in the baffle. Five dram vials containing 70 per cent isopropyl alcohol were fitted around the mouth of each funnel. Two refrigerator shelves containing 18 funnels were used (Fig. 4) while the remaining samples were temporarily stored in a refrigerator held at 5°C.

#### Efficiency Check of the Berlese Funnels

Berlese funnel extraction efficiency was checked to determine if all invertebrates were being removed. A flotation method was used on several samples after they were removed from the funnels. The soil core was placed in a pint Mason jar filled with water. A teaspoon of kerosene was added and mixed for 2 to 3 minutes. When most of the plant material had settled to the bottom of the beaker, the kerosene phase containing the invertebrates was removed and placed in a Petri dish containing 70 per cent isopropyl alcohol for examination. This efficiency check was employed for one collection only. Due to low numbers and inadequacy of the settling of vegetation, the process was abandoned.

#### **RESULTS AND DISCUSSION**

To this point, 3 Arthropod classes, 13 orders, and 46 families have been identified (Table 3). The total number of acarine families is relative as it was a result of only two collections in 1972. Specialists have determined 25 genera of mites (Table 4).

| Order               | Family                 | Trophic Level                     |
|---------------------|------------------------|-----------------------------------|
| Acarina             | Ameroseiidae           | Fungivore                         |
|                     | Ascidae                | Predator                          |
|                     | <b>Parholaspididae</b> | Predator                          |
|                     | Laelapidae             | Predator                          |
|                     | Rhodacar i dae         | Predator                          |
|                     | Oplitidae              | Myrmecophilore                    |
|                     | Trachyuropodidae       | Myrmecophilore                    |
|                     | Pyemotidae             | Unknown                           |
|                     | Cunaxidae              | Predator                          |
|                     | Trombidiidae           | Predator                          |
|                     | Eniochthoniidae        | Undetermined                      |
|                     | Epilohmanniidae        | Undetermined                      |
|                     | Euphthiracaridae       | Undetermined                      |
|                     | Lohmanniidae           | Undetermined                      |
|                     | Nanhermanniidae        | Undetermined                      |
|                     | Nothridae              | Undetermined                      |
|                     | Opiidae                | Undetermined                      |
|                     | Eremobelbidae          | Undetermined                      |
|                     | Oribatulidae           | Undetermined                      |
|                     | Haplozetidae           | Undetermined                      |
|                     | Ceratozetidae          | Undetermined                      |
|                     | Galumnidae             | Undetermined                      |
| raneida             |                        | Predator                          |
| heloneth <b>ida</b> |                        | Predator                          |
| ymphyla             |                        | Herbivore, Scavenger              |
| oleoptera           | Carabidae              | Predator, Herbivore               |
|                     | Curculionidae          | Herbivore                         |
|                     | Dermestidae            | Scavenger                         |
|                     | Elateridae             | Predator                          |
|                     | Nitidulidae            | Herbivore                         |
|                     | Staphylinidae          | Predator                          |
| ollembola           | Entomobryidae          | Horbiyora Securren                |
|                     | Onichiuridae           | Herbivore, Scavenger<br>Scavenger |
|                     | Poduridae              |                                   |
|                     | Sminthuridae           | Herbivore, Scavenger              |
| _                   |                        | Scavenger                         |
| İptera              | Cecidomyidae           | Herbivore                         |
|                     | Immatures              | Undetermined                      |
|                     | Psychodidae            | Undetermined                      |
|                     | Sciaridae              | Herbivore                         |
| emiptera            | Anthocoridae           | Durada ta m                       |
| en record           | Miridae                | Predator                          |
|                     | Pentatomidae           | Herbivore                         |
|                     |                        | Herbivore, Predator               |
|                     | Scutelleridae          | Herbivore                         |

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## Table 3. Arthropod orders and families determined from Osage Site from June 18, 1971, through November 21, 1972.<u>a,b,c</u>/

Table 3 (concluded).

| Order        | Family                               | Trophic Level                       |  |
|--------------|--------------------------------------|-------------------------------------|--|
| Homoptera    | Aphididae<br>Coccidae<br>Delphacidae | Herbivore<br>Herbivore<br>Herbivore |  |
| Hymenoptera  | Formicidae                           | Scavenger, Predator                 |  |
| Lepidoptera  |                                      | Herbivore                           |  |
| Orthoptera   | Blattidae                            | Scavenger                           |  |
| Protura      |                                      | Scavenger                           |  |
| Psocoptera   |                                      | Scavenger                           |  |
| Thysanoptera | Phloeothripidae<br>Thripidae         | Herbivore, Predator<br>Herbivore    |  |

<sup>a</sup>All orders were not determined to family.

<sup>b</sup>All immatures were determined to order.

<sup>C</sup>Acarina classification by Dr. Donald E. Johnson, Ohio State University.

Table 4. Acarina collected from the Osage Site, August 8 and November 21, 1972.<sup>a</sup>

#### MESOSTIGMATA

Cohort Gamasina

Ameroseius sp. Antennoseius sp. Cheiroseius sp. Holaspina sp. Hypoaspis sp. 1 Hypoaspis sp. 2 Hypoaspis sp. 3 Rhodacarus sp.

Cohort Uropodina

<u>Oplitis</u> sp. Trachyuropoda sp.

#### PROSTIGMATA

Cohort Tarsonemina

Pygmephorus sp.

Cohort Eleutherengona

Bonzia sp. Cunaxa sp. Cunaxoides sp.

Supercohort Parasitengona

Allothrombium sp.

#### CRYPTOSTIGMATA

Supercohort Oribatei

Hypochthoniella sp. Epilohmannia sp. Thysotritia sp. Lohmannia sp. Masthermannia sp. Nothrus sp. Eremobelba sp. Scheloribates sp. 1 Scheloribates sp. 3 Rostrozetes sp. Trichoribates sp. Galumna sp.

<sup>a</sup>Acarina classification by Dr. Donald E. Johnson, Ohio State University.

All biomass figures  $(g/m^2)$  and numbers  $(mean/m^2)$  of the reported groups were obtained using the power probe furnished by the Oklahoma Soil Conservation Service and the mechanical "T" handle probe prescribed by IBP. Trophic levels of many groups are incomplete due to problems of classifying some insects and the absence of information on others. Many of the immature forms were classified only to order.

Major orders of insects at specific depths (Table 5) were determined for 1971 and 1972. The 1971 collection revealed a seasonal distribution. About 80% of the major soil invertebrate orders Acarina and Collembola were represented at the depth of 0-10 cm for the June 18 collection with 60 to 70% located in the 0-5 cm depth for both treatments. In contrast, only 40 to 55% of the Acarina and only 10% of Collembola were represented at the depth of 0-10 cm for the October 10 collection. The 1972 collection showed no seasonal variation as percentages of Collembola and Acarina were consistently well above the 50% level at the 0-10 cm depth for both treatments. Comparison of the 1971 and 1972 collections indicated that the variation in numbers at different depths was due to physical changes of the soil as effected by temperature and humidity.

Wallwork (1970) stated that the diameter of the soil spaces is an important limiting factor in the depth distribution of the soil Acari with the richest fauna occurring in the top 6 cm of the profile. The rather general distribution of many of the medium-sized and smaller species in the soil profile may be a reflection of their ability to undertake vertical migrations. Wallwork (1959) and Tarras-Wahlberg (1961) reported seasonal movements occurring among the Cryptostigmata and these probably occurred as a response to changing temperature or moisture conditions.

Poole (1963) established that correlations existed between Collembola

|      | Date     | ę                | Treatment | Dept  | th                                               | Order                                                        | Number                                      | Per Cen<br>Total<br>Core           |
|------|----------|------------------|-----------|-------|--------------------------------------------------|--------------------------------------------------------------|---------------------------------------------|------------------------------------|
| June | 18, 1971 | 3, 1971 Ungrazed | 0-5       | cm    | Acarina<br>Collembola<br>Diplura<br>Thysanoptera | 12817.3<br>1358.1<br>339.5<br>254.6                          | 51.9<br>80.0<br>33.3<br>21.4                |                                    |
|      |          |                  |           | 5-10  | <b>C</b> M .                                     | Acarina<br>Collembola<br>Diplura<br>Thysanoptera             | 8997.6<br>169.8<br>339.5<br>763.9           | 36.4<br>10.0<br>33.3<br>64.3       |
|      |          |                  |           | 10-20 | СП                                               | Acarina<br>Collembola<br>Diplura<br>Thysanoptera             | 2886.0<br>169.8<br>339.5<br>169.8           | 11.7<br>10.0<br>33.3<br>14.3       |
|      |          |                  |           | 20-50 | cm                                               |                                                              |                                             |                                    |
|      |          |                  |           | 0-50  | cm                                               | Acarina<br>Collembola<br>Diplura<br>Thysanoptera             | 24700.8<br>1697.7<br>1018.6<br>1188.4       |                                    |
|      |          |                  | Grazed    | 0-5   | cm                                               | Acarina<br>Collembola<br>Diplura<br>Thysanoptera             | 10949.9<br>3225.5<br>169.8<br>509.3         | 69.4<br>79.2<br>16.7<br>100.0      |
|      |          |                  |           | 5-10  | cm                                               | Acarina<br>Collembola<br>Diplura                             | 2037.2<br>594.2<br>594.2                    | 12.9<br>14.6<br>58.3               |
|      |          |                  |           | 10-20 | cm                                               | Acarina<br>Collembola<br>Diplura                             | 2801.1<br>254.6<br>254.6                    | 17.7<br>6.2<br>25.0                |
|      |          |                  |           | 20-50 | cm                                               |                                                              |                                             |                                    |
|      |          |                  |           | 0-50  | cm                                               | Acarina<br>Collembola<br>Diplura<br>Thysanoptera             | 15788.2<br>4074.4<br>1018.6<br>509.3        |                                    |
|      |          |                  | Ungrazed  | 0-5   | cm                                               | Acarina<br>Collembola<br>Protura<br>Symphyla<br>Thysanoptera | 52117.9<br>1103.5<br>254.6<br>84.8<br>169.7 | 44.0<br>9.7<br>27.2<br>6.7<br>33.3 |

