Soil Biology and Ecology

Introduction

Lecture 1: Soil Biology and Ecology

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   Step-by-Step Instructions for Students

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Introduction: Soil Biology & Ecology

UNIT OVERVIEW
This unit introduces students to the biological properties and ecosystem processes of agricultural soils.

The lecture reviews the constituents of soils and the physical characteristics and soil ecosystem processes that can be managed to improve soil quality. Demonstrations and exercises introduce students to techniques used to assess the biological properties of soils. Such assessments help inform decisions about soil management with the goal of maintaining crop productivity and soil health in organic farming and gardening systems.

MODES OF INSTRUCTION
> LECTURE (1 LECTURE, 1.5 HOURS)
The lecture covers the basic biology and ecosystem processes of soils, focusing on ways to improve soil quality for organic farming and gardening systems.

> DEMONSTRATION 1: ORGANIC MATTER DECOMPOSITION (1.5 HOURS)
In Demonstration 1, students will learn how to assess the capacity of different soils to decompose organic matter. Discussion questions ask students to reflect on what environmental and management factors might have influenced the test results and what the results suggest about nutrient cycling rates and the quality/health of the soils tested.

> DEMONSTRATION 2: SOIL RESPIRATION (1 HOUR)
Demonstration 2 covers the use of Draeger gas detection tubes for measuring carbon dioxide levels liberated from soils as an indicator of soil biological activity and soil quality/health.

> DEMONSTRATION 3: EARTHWORM POPULATION (1 HOUR)
Demonstration 3 takes students through the process of sampling soil for earthworm types. Discussion questions ask students to consider the presence and abundance of certain earthworm types as indicators of soil quality/health.

> DEMONSTRATION 4: SOIL ARTHROPODS (1 HOUR)
Demonstration 4 covers the preparation and materials used to collect and identify soil arthropods. Discussion questions ask students to consider the presence and diversity of soil arthropods as indicators of soil quality/health.

> ASSESSMENT QUESTIONS (1 HOUR)
Assessment questions reinforce key unit concepts and skills.

LEARNING OBJECTIVES
CONCEPTS
- Soil quality/soil health
- Mineralization/immobilization
- Autotrophic/heterotrophic food webs
- Functional groups of soil biota
- Rhizosphere ecology
- Management effects on soil ecosystems
SKILLS

- How to assess soils for biological activity through measuring the rate of decomposition of cellulose
- How to assess soil biological activity through measuring soil respiration
- How to assess soil biological activity through earthworm census
- How to assess the soil ecosystem structure through a soil arthropod census
Lecture 1: Soil Biology & Ecology

Pre-Assessment Questions

1. What is soil?
2. What forms of life exist in soil ecosystems?
3. How would you define a “healthy” agricultural soil?
4. What is a food web?
5. Can you describe a decomposer food web that may exist in the soil?
6. What might be some negative effects of the long-term practice of monoculture cropping and the use of synthetic chemical fertilizers and pest control agents on the soil ecosystem?

A. What Is Soil? (should be a review in part; see also Unit 2.1, Soils and Soil Physical Properties)

1. Soil components
   a) Mineral
      i. Derived from parent material
   b) Soil organic matter
   c) Water and air
      i. 1/2 soil volume = pore space
      ii. Importance of gas diffusion: When diffusion is slow, as with water-saturated soil, respiration byproducts (such as CO₂) accumulate and inhibit aerobic processes (such as respiration itself)
      iii. CO₂ is about 1% in dry soil, up to 10% in saturated soil
   d) Biota: The smallest life forms are inseparable from soil organic matter

2. Soil structure vs. soil texture
   a) Soil texture, a native characteristic
      i. Soil texture: The relative percentage of sand, silt, and clay particles
      ii. The bricks, boards, and mortar (the physical materials) that make up soil
      iii. The particle sizes have surface area:volume effects. This influences properties such as cation exchange capacity (CEC), pore space, water holding capacity, and aggregate formation.
   b) Soil structure, a manageable characteristic
      i. Soil structure: The arrangement of soil particles. The “architecture” of soil—what shapes you build with the “bricks, boards and mortar.”
      ii. Determines movement of gases and water in soil
      iii. Creates small habitat spaces
      iv. Water stability: Aggregates that retain shape when wetted maintain a more stable soil structure
      v. Influences soil tilth/soil health

B. What Is a Healthy Soil? (see also Unit 1.1, Managing Soil Health)

1. Question: Is soil merely a solid medium that holds nutrients for plant growth or does soil serve other functions?
2. Soil health and soil quality are generally synonymous
3. Definition of soil health: “Capacity of a soil to function, within land use and ecosystem boundaries, to sustain biological productivity, maintain environmental quality, and promote plant, animal, and human health.”
a) Soil is recognized as an essential component of the biosphere
b) Soil is required for significant production of food and fiber
c) Soil contributes to maintaining and enhancing air and water quality
d) Soil filters and chemically alters water
e) The definition of soil health must be broad enough to encompass the many functions of soil