Table 5. Number (mean number/m<sup>2</sup>) and per cent at given depths for selected invertebrate groups collected, Osage Site, June 18, 1971, through November 21, 1972.

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| Date | Treatment | Depth    | Order                                                                   | Number                                                    | Per Cen<br>Total<br>Core                     |
|------|-----------|----------|-------------------------------------------------------------------------|-----------------------------------------------------------|----------------------------------------------|
|      |           | 5-10 cm  | Acarina<br>Collembola<br>Symphyla<br>Thysanoptera                       | 14090.5<br>254.6<br>169.8<br>84.9                         | 11.9<br>2.2<br>13.3<br>16.7                  |
|      |           | 10-20 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 28775.2<br>2631.4<br>679.1<br>339.5<br>594.2<br>84.9      | 24.3<br>23.2<br>34.8<br>36.4<br>46.7<br>16.7 |
|      |           | 20-50 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 23512.5<br>7384.8<br>1273.2<br>339.5<br>424.4<br>169.8    | 19.8<br>64.9<br>65.2<br>36.4<br>33.3<br>33.3 |
|      |           | 0-50 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 118496.2<br>11374.3<br>1952.3<br>933.7<br>1273.2<br>509.3 |                                              |
|      | Grazed    | 0-5 cm   | Acarina<br>Collembola<br>Protura<br>Thysanoptera                        | 8573.1<br>594.1<br>254.6<br>424.4                         | 22.2<br>6.7<br>33.3<br>55.6                  |
|      |           | 5-10 cm  | Acarina<br>Collembola<br>Diplura                                        | 6366.2<br>254.6<br>169.8                                  | 16.5<br>2.9<br>3.4                           |
|      |           | 10-20 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 6366.2<br>594.2<br>679.1<br>84.9<br>339.5<br>84.9         | 16.5<br>6.7<br>13.8<br>11.1<br>33.3<br>11.1  |
|      |           | 20-50 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 17316.1<br>7384.8<br>4074.4<br>424.4<br>679.1<br>254.6    | 44.8<br>83.7<br>82.8<br>55.6<br>66.7<br>33.3 |

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## Table 5 (cont'd).

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| Date         | Treatment | Depth    | Order                                                                                                            | Number                                     | Per Cen<br>Total<br>Core              |
|--------------|-----------|----------|------------------------------------------------------------------------------------------------------------------|--------------------------------------------|---------------------------------------|
|              | <u></u>   | 0-50 cm  | Acarina 38621.6<br>Collembola 8827.8<br>Diplura 4923.2<br>Protura 763.9<br>Symphyla 1018.6<br>Thysanoptera 763.9 | <b></b>                                    |                                       |
| 1ay 16, 1972 | Ungrazed  | 0-5 cm   | Acarina<br>Collembola<br>Diplura<br>Protura<br>Thysanoptera                                                      | 3649.9<br>169.8<br>84.9<br>84.9<br>84.9    | 56.6<br>50.0<br>50.0<br>100.0<br>33.3 |
|              |           | 5-10 cm  | Acarina<br>Collembola<br>Thysanoptera                                                                            | 2037.2<br>84.9<br>84.9                     | 31.6<br>25.0<br>33.3                  |
|              |           | 10-20 cm | Acarina<br>Diplura<br>Thysanoptera                                                                               | 509.3<br>84.9<br>84.9                      | 7.9<br>50.0<br>33.3                   |
|              |           | 20-50 cm | Acarina<br>Collembola                                                                                            | 254.6<br>84.9                              | 3.9<br>25.0                           |
| ·            |           | 0-50 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Thysanoptera                                                      | 6451.1<br>339.5<br>169.8<br>84.9<br>254.6  |                                       |
|              | Grazed    | 0-5 cm   | Acarina<br>Collembola<br>Thysanoptera                                                                            | 4159.2<br>339.5<br>84.9                    | 26.3<br>33.3<br>33.3                  |
|              |           | 5-10 cm  | Acarina<br>Collembola<br>Diplura                                                                                 | 4329.0<br>254.6<br>254.6                   | 27.4<br>25.0<br>10.3                  |
|              |           | 10-20 cm | Acarina<br>Collembola<br>Diplura                                                                                 | 2631.4<br>169.8<br>424.4                   | 16.7<br>16.7<br>17.2                  |
|              |           | 20-50 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Thysanoptera                                                      | 4668.5<br>254.6<br>1782.5<br>84.9<br>339.5 | 29.6<br>25.0<br>72.5<br>100.0<br>80.0 |

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# Table 5 (cont'd)

| Date   | Treatment | Depth    | Order                                                                   | Number                                               | Per Cent<br>Total<br>Core            |
|--------|-----------|----------|-------------------------------------------------------------------------|------------------------------------------------------|--------------------------------------|
|        |           | 0-50 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Thysanoptera             | 15788.2<br>1018.6<br>2461.6<br>84.9<br>424.4         |                                      |
| July 5 | Ungrazed  | 0-5 cm   | Acarina<br>Collembola<br>Protura<br>Symphyla                            | 3734.8<br>763.9<br>169.8<br>169.8                    | 38.9<br>64.3<br>28.6<br>100.0        |
|        |           | 5-10 cm  | Acarina<br>Diplura<br>Protura<br>Thysanoptera                           | 2206.9<br>169.8<br>84.8<br>254.6                     | 23.0<br>25.0<br>14.3<br>100.0        |
|        |           | 10-20 cm | Acarina<br>Collembola<br>Diplura<br>Protura                             | 2546.5<br>84.9<br>509.3<br>84.9                      | 26.5<br>7.0<br>75.0<br>14.3          |
|        |           | 20-50 cm | Acarina<br>Collembola<br>Protura                                        | 1103.5<br>339.5<br>254.6                             | 11.6<br>28.7<br>42.8                 |
| •      |           | 0-50 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 9591.7<br>1188.4<br>679.1<br>594.2<br>169.8<br>254.6 |                                      |
|        | Grazed    | 0-5 cm   | Acarina<br>Collembola<br>Diplura<br>Protura<br>Thysanoptera             | 7299.9<br>1273.2<br>254.6<br>339.5<br>169.8          | 36.8<br>46.9<br>10.7<br>26.7<br>66.7 |
|        |           | 5-10 cm  | Acarina<br>Collembola<br>Diplura<br>Symphyla<br>Thysanoptera            | 4753.4<br>84.9<br>254.6<br>84.9<br>84.9<br>84.9      | 24.0<br>3.0<br>10.7<br>50.0<br>33.3  |
|        |           | 10-20 cm | Acarina<br>Collembola<br>Diplura<br>Protura                             | 3649.9<br>169.8<br>424.4<br>84.9                     | 18.4<br>6.2<br>17.9<br>6.7           |

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| Date   | Treatment | Depth    | Order                                                                   | Number                                                  | Per Cent<br>Total<br>Core                    |  |
|--------|-----------|----------|-------------------------------------------------------------------------|---------------------------------------------------------|----------------------------------------------|--|
|        |           | 20-50 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla                 | 4159.2<br>1188.4<br>1433.0<br>848.8<br>848.9            | 20.8<br>43.9<br>60.7<br>66.6<br>50.0         |  |
|        |           | 0-50 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 19862.5<br>2716.2<br>2376.7<br>1273.2<br>169.8<br>254.6 |                                              |  |
| August | Ungrazed  | 0-5 cm   | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla                 | 4074.4<br>1018.6<br>84.9<br>509.3<br>424.4              | 36.9<br>54.5<br>20.0<br>54.5<br>62.5         |  |
|        |           | 5-10 cm  | Acarina<br>Collembola<br>Symphyla                                       | 3225.5<br>169.8<br>84.9                                 | 29.2<br>9.1<br>12.5                          |  |
|        |           | 10-20 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 1782.5<br>169.8<br>169.8<br>254.6<br>84.9<br>169.8      | 16.2<br>9.1<br>40.0<br>27.3<br>12.5<br>100.0 |  |
|        |           | 20-50 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla                 | 1952.3<br>509.3<br>169.8<br>169.8<br>84.9               | 17.7<br>27.3<br>40.0<br>18.2<br>12.5         |  |
|        |           | 0-50 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 11034.7<br>1867.4<br>424.4<br>933.7<br>679.1<br>169.8   | -                                            |  |
|        | Grazed    | 0-5 cm   | Acarina<br>Collembola<br>Diplura<br>Thysanoptera                        | 3395.3<br>1188.4<br>84.9<br>594.2                       | 42.1<br>43.8<br>5.0<br>53.8                  |  |

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| Date        | Treatment | Depth    | Order                                                                   | Number                                                 | Per Cent<br>Total<br>Core                    |
|-------------|-----------|----------|-------------------------------------------------------------------------|--------------------------------------------------------|----------------------------------------------|
|             |           |          | Acarina<br>Collembola<br>Thysanoptera                                   | 848.8<br>594.2<br>169.8                                | 10.5<br>21.9<br>15.4                         |
|             |           | 10-20 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 2546.5<br>679.1<br>169.8<br>169.8<br>84.9<br>169.8     | 31.6<br>25.0<br>10.0<br>40.0<br>20.0<br>15.4 |
|             |           | 20-50 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 1273.2<br>254.6<br>1433.0<br>254.6<br>339.5<br>169.8   | 15.8<br>9.3<br>85.0<br>60.0<br>80.0<br>15.4  |
|             |           | 0-50 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 8063.9<br>2716.2<br>1697.7<br>424.4<br>424.4<br>1103.5 |                                              |
| November 21 | Ungrazed  | 0-5 cm   | Acarina<br>Collembola<br>Diplura<br>Thysanoptera                        | 5772.0<br>848.8<br>84.9<br>3055.8                      | 42.5<br>52.6<br>6.3<br>85.7                  |
|             |           | 5-10 cm  | Acarina<br>Collembola<br>Symphyla<br>Thysanoptera                       | 4159.4<br>424.4<br>169.8<br>84.9                       | 30.6<br>26.3<br>33.3<br>2.4                  |
| ·<br>·      |           | 10-20 cm | Acarina<br>Collembola<br>Diplura<br>Symphyla<br>Thysanoptera            | 2122.1<br>254.6<br>254.6<br>254.6<br>169.8             | 15.6<br>15.8<br>18.7<br>50.0<br>4.8          |
|             |           | 20-50 cm | Acarina<br>Collembola<br>Diplura<br>Symphyla<br>Thysanoptera            | 1527.9<br>84.9<br>1018.6<br>254.6<br>254.6             | 11.3<br>5.3<br>75.0<br>50.0<br>7.1           |

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| Date         | Treatment | Depth Order |                                                                         | Number                                                 | Per Cent<br>Total<br>Core                    |
|--------------|-----------|-------------|-------------------------------------------------------------------------|--------------------------------------------------------|----------------------------------------------|
|              |           | 0-50 cm     | Acarina<br>Collembola<br>Diplura<br>Symphyla<br>Thysanoptera            | 13581.2<br>1612.8<br>1358.1<br>509.3<br>3565.1         |                                              |
|              | Grazed    | 0-5 cm      | Acarina<br>Collembola<br>Protura<br>Symphyla<br>Thysanoptera            | 4923.2<br>424.4<br>84.9<br>84.9<br>84.9                | 49.6<br>41.7<br>50.0<br>16.7<br>7.7          |
|              |           | 5-10 cm     | Acarina<br>Collembola<br>Protura<br>Symphyla<br>Thysanoptera            | 1433.0<br>339.5<br>84.9<br>84.9<br>848.8               | 14.4<br>33.3<br>50.0<br>16.7<br>76.9         |
|              |           | 10-20 cm    | Acarina<br>Collembola<br>Diplura<br>Symphyla                            | 2122.1<br>84.9<br>1103.5<br>84.9                       | 15.6<br>8.3<br>37.1<br>16.7                  |
|              |           | 20-50       | Acarina<br>Collembola<br>Diplura<br>Symphyla<br>Thysanoptera            | 1103.5<br>169.8<br>1867.4<br>254.6<br>169.8            | 11.2<br>16.7<br>62.9<br>50.0<br>15.4         |
|              |           | 0-50 cm     | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 9931.3<br>1018.6<br>1970.9<br>169.8<br>509.3<br>1103.5 |                                              |
|              |           | IBP Separ   | rator                                                                   |                                                        |                                              |
| July 5, 1972 | Ungrazed  | 0-5 cm      | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 8403.4<br>1358.1<br>169.8<br>594.2<br>594.2<br>509.3   | 83.9<br>76.2<br>50.0<br>70.0<br>87.5<br>75.0 |

| Date     | Treatment | Depth   | Order                                                                   | Number                                                | Per Cent<br>Total<br>Core                     |
|----------|-----------|---------|-------------------------------------------------------------------------|-------------------------------------------------------|-----------------------------------------------|
|          |           | 5-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 1612.8<br>424.4<br>169.8<br>254.6<br>84.9<br>169.8    | 16.1<br>23.8<br>50.0<br>30.0<br>25.0<br>25.0  |
|          |           | 0-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 10016.2<br>1782.5<br>339.5<br>848.8<br>679.1<br>679.1 |                                               |
|          | Grazed    | 0-5 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 8318.5<br>1188.4<br>169.8<br>848.8<br>254.6<br>84.9   | 70.6<br>53.8<br>66.7<br>83.3<br>75.0<br>100.0 |
|          |           | 5-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla                 | 2886.0<br>1018.6<br>84.9<br>169.8<br>84.9             | 29.4<br>46.2<br>33.3<br>16.7<br>25.0          |
|          |           | 0-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 11204.5<br>2206.9<br>254.6<br>1018.6<br>339.5<br>84.9 |                                               |
| August 8 | Ungrazed  | 0-5 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 6366.2<br>2291.8<br>84.9<br>763.9<br>679.1<br>84.9    | 65.8<br>77.1<br>50.0<br>52.9<br>61.5<br>20.0  |
|          |           | 5-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 3310.4<br>679.1<br>84.9<br>424.4<br>424.4<br>339.5    | 34.2<br>22.9<br>50.0<br>41.7<br>28.5<br>80.0  |

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| Date        | Treatment | Depth   | Order                                                                   | Number                                                 | Per Cent<br>Total<br>Core             |
|-------------|-----------|---------|-------------------------------------------------------------------------|--------------------------------------------------------|---------------------------------------|
|             |           | 0-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla<br>Thysanoptera | 9676.6<br>2970.9<br>169.8<br>1443.0<br>1103.5<br>424.4 |                                       |
|             | Grazed    | 0-5 cm  | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla                 | 8148.7<br>1443.0<br>339.5<br>594.2<br>679.1            | 74.2<br>63.0<br>75.0<br>58.3<br>80.0  |
|             |           | 5-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla                 | 2546.5<br>848.8<br>169.8<br>424.4<br>169.8             | 25.8<br>37.0<br>25.0<br>41.7<br>20.0  |
|             |           | 0-10 cm | Acarina<br>Collembola<br>Diplura<br>Protura<br>Symphyla                 | 10695.2<br>2291.8<br>509.3<br>1018.6<br>848.8          |                                       |
| November 21 | Ungrazed  | 0-5 cm  | Acarina<br>Collembola<br>Protura<br>Thysanoptera                        | 7724.3<br>679.1<br>424.4<br>169.8                      | 67.4<br>52.3<br>62.5<br>100.0         |
|             |           | 5-10 cm | Acarina<br>Collembola<br>Protura<br>Symphyla                            | 3734.8<br>594.2<br>254.6<br>84.9                       | 32.6<br>47.7<br>37.5<br>100.0         |
|             |           | 0-10 cm | Acarina<br>Collembola<br>Protura<br>Symphyla<br>Thysanoptera            | 11459.2<br>1273.3<br>679.1<br>84.9<br>169.8            |                                       |
|             | Grazed    | 0-5 cm  | Acarina<br>Collembola<br>Protura<br>Symphyla<br>Thysanoptera            | 5687.1<br>254.6<br>254.6<br>254.6<br>84.9              | 71.3<br>75.0<br>60.0<br>100.0<br>24.3 |

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## Table 5 (concluded).

| Date | Treatment | Depth   | Order                                                        | Number                                     | Per Cent<br>Total<br>Core    |
|------|-----------|---------|--------------------------------------------------------------|--------------------------------------------|------------------------------|
|      |           | 5-10 cm | Acarina<br>Collembola<br>Protura<br>Thysanoptera             | 2291.8<br>84.9<br>169.8<br>509.3           | 28.7<br>25.0<br>40.0<br>85.7 |
|      |           | 0-10 cm | Acarina<br>Collembola<br>Protura<br>Symphyla<br>Thysanoptera | 7979.0<br>339.5<br>424.4<br>254.6<br>594.2 |                              |

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numbers and depth and moisture content of the organic layer in a coniferous woodland. Dhillon and Gibson (1962) stated that in undisturbed grassland soils, where there is no appreciable accumulation of surface litter and no well-defined fermentation zone, Collembola numbers showed a progressive decrease with depth which was related to the decreasing porosity of the soil. Hale (1963) reported that the influence of soil water could best be observed in localities where a gradient in this factor occurred as in common calcareous areas, bogs, and moorland. Pryor (1962) noted the direct influence of the temperature factor on distribution patterns was hard to evaluate, if not meaningless, if not considered in conjuction with the effect of moisture. Christiansen (1964) and Wood (1967) found that in most permanent grassland, moorland, and woodland sites the Collembola fauna were best represented in organic layers of 10-15 cm.

Symphylids were encountered, for the most part, below 10 cm. Edwards (1958) reported that although Symphyla may be encountered in a variety of soils, ranging from cultivated plots to grassland forest litter, they are especially abundant in glasshouses, evidently preferring moist, organic soils of the loam type with open texture. They often occur below the soil surface sometimes at a depth of several cm, although seasonal vertical migrations complicate the pattern of depth distribution. Edwards (1958) also described the migrations of <u>Scutigerella immaculata</u> and <u>Symphylella vulgaris</u>, and correlated these with changes in environmental conditions. He reported that surface layers of the soil became dessicated during the summer when high surface temperatures also occurred, and the combined effect of these two factors evidently provoked a downward migration of the population in the litter. This trend was reversed in the spring and autumn seasons. Wallwork (1970), however, found the <u>Scutigerella</u> <u>immaculata</u> migrated upwards to the surface, even when temperature and moisture conditions were unfavorable, provided suitable food material was available. He noted this species reached pest proportions in glasshouses, where it fed on the root systems of cultivated plants.

In a comparative ecological study involving the genera <u>Scutigerella</u>, <u>Symphylella</u>, and <u>Symphylelopsis</u> in cultivated fields in south-western France, Anglade (1967) demonstrated differences between these three genera in their vertical distribution patterns. <u>Scutigerella</u> and <u>Symphylella</u> were much more abundant in the top 15 cm of the profile than at greater depths. In contrast, the bulk of the <u>Symphylellopsis</u> population was generally located lower down the profile at a depth of 15-45 cm. These differences may be explained, in part, by differences in feeding habits; <u>Scutigerella</u> is phytophagous while the other two are saprophagous.

In several surveys of soil arthropods, symphylids were the most numerous myriapods present (Salt et al. 1948; Edwards 1958). Michelbacher (1949) stated that they were widely distributed through temperate and tropical regions and occurred in both cultivated and uncultivated soils, being particularly abundant in warm, moist organic soils.

Edwards (1958) sampled 415 sites in southern England and found symphylids in 46% of them. Of the various habitats sampled, they were found in 32% of the grassland sites, 26% of the forest litter sites, 44% of the cultivated soils, and 53% of the grassland soils. <u>Symphylella</u> spp. were more widespread than <u>Scutigerella</u> spp. Loams appeared to be the most favorable soil type, with clay loams and sandy loams intermediate, and sandy loams, sands, and clays the least favorable. These would be soils with open texture, good moisture holding capacity, and high organic matter content.

Raw (1967) reported that symphylids made seasonal vertical migrations in soil in response to changes in soil water and temperature, and to=feeding, molting, and oviposition cycles.

Representatives of Protura and Thysanoptera were found, as could be expected, in the upper 10 cm.

Members of Diplura were restricted to Japygidae. At least 60% of the total japygids for each treatment and collection were found at the depth of 20-50 cm. The decrease in soil porosity would seem to handicap the predaceous japygid in regard to food availability. The distribution must therefore be related to a high and stable moisture regimen, as suggested by Kuhnelt (1961).

Total arthropods according to numbers, biomass, and percentage (Table 6) were determined for 1972. It must be noted that the reliability of certain macroarthropods to be soil inhabitors is questionable, yet they are listed as a matter of reference. Although the extraction technique did not favor either group, microarthropods dominated the grassland soil according to numbers while macroarthropods presumably had the larger biomass.

One of the prime objectives of this paper was to compare the extraction efficiency of two techniques (Table 7). The numbers extracted from the IBP Separator were larger in almost every case as compared to the Regular Berlese Separator. This difference was not significant, however. The most proficient extractor of soil arthropods seems to vary from order to order. To date, the most efficient methods for the extraction of Collembola are Macfayden's (1961b) high gradient cylinder and Raw's (1955) flotation method. The latter method has the disadvantage of being suitable only for mineral soils, but Hale (1963) devised a similar method for use with organic soils. Symphylids are most successfully extracted from the soil by the flotation process described by Edwards (1955). Edwards and Fletcher (1970) also reported no single extraction method or size of sample ideal for all groups of soil

|                                                                          |                                                          | Per Cent                             |                                                | Per Cent                            |  |
|--------------------------------------------------------------------------|----------------------------------------------------------|--------------------------------------|------------------------------------------------|-------------------------------------|--|
|                                                                          |                                                          | of                                   |                                                | of                                  |  |
| Group                                                                    | Number                                                   | Numbers                              | Biomass                                        | Biomass                             |  |
| MICROARTHROPODS                                                          |                                                          |                                      |                                                |                                     |  |
| Acarina                                                                  | 94304.608                                                | 51.8                                 | .52650                                         | 9.0                                 |  |
| Collembola<br>Entomobryidae<br>Onychiuridae<br>Poduridae<br>Sminthuridae | 12478.749<br>1018.593<br>7385.789<br>3819.719<br>254.648 | (6.9)<br>0.6<br>4.1<br>2.1<br>0.1    | .89288<br>.04629<br>.66567<br>.18092<br>.00000 | (15.1)<br>0.8<br>11.3<br>3.0<br>0.0 |  |
| Chelonethida                                                             | 748.030                                                  | 0.4                                  | .11816                                         | 2.0                                 |  |
| Diplura<br>Japygidae                                                     | 11713.803                                                | 6.4                                  | . 59394                                        | 10.0                                |  |
| Homoptera<br>Coccidae                                                    | 3395.305                                                 | 1.9                                  | .26408                                         | 4.5                                 |  |
| Protura                                                                  | 3565.071                                                 | 2.0                                  | .17825                                         | 3.0                                 |  |
| Psocoptera                                                               | 676.409                                                  | 0.4                                  | .00000                                         | 0.0                                 |  |
| Symphyla                                                                 | 2122.066                                                 | 1.2                                  | .20104                                         | 3.4                                 |  |
| Thysanoptera<br>Phloeothripidae<br>Thripidae                             | 7130.142<br>6535.963<br>594.179                          | (3.9)<br>3.6<br>0.3                  | .70860<br>.48790<br>.22070                     | (12.0)<br>8.3<br>3.7                |  |
| MACROARTHROPODS                                                          |                                                          |                                      |                                                |                                     |  |
| Araneida                                                                 | 822.302                                                  | 0.5                                  | . 49338                                        | 8.3                                 |  |
| Coleoptera<br>Carabidae<br>Chrysomelidae<br>Pselaphidae<br>Staphylinidae | 822.302<br>82.230<br>493.382<br>82.230<br>164.460        | (0.5)<br>0.04<br>0.3<br>0.04<br>0.1r |                                                | -                                   |  |
| Diptera<br>Cecidomyidae<br>Immatures<br>Psychodidae<br>Sciaridae         | 1233.451<br>657.841<br>328.920<br>82.230<br>164.460      | (0.7)<br>0.4<br>0.2<br>0.04<br>0.1   |                                                |                                     |  |
| demiptera<br>Miridae<br>Pentatomidae<br>Scutellaridae                    | 328.920<br>82.230<br>164.460<br>82.230                   | (0.2)<br>0.04<br>0.1<br>0.04         |                                                |                                     |  |

Table 6. Number (mean number/m<sup>2</sup>), biomass (g/m<sup>2</sup>), and per cent of total biomass and numbers for total invertebrates collected, Osage Site, May 16, 1972 through November 21, 1972.\*

## Table 6 (concluded).

| Group                                            | Number                              | Per Cent<br>of<br>Numbers | Biomass                      | Per Cent<br>of<br>Biomass |
|--------------------------------------------------|-------------------------------------|---------------------------|------------------------------|---------------------------|
| Homoptera<br>Aphidae<br>Delphacidae              | 246.690<br>164.460<br>82.230        | (0.1)<br>0.1<br>0.04      |                              |                           |
| Hymenoptera<br>Immature Formicidae<br>Formicidae | 41444.030<br>20804.245<br>20639.785 | (22.7)<br>11.4<br>11.3    | 1.93451<br>.45391<br>1.48060 | (32.7)<br>7.7<br>25.0     |
| Lepidoptera<br>Immatures                         | 82.230                              | 0.04                      |                              | -,                        |
| Orthoptera<br>Immature Blattidae                 | 904.532                             | 0.5                       |                              |                           |

\*Only major groups of macroarthropods weighed for biomass.

|              |            | <u>.                                    </u> | NUMBE     | RS        |           |
|--------------|------------|----------------------------------------------|-----------|-----------|-----------|
| <b>D</b> .   | <b>.</b> . | Ungr                                         | azed      | Gr        | azed      |
| Date         | Depth      | IBP                                          | Regular   | IBP       | Regular   |
| July 5, 1972 | 0-5 cm     | 11713.804                                    | 4838.310  | 10864.977 | 9337.090  |
|              | 5-10 cm    | 2801.127                                     | 2886.010  | 4244.132  | 5262.723  |
|              | 0-10 cm    | 14514.931                                    | 7724.320  | 15109.109 | 14599.813 |
| August 8     | 0-5 cm     | 10355.682                                    | 6196.432  | 11289.391 | 5347.606  |
|              | 5-10 cm    | 5687.137                                     | 3565.071  | 4159.249  | 1867.418  |
|              | 0-10 cm    | 16042.818                                    | 9761.503  | 15448.640 | 7215.024  |
| November 21  | 0-5 cm     | 8997.559                                     | 10016.151 | 6535.963  | 5602.254  |
|              | 5-10 cm    | 4838.310                                     | 5092.958  | 3055.775  | 2970.892  |
|              | 0-10 cm    | 13835.870                                    | 15109.109 | 9591.738  | 8573.146  |

Table 7. Comparison of numbers (mean number/m<sup>2</sup>) and biomass (g/m<sup>2</sup>) of total invertebrates obtained by two separation techniques, Osage Site, July 5, 1972, through November 21, 1972.

|              |         | BIOMASS |         |             |         |  |
|--------------|---------|---------|---------|-------------|---------|--|
| De tra       |         |         | razed   | Gr          | azed    |  |
| Date         | Depth   | IBP     | Regular | <b>I</b> BP | Regular |  |
| July 5, 1972 | 0-5 cm  | *.24567 | *.09441 | . 20369     | *.21239 |  |
|              | 5-10 cm | .08061  | .05739  | . 12072     | .04579  |  |
|              | 0-10 cm | .32629  | .15180  | . 32441     | .25818  |  |
| August 8     | 0-5 cm  | .31091  | *.16524 | *.26324     | *.14454 |  |
|              | 5-10 cm | .18565  | *.04069 | *.10727     | .06536  |  |
|              | 0-10 cm | .49656  | .20593  | *.37052     | .20990  |  |
| November 21  | 0-5 cm  | *.11975 | *.34950 | *.08684     | *.07710 |  |
|              | 5-10 cm | *.09750 | *.12442 | *.06695     | *.10342 |  |
|              | 0-10 cm | .21725  | .47392  | *.15380     | .18052  |  |

\*The standard error of the mean dry weight exceeds 25 per cent of the mean.

invertebrates, but of the methods tested, the Macfadyen air-conditioned funnel was usually the most efficient. If this apparatus could not be used, then simpler funnels with a controlled temperature gradient would give reasonable results for most groups of animals. For the larger arthropods, he noted both flotation and funnel methods were satisfactory but flotation methods, which extracted more animals from fallow or arable soils, were more difficult to use with soils containing much organic matter. Except for onychiurid Collembola, most microarthropods were recovered better by funnel than by flotation methods.

A comparison of treatment areas (Table 8) was determined for 1971 and 1972 using the Regular Berlese Separator. The ungrazed treatment showed the higher figures for 1971 while the grazed treatment had the larger figures for 1972. This contradicts the work of Ford (1935), Thompson (1924), and Morris (1920) who have noted that the soil of fields open to grazing cattle had a definitely smaller population than that in fields in the same locality not grazed. Upon investigation of the soil cores and personal communication with agronomists at Kansas State University, however, the effect of grazing may not be significant in mineral soils. The grazed soil core also indicated the greater extension of plant roots which is related to food availability and increased numbers.

Analyses of variance and covariance were run by the statistical collaborators at the Grassland Biome Central Office located at Fort Collins, Colorado considering the effect of environmental conditions and extraction techniques employed upon the number of mites, springtails, and total microarthropods.

An analysis of covariance considered only the number of mites obtained using the Berlese extraction technique. The core depths used were 0-5, 5-10, and 0-50 cm and the sampling dates were 16 May, 5 July, 8 August, and 21

| Date          | Treatment | Depth    | Number     | Per Cent |
|---------------|-----------|----------|------------|----------|
| June 18, 1971 | Ungrazed  | 0~5 cm   | 15278.875  | 49.7     |
|               | -         | 5-10 cm  | 10949.860  | 35.6     |
|               |           | 10-20 cm | 4498.780   | 14.7     |
|               |           | 20-50 cm |            |          |
|               |           | 0-50 cm  | 30727.514  |          |
|               | Grazed    | 0-5 cm   | 14854.461  | 68.1     |
|               |           | 5-10 cm  | 3225.540   | 14.8     |
|               |           | 10-20 cm | 3734.836   | 17.1     |
|               |           | 20-50 cm |            | •        |
|               |           | 0-50 cm  | 21814.838  |          |
| October 10    | Ungrazed  | 0-5 cm   | 53730.709  | 38.9     |
|               | -         | 5-10 cm  | 14599.813  | 10.6     |
|               |           | 10-20 cm | 34547.233  | 25.0     |
|               |           | 20-50 cm | 35396.059  | 25.5     |
|               |           | 0-50 cm  | 138273.815 |          |
|               | Grazed    | 0-5 cm   | 9846.386   | 17.6     |
|               |           | 5-10 cm  | 7045.259   | 12.6     |
|               |           | 10-20 cm | 8233.616   | 14.7     |
|               |           | 20-50 cm | 30727.514  | 55.1     |
|               |           | 0-50 cm  | 55852.775  |          |
| 1ay 16, 1972  | Ungrazed  | 0-5 cm   | 4244.132   | 56.8     |
|               | -         | 5-10 cm  | 2206.949   | 29.5     |
|               |           | 10-20 cm | 679.061    | 9.1      |
|               |           | 20-50 cm | 339.531    | 4.6      |
|               |           | 0-50 cm  | 7469.672   |          |
|               | Grazed    | 0-5 cm   | 4583.662   | 23.0     |
|               |           | 5-10 cm  | 4838.310   | 24.2     |
|               |           | 10-20 cm | 3310.423   | 16.6     |
|               |           | 20-50 cm | 7215.024   | 36.2     |
|               |           | 0-50 cm  | 19947.420  | -        |
| luly 5, 1972  | Ungrazed  | 0-5 cm   | 4838.310   | 36.3     |
|               | -         | 5-10 cm  | 2886.010   | 21.7     |
|               |           | 10-20 cm | 3734.836   | 28.0     |
|               |           | 20-50 cm | 1867.418   | 14.0     |
|               |           | 0-50 cm  | 13326.574  | -        |
|               | Grazed    | 0-5 cm   | 9337.090   | 35.1     |
|               |           | 5-10 cm  | 5262.723   | 19.7     |
|               |           | 10-20 cm | 4329.014   | 16.2     |
|               |           | 20-50 cm | 7724.320   | 29.0     |
|               |           | 0-50 cm  | 26653.148  | -2       |

Table 8. Number (mean number/m<sup>2</sup>) and per cent of total invertebrates at given depths, Osage Site, June 18, 1971, through November 21, 1972.

## Table 8 (concluded).

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| Date        | Treatment | Depth    | Number    | Per Cent |  |
|-------------|-----------|----------|-----------|----------|--|
| August 8    | Ungrazed  | 0-5 cm   | 6196.432  | 40,1     |  |
|             | -         | 5-10 cm  | 3565.071  | 23.1     |  |
|             |           | 10-20 cm | 2801,127  | 18.1     |  |
|             |           | 20-50 cm | 2886.010  | 18.7     |  |
|             |           | 0-50 cm  | 15448.640 |          |  |
|             | Grazed    | 0-5 cm   | 5347.606  | 34.4     |  |
|             |           | 5-10 cm  | 1867.418  | 12.0     |  |
|             |           | 10-20 cm | 3819.719  | 24.6     |  |
|             |           | 20-50 cm | 4498.780  | 29.0     |  |
|             |           | 0-50 cm  | 15533.522 | -        |  |
| November 21 | Ungrazed  | 0-5 cm   | 10016.151 | 45.7     |  |
|             |           | 5-10 cm  | 5092.958  | 23.3     |  |
|             |           | 10-20 cm | 3310.423  | 15.1     |  |
|             |           | 20-50 cm | 3480.188  | 15.9     |  |
|             |           | 0-50 cm  | 21899.720 |          |  |
|             | Grazed    | 0-5 cm   | 5602.254  | 34.9     |  |
|             |           | 5-10 cm  | 2970.892  | 18.5     |  |
|             |           | 10-20 cm | 3734.836  | 23.3     |  |
|             |           | 20-50 cm | 3734.836  | 23.3     |  |
|             |           | 0-50 cm  | 16042.818 | -2.2     |  |

November. Both the ungrazed and grazed treatments, by replicates, were involved in the analysis. In addition, the variables of soil water and root biomass were considered as a function of total mites collected. The covariates showed no significance for any of the analyses. For each analysis, neither the root biomass nor the soil water influenced the number of mites at Osage for 1972. In addition, none of the main effects or interactions showed a significant effect on the number of mites.

Three variables, number of mites, springtails, and total microarthropods, were used under both the Berlese extraction and the IBP extraction techniques as an analysis of variance. The core depths used were 0-5, 5-10, and 0-10 cm and the sampling dates were 5 July, 8 August, and 21 November. Again, both the ungrazed and grazed treatments, by replicates, were involved in the analysis. At best, the two techniques elicited a significant difference at the 10% level in extraction comparison--more specifically for mites and total microarthropods at depths of 0-5 and 5-10 cm. There were, however, significant depth differences for all three microarthropod classifications. Other significances occurred in the Date x Treatment interaction and Technique x Depth interaction as related to the number of mites collected.

#### SUMMARY AND CONCLUSIONS

Invertebrate data were collected at the IBP Comprehensive Osage Site, Foraker, Oklahoma in 1971-1972. Six collections were taken between June 18, 1971, and November 21, 1972 in grazed and ungrazed treatments. Each treatment consisted of two replications of five quadrats each of which only the first three were sampled.

Samples were obtained using a hydraulic-powered probe furnished by the Oklahoma Soil Conservation Service and a hand probe prescribed by IBP.

Modified Berlese funnels were used to extract the invertebrates from the samples Although immatures were classified only to order, most insects were sorted to family, dried at 60 C for 24 hr and weighed. Fifteen orders and 46 families were identified. Major groups in numbers for the grazed and ungrazed treatments were Acarina, Collembola, Diplura, Protura, Symphyla, and Thysanoptera. Major groups according to biomass were Formicidae, Collembola, Diplura, Thysanoptera, Araneida, and Acarina.

Contradictory to previous work, microarthropods were found in large numbers below 20 cm. There proved to be little significance in the comparison of the two extraction techniques statistically although the IBP Separator extracted the greater numbers. There also was Little significance in the comparison of ungrazed and grazed treatments as mineral soils show little compaction from grazing effects. Ironically, figures showed the grazed treatment to have the greater numbers.

Analyses from the study indicated little significance between limiting environmental factors and numbers and biomass. The degree of standard error, for the most part, indicated the need for increased observations over a greater area.

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#### APPENDIX I

#### THE ROLE OF SELECTED INVERTEBRATES IN GRASSLAND SOIL

The activity of soil animals, especially arthropods and annelids, has been studied in forests and agricultural land sufficiently to develop certain principles concerning their relationships to soil fertility. One of these is that the great majority of individuals live in the surface litter; immediately below it, their numbers decline rapidly (Jacot 1936). Another is that since about 80% of the animals feed on the litter and organic residues of it (the remainder being predatory), the most important function they perform is to initiate the disintegration of litter into humus, both mechanically and chemically. Buckle (1921) found that in England the distribution and numbers of soil animals were more stable in grass than in cultivated land because the vegetative cover increased accordingly, and because there was no close correlation between insect species and soil types. The importance of microarthropods in soils and their relations to detritus, other fauna, and microflora, have been reviewed recently by Christiansen (1970), Crossley (1970), and Witkamp (1971).

#### Acarina

Mites are the most numerous of soil arthropods. Most are saprophagous though many are predaceous and parasitic. Other species eat out dead roots and rootlets, thus secondarily channeling the soil. They undoubtedly go as deep as plant rootlets (240 to 430 or more centimeters) provided they do not encounter the water table.

Although the group comprises minute species, they are so generally distributed throughout the mineral soil and so well provided for digging and channeling the soil, that they are the most important group of soil microarthropods (Jacot 1940). Wallwork (1970) included in the predatory Mesostigmata the genera <u>Macro-</u> <u>cheles</u>, <u>Veigaia</u>, <u>Parasitus</u>, <u>Pergamasus</u>, and <u>Gamasolaelaps</u>. Hurlbutt (1964) noted that macrochelids and veigaiids feed on a variety of animals including Collembola, Protura, and Pauropoda and have also been observed to attack cryptostigmatid mites. Singer and Krantz (1967) noted them feeding on nematodes and enchytraeids. Wallwork (1957) has found that the particular diet will undoubtedly depend on what is available. Wallwork (1967) also noted that some of these predators, such as <u>Pergamasus</u> spp., may be cannibalistic when no other food is available.

Members of the mesostigmatid Uropodina are much less active than their predatory counterparts, and, although <u>Fuscuropoda marginata</u> has been shown to prey on the astigmatid mite, <u>Caloglyphus mycophagus</u>, the majority of the group is regarded as scavengers on a variety of decaying plant and animal material (Rohde 1959). Evans et al. (1961) has noted, however, that the Uropodina are largely fungal feeding mites. Kuhnelt (1961) reported another Uropodina, <u>Prodinychus</u> sp., to have predatory tendencies although it is also a carrionfeeder, a common habit among predatory Mesostigmata.

Important predators of the Prostigmata, such as members of the families Bdellidae, Cheyletidae, Rhagidiidae, Erythraeidae, Cunaxidae, and Trombidiidae, have much the same kind of feeding habit as the predatory Mesostigmata (Wallwork 1970). Baker and Wharton (1952) reported predatory activities of Bdellidae, Cheyletidae, Rhagidiidae, Trombidiidae, and Tydeidae. Kuhnelt (1961) noted the family Eupodidae to be predatory while Evans et al. (1961) considered it a fungal feeder. Baker and Wharton (1952) also cited a number of phytophagous species which are agricultural pests in certain countries. Newell (1963) found the genus <u>Balaustuim</u> of Erythraeidae to be an active predator of scale insects, thrips, and other insects associated with vegetation while others were Phytophagous on green leaves or pollen.

Wallwork (1970) reported members of the Cryptostigmata feeding on a variety of decomposing plant material although fungal hyphae and spores form a part of the natural diet of many species such as Ceratoppia bipilis, Adoristes ovatus, Belba spp., Oribatula tibialis, Scheloribates laevigatus, Ceratozetes gracilis, and Galumna elimata. Jacot (1939) and Murphy (1955) noted that the 'box mites' of the families Phthiracaridae and Euphthiracaridae usually feed on decaying leaf and woody tissue. Englemann (1961) found that many species feed on soil fungi and bacteria making them a possible important element in the decomposer food chain. Results of feeding experiments made by Hartenstein (1962) on 20 species indicated that about half of them preferred fungi as food; the others preferred decomposing plant material. Some Cryptostigmata are coprophagous, and this may be of general occurrence among larval stages of phthiracarids that burrow in woody tissue. Animal food was taken by a number of species, although this was more usually in the form of carrion than living prey (Wallwork 1970). A lack of specificity of many species has been noted in culture and gut content studies by Forsslund (1939), Schuster (1956), Wallwork (1957), and Woodring and Cook (1962) although some tentative conclusions were drawn.

Karg (1963) reported the Astigmata to feed on plant detritus, fungi, algae, and the liquified products of putrefaction processes. The <u>Glycypha-</u> <u>gidae</u> and <u>Histiogaster</u> species (Burgess and Raw 1967) are considered to be mainly fungal feeders. Woodring (1963) recorded this genus as a carrionfeeder. Karg (1963) noted many of the Anoetidae to be filter feeders on liquids rich in micro-organisms. Burgess and Raw (1967) reported the probability that soil astigmata require organic material in an advanced state of decay, low oxygen concentration, and a plentiful supply of moisture. As a role of decomposer in breakdown processes occurring in meadow soil, Macfadyen (1963) calculated that Acarina attracted considerably more of the energy flow than the herbivore/carnivore food chain. It has been suggested that mites assist the decomposition processes by promoting the growth and distribution of micro-organisms and fungi and by transporting the products of decomposition to the lower layers and root zone of the soil profile (Burgess and Raw 1967). Ghilarov (1963) and Kuhnelt (1963) suggested that the symbiotic relationship between mites and micro-organisms may be associated with decomposition, because there is evidence that litter or decomposing material may be more susceptible microbial activity after passing through the gut of mites. Macfadyen (1961a), however, noted that the "wasteful" feeding behavior of the Cryptostigmata may in fact be the reverse.

#### Collembola

The majority of soil Collembola are saprophagous, living on the humus in thick, uncut and damp grass that would be plentiful. MacLagan (1933) remarked that the role of Collembola in soil formation has been underestimated, it being next in importance to that of earthworms. He also noted that certain species, <u>Synthurus viridis</u>, for example, were phytophagous and appeared to be most active at night.

A strong feeding preference for fungal hyphae and spores has been shown by some Collembola, and perhaps the distribution is influenced by the distribution of fungi on which they feed (Knight 1961). Poole (1959) found differences within the species <u>Folsomia quadrioculata</u>, some individuals preferring fungi, others selecting higher plant material. Sharma and Kevan (1963) reported <u>Folsomia similis</u> could be bred on a diet consisting of the leaves of sugar maple and elm and yeasts, but that populations did not reproduce on a

diet in which either the yeast or the higher plant material was lacking. In some cases, the predaceous habitat has been reportedly facultative, as in <u>Pseudosinella alba</u>, which will thrive on decaying plant material and yeasts but occasionally is cannibalistic on its own eggs (Sharma and Kevan 1963).

Christiansen (1964) suggested that bacteria may form an important source of food. Dunger (1956) showed that while Collembola were able to feed on fresh leaves, leaves which had been attacked by micro-organisms were eaten more rapidly. Shaller (1950) found a preference for decaying leaves in <u>Tonocerus flavescens</u>.

Healey (1965) noted that some species of Collembola can survive for a period of 18 months, apparently without feeding, in a state of facultative diapause.

Poole (1959) concluded, from gut analysis, that the larger species of Collembola fed mainly on soil fungi, whereas the smaller forms appeared to feed directly on humus. Angrell (1940) reported no food preferences for different species as he found the gut contents of some species to consist entirely of fungal mycelia in one area, and of amorphous detritus in another area. Strebel (1928) recorded decaying plant material, fungal mycelia, spores, dipteran pupae, other Collembola, parts of decaying earthworms and their own cast cuticles from individuals of <u>Hypogastrura purpurescens</u>.

In the living state, Collembola contribute to the soil in two different ways. First, they remove from it material which is ingested into the gut and, second, they produce fecal pellets which are added to the soil (Burgess and Raw 1967). Poole (1959) suggested that Collembola play an important part in the dissemination of fungi, and in the breakdown of the feces of larger arthropods.

#### Diplura

Kuhnelt (1961) reported <u>Japyx</u> spp. in ground litter, in deep humus beds, and beneath stones. They were predaceous feeders on various soil dwelling insects, but preferred the soft sluggish Collembola of the genus Onychiurus. He also noted that they were strongly thigmotactic and showed a preference for sites with a high and relatively stable moisture regimen; members of the genera <u>Campodea</u>, <u>Plusiocampa</u>, and <u>Japyx</u> favored deep, porous litter and humus layers and the moist cavities beneath stones and decaying wood.

Engleman (1961) recorded japygids from an old field in Michigan. Jacot (1940) noted that <u>Japyx</u> was not uncommon in mull soils of the southern states and had but a negligible effect on the soil.

#### Protura

Protura are seldom found in large numbers but are regularly detectable in certain permanently moist forest and meadow soils. A moderate soil water content is necessary for them to thrive normally; they avoid dryness as well as too much wetness (Raw 1956b).

Little is known about the food of protura. Sturm (1959) observed <u>Aceren-tomon</u> piercing the hyphae of mycorhiza-fungus which grows on roots of oak and hornbeam. Borror and DeLong (1971) noted that proturans live in moist soil or humus and feed on decomposing organic matter.

Raw (1956b) encountered Protura quite frequently in moist forest and grassland soils. Nosek (1963) recorded eight species, mainly from beach and oak forest soils, of which <u>Eosentomon transitorum</u>, <u>Acerella remyi</u>, <u>Acerentomon gallicum</u>, <u>Acerentulus confinis</u>, and <u>Acerentomon carpaticum</u> occurred in frequencies of

57.2 per cent, 25.0 per cent, 14.3 per cent, and 10.7 per cent, respectively. Kuhnelt (1961) found Protura to favor moist organic soils, provided they were not too acid and although they were usually restricted to the upper 10 cm of the profile, a distinction could be made between surface-dwelling species such as <u>Acerentomon doderoi</u>, <u>Eosentomon spinsosum</u>, and <u>Eosentomon armatum</u>, which have relatively long legs, and the inhabitants of deeper levels, such as <u>Acerentulus</u> <u>minimus</u> and <u>Protentomon</u> spp., which have shorter legs.

Jacot (1940) obtained two specimens from 90 in<sup>3</sup> of soil 27.5 to 32.5 cm deep under <u>Andropogon</u> sod, as well as an occasional individual 10.0 to 17.5 and 27.5 to 32.5 cm deep under woodland in the southern Appalachians. He found their effect similar to Collembolans but not as intensive. He noted them to feed on dead roots and animals.

#### Symphyla

Burgess and Raw (1967) reported that phytophagous species, such as <u>Scutigeralla immaculata</u> Newport, feed on young roots near the soil when soil conditions are otherwise unfavorable. Symphylids feed mainly on vegetable material and soil micro-organisms, and some, such as <u>Scutigeralla immaculata</u>, are serious pests of horticultural crops, particularly in glasshouses. They contribute to the breakdown of soil organic matter but this is limited due to the relatively small part of the total biomass of the soil fauna.

Friedel (1928) found symphylids under stones, in leaves, and humus, very sensitive to dessication. The presence of suitable cavities in the soil was very important for the occurrence of <u>Scutigella immaculata</u>, since it was not capable of burrowing. According to Friedel, these animals were little sensitive to temperature and did not hibernate as long as the soil had not frozen. He also noted them to feed exclusively on plant detritus like leaf and moss

residues. They also ate the soft parts of leaves, leaving a venal skeleton.

#### Thysanoptera

Wallwork (1970) recorded Thysanoptera present as adults, immatures, or both but rarely regarded it as an important member of the soil fauna. Jacot (1940) has suggested that although thrips have been found in litter, even to the extent of a dozen per square foot of surface, they are not known to affect soil structure.

Kuhnelt (1961) noted a distinction between the two subgroups of Thysanoptera. The Terebrantia, characterized by a non-tubular, posterior end of the body, fed on living plants and occurred only occasionally in leaf litter, mainly for hibernation. The Tubulifera, which have a tubular extension to the posterior end, were normal inhabitants of ground litter and were found in moss cushions, lichen beds, under the bark of trees and in and upon tree fungi. He noted the nymphs in particular to feed on tree fungi. In addition, mites, insect eggs, and soft insects were consumed.

## APPENDIX II

# 1972 OSAGE SOIL MACROARTHROPODS

Soil macroarthropod data collected at the Osage Site were recorded on form NREL-35. These data are stored as Grassland Biome Data set A2U30F9. A sample data form and a listing of the data are attached.

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# GRASSLAND BIOME

U.S. INTERNATIONAL BIOLOGICAL PROGRAM

FIELD DATA SHEET--MACROARTHROPOD CORES

| Data Type                                                                                                                                                             | Site                                 | Initials    | Day              | Date<br>Month                                                                                        | Year                                                                                                                        | Treatment                      | Rep]icate     | Core<br>Diameter | Quadrat | Trophic Leve | Top Depth | Bottom Depth | Condition | Class    | Order | Fami ly | Genus | Species | Life<br>Stage |   |
|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------|-------------|------------------|------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------|--------------------------------|---------------|------------------|---------|--------------|-----------|--------------|-----------|----------|-------|---------|-------|---------|---------------|---|
| 1-2                                                                                                                                                                   | 3-4                                  | 5-7         | 8-9              | 10-11                                                                                                | 12-13                                                                                                                       | 14                             | 15            | 16-18            | 19-21   | <u> </u> -   |           | 28-29        |           | <u> </u> | 36-39 | 46-49   |       |         |               | - |
| 35                                                                                                                                                                    |                                      |             | -                |                                                                                                      |                                                                                                                             |                                |               |                  |         |              | 20-27     | 20-29        |           | 51-54    | 30-39 | 46-49   | 51-54 | 56-58   | 60-61         | + |
| 1 /                                                                                                                                                                   | ditio<br>Alive<br>Dead<br>Unkno      |             |                  |                                                                                                      | ·                                                                                                                           | 1                              | <b>_</b>      |                  |         |              |           |              |           |          |       |         |       |         |               |   |
| Life Stage<br>00 Undetermined<br>10 Adult<br>11 Adult male                                                                                                            |                                      |             |                  | <u>Tr</u><br>0<br>1                                                                                  |                                                                                                                             |                                |               |                  |         |              |           |              |           |          |       |         |       |         |               |   |
| 12Adult female2Plant feed20Pupa(sap)30Egg3Plant feed31Egg case(pollen &32Egg case, with(seed)nymphs5Predator40Nymph or larva641Nymph or larva,parasitoiearly7Omnlyore |                                      |             |                  |                                                                                                      | <ol> <li>Plant feeding         (sap)</li> <li>Plant feeding         (pollen &amp; nectar)</li> <li>Plant feeding</li> </ol> |                                | ng<br>Dectar) |                  |         |              |           |              |           |          |       |         |       |         |               |   |
|                                                                                                                                                                       |                                      |             |                  |                                                                                                      | bid                                                                                                                         |                                |               |                  |         |              |           |              |           |          |       |         |       |         |               |   |
| 42 Nymph or larva,<br>middle<br>43 Nymph or larva,<br>late<br>50 Instar                                                                                               |                                      |             | 9<br>A           | <ul> <li>9 Non-feeding stage</li> <li>A Root feeding<br/>(tissue)</li> <li>B Root feeding</li> </ul> |                                                                                                                             |                                |               |                  |         |              |           |              |           |          |       |         |       |         |               |   |
| 51 Instar, 1st<br>52 Instar, 2nd<br>53 Instar, 3rd<br>54 Instar, 4th<br>55 Instar, 5th<br>56 Instar, 6th                                                              |                                      | 1 2         | 2 Lightly grazed |                                                                                                      |                                                                                                                             | ed                             |               |                  |         |              |           |              |           |          |       |         |       |         |               |   |
| 57  <br>11 A<br>12 B                                                                                                                                                  | 7 Instar, 7th                        |             |                  | 4  <br>5  <br>A [<br>B [                                                                             | 4 Heavily graz<br>5 Ungrazed cur<br>year only                                                                               | raze<br>curr<br>/<br>:<br>rate | ed<br>rent    |                  |         |              |           |              |           |          |       |         |       |         |               |   |
| 3 Bridger<br>4 Cottonwoo<br>5 Dickinson<br>6 Hays<br>7 Annual                                                                                                         |                                      | wood<br>son | rod<br>n         | C (<br>D E<br>E E<br>F E                                                                             | C Diet heav<br>D ESA - O<br>E ESA - W<br>F ESA - N                                                                          | avy                            | avy           |                  |         |              |           |              |           |          |       |         |       |         |               |   |
| 8 Jorn<br>9 Osag<br>0 Panta                                                                                                                                           | Jornada<br>Osage<br>Pantex<br>Pawnee |             |                  |                                                                                                      |                                                                                                                             |                                |               |                  |         |              |           |              |           |          |       | -       |       |         |               |   |
|                                                                                                                                                                       |                                      |             |                  |                                                                                                      |                                                                                                                             |                                |               |                  |         |              |           |              |           |          |       |         |       |         |               |   |
|                                                                                                                                                                       |                                      |             |                  |                                                                                                      |                                                                                                                             |                                |               |                  |         |              |           |              |           |          |       |         |       |         |               |   |
|                                                                                                                                                                       |                                      |             |                  |                                                                                                      |                                                                                                                             |                                |               |                  |         |              |           |              |           |          |       |         |       | I       | T             |   |

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## +++ EXAMPLE OF DATA +++

| 1 2                    | 3         | 4               | c            | ,              |
|------------------------|-----------|-----------------|--------------|----------------|
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| 3509RMS160572115080030 | 00103     | DIPT            | PSYC         |                |
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| 3509RMS160572115080027 | 20503     |                 |              | 10 03          |
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| 3509RMS160572515080037 | 00103     | HYME            | FORM         | 10 51          |
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| 3509RMS160572515080028 | 10203     | ORTH            | BLAT         | 00 01          |
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| 3509RMS160572515080020 | 20503     | DIPT            | CECI         | 10 01          |
| 3509RMS16057251508003  | 2050      |                 | CLCI         | 10 01          |
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| 3509RMS16057252508002  | 0010      | UK I II         | DLAT         | 40 01          |
| 3509RMS160572525080037 | 00103     | HYME            | FORM         | • •            |
| 3509RMS160572525080037 | 00103     | HYME            |              | 10 21          |
| 3509RMS160572525080015 | 10203     | ARAN            | FORM         | 40 102         |
| 3509RMS160572525080018 | 10203     | ORTH            |              | 00 01          |
| 3509RMS160572525080025 | 10203     | ARAN            | BLAT         | 40 01          |
| 3509RMS160572525080020 | 10203     | DIPT            | 05.01        | 00 01          |
| 3509RMS160572525080028 | 10203     | ORTH            | CECI<br>BLAT | 10 02          |
| 3509RMS160572525080037 | 10203     | HYME            |              | 40 01          |
| 3509RMS160572525080037 | 10203     | HYME            | FORM         | 10 45          |
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| 3509RMS160572525080020 | 20503     | DIPT            | CECI         | 10 01          |
|                        | ~~~~      | DIFI            | SCIA         | 10 01          |
|                        |           |                 |              |                |

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|--------------------------|---------|--------------|----------------|----|----------|
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| 3509RMS160572525080032   | 20503   | HOMO         | APHI           | 10 | 01       |
| 3509RMS160572525080037   | 20503   | HYNE         | FORM           | 10 | 02       |
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| 3509RMS050772115080027   | 00103   | HYME         | FORM           |    | Q1       |
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| 3509RMS05077211508003    | 1020    |              |                |    |          |
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| 3509RMS05077212508003    | 1020    | LULE         | CURC           | 10 | 01       |
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| 3509RMS05077251508003    | 1020    |              |                |    |          |
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| 3509RMS050772525080037   | 00103   |              |                |    |          |
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| 3509RMS05077252508002    | 1020    |              |                |    |          |
| 3509RMS05077252508003    | 1020    |              |                |    |          |
| 3509RMS05077252508001    | 2050    |              |                |    |          |
| 3509RMS05077252508002    | 2050    |              |                |    |          |
| 3509RMS050772525080031   | 20503   | COLE         | CU.D.C         |    |          |
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| 3509RMS08087211508002    | 1020    |              |                |    |          |
| 3509RMS08087211508003    | 1020    |              |                |    |          |
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| 3509RMS080872125080032   | 0010    | HONO         | DELP           | 10 | 01       |
| 3509RMS080872125080037   | 0010    | HYME         | FORM           | 10 | 01       |
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| 3509RMS080872125080020   | 1020    | CHEL         |                | 00 | 01       |
| 3509RM508087212508003    | 1020    | UNEL         |                | 00 | 01       |
| 3509RMS08087212508001    | 2050    |              |                |    |          |
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| 3509RMS08087212508003<br>3509RMS080872515 <b>0</b> 80010 | 2050           |      |              |          |          |
|----------------------------------------------------------|----------------|------|--------------|----------|----------|
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| 3509RMS080872515080037                                   | 0010           | HYME | FORM         | 00       | 01       |
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| 3509RMS080872515080027                                   | 1020           | HYME | FORM         | 00<br>10 | 02<br>29 |
| 3509RMS080872515080027                                   |                | HYME | FORM         | 40       | 22       |
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| 3509RM\$08087252508003                                   | 0010           |      |              |          |          |
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| 3509RMS21117211508003                                    | 0010           |      |              |          |          |
| 3509RMS21117211508001                                    | 1020           |      |              |          |          |
| 3509RMS21117211508002                                    | 1020           |      |              |          |          |
| 3509RMS21117211508003<br>3509RMS21117211508001           | 1020           |      |              |          |          |
| 3509RMS21117211508001                                    | 2050           |      |              |          |          |
| 3509RMS21117211508002                                    | 2050<br>2050   |      |              |          |          |
| 3509RMS21117212508001                                    | 0010           |      |              |          |          |
| 3509RMS21117212508002                                    | 0010           | HEMI | DENT         | • •      |          |
| 3509RMS21117212508003                                    | 0010           | COLE | PENT<br>CARA | 10       | 01       |
| 3509RMS21117212508003                                    | 0010           | ORTH | BLAT         | 40       | 01       |
| 3509RMS21117212508001                                    | 1020           |      | ULA I        | 40       | 01       |
| 3509RMS21117212508002                                    | 1020           |      |              |          |          |
| 3509RMS21117212508003<br>3509RMS21117212508001           | 1020           |      |              |          |          |
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| 3509RMS21117212508003                                    | 2050           |      |              |          |          |
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| 3509RMS21117251508002                                    | 0010           | HEMI |              | ( •      |          |
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| 3509RMS21117251508002                                    | 2050           |      |              |          |          |
| 3509RMS21117251508003                                    | 2050           |      |              |          |          |
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| 3509RMS21117252508002                                    | 0010           | DIPT |              | 40       | 01       |
| 3509RMS21117252508002<br>3509RMS21117252508002           | 0010           | LEPI |              | 40       | 01       |
| 3509RMS21117252508002                                    | 0010           | HYME | FORM         |          | 23       |
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| 3509RMS21117252508003                                    | 1020           |      |              |          |          |
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| 3509RMS21117252508002                                    | 2050           |      |              |          |          |
| 3509RMS21117252508003                                    | 2050           |      |              |          |          |
|                                                          | ··             | ·    |              |          |          |
|                                                          |                |      |              |          |          |