4. Assessment of soil health
   a) Analogous to monitoring human health
   b) Indicators needed to identify problems and to monitor the effects of management
   c) Requires a holistic approach
   d) Should include physical, chemical, and biological attributes of soil
   e) Indicators must be measurable by as many people as possible, at many different skill levels
   f) Definition and assessment of soil quality is complicated by the fact that soil is not (typically) directly consumed by animals and humans, unlike air and water
   g) Basic data set of soil health indicators
      i. Soil texture
      ii. Rooting depth
      iii. Water infiltration
      iv. Bulk density
      v. Water holding capacity
      vi. Soil organic matter
      vii. pH
      viii. Cation exchange capacity (CEC)
      ix. Extractable N, P, and K
      x. Microbial biomass C and N
      xi. Potentially mineralizable N
      xii. Soil respiration
      xiii. Soil temperature

5. Protection of soil health as a national priority
   a) National Research Council recommendation (1993): “Protecting soil quality, like protecting air and water quality, should be a fundamental goal of national environmental policy”

C. Nutrient Cycling and Decomposition
   1. Mineralization/immobilization
      a) Soil nutrients occur as parts of:
         i. Inorganic compounds: Some of these are available to plants
         ii. Organic compounds: Are part of living organisms and decaying organic matter. These nutrients are stored (“immobilized”) in the biomass of the organisms and are unavailable until released during decay or consumption.
      b) Soil organisms are constantly transforming nutrients between these 2 forms
c) Mineralization: Soil organisms excrete inorganic waste compounds that may adhere to CEC sites and/or dissolve in soil water (soil solution) for possible uptake by crop plants. Net mineralization must be greater than net immobilization for nutrients to be available to crop plants.

d) Immobilization: Soil organisms consume inorganic compounds to construct living tissues. These nutrients are temporarily stored and unavailable for plant uptake.

2. Soil organic matter (SOM): Includes all organic substances in or on the soil
   a) Living organisms—include plant roots and all soil biota (< 5% of SOM)
      i. Cellulose, the major carbohydrate structural building block for plants, is the most abundant compound on earth and the major component of soil organic matter
      ii. Lignin is the second largest input into SOM
   b) Fresh and decomposing organic residues (40–60% of SOM)
      i. Easily decomposable (active, labile) fraction: The quantity of this fraction of SOM changes quickly in response to management practices and is the organic matter fraction from which the majority of plant nutrients are liberated into the soil solution for uptake by plants
      ii. Moderately decomposable fraction: This fraction is physically and/or chemically more complex than labile OM. Its decomposition is slower and therefore fewer nutrients are mineralized from it in a given season.
   c) Resistant (recalcitrant) fraction: Also called humus, and is resistant to further decomposition (33–50% of SOM). Has greater influence on the structure/physical properties of soils than on nutrient availability.
   d) See Appendix 1, Major Organic Components of Typical Decomposer Food Sources

3. Nitrogen cycle (see Figure 2.10 in Unit 2.2, Soil Chemistry and Fertility)
   a) Proteins break down —> amino acids —> ammonium (form of N usable by some plants) —> nitrate (form of N usable by most plants)
   b) Ammonification (aerobic or anaerobic): The biochemical process in the N cycle above whereby ammonium is released from nitrogen-containing organic compounds (amino acids)
c) Nitrification (aerobic): The biochemical process in the N cycle above whereby bacteria convert ammonium to nitrate
   i. Inhibited by low oxygen or low temperatures
   ii. This leads to ammonium build-up in cold, wet soils

D. Soil Food Webs

1. Soil food web ecology
   a) Food webs trace the path of energy or nutrients passing from one organism to the next

2. Heterotrophs vs. autotrophs in food webs
   a) Autotrophs form the base of food webs, and acquire their own C from the atmosphere. In the soil food web, this begins with C fixation by plants, which is photosynthesis. Energy for most life is derived from sunlight that has been transformed by photosynthetic plants into organic compounds.
   b) Heterotrophs in food webs consume organic matter to acquire carbohydrates for respiration. By consuming organic matter, they release nutrients, making them available to other plants and animals, or become food themselves for other organisms.

3. Energy loss = 80–90% at each step in the food chain

4. Food web structure and properties
   i. Resilience = speed of recovery after disturbance. Resilience decreases with increasing number of trophic levels due to increasing complexity—it takes longer to reestablish complex food web relationships
   ii. Disturbance selects for shorter food chains: In farmed soils, disturbance can be chemical (pesticides, fertilizers) or physical (cultivation, organic matter incorporation, removal of surface organic layer)
      *The frequency of soil disturbance by physical or chemical agricultural inputs and other disturbances is important to the overall assemblage of soil biota and food chain length
   iii. Fungi:bacteria biomass ratio characteristics of soil ecosystems
      • Productive agricultural soils have a ratio of 1:1 or less (higher in no-till). These are bacterial-dominated food webs with rapid cycling of nutrients.
      • Deciduous forest soils, 5:1 to 10:1 (fungal dominated)
      • Coniferous forest soils, 100:1 to 1000:1 (fungal dominated)

5. Some heterotrophic roles in soil food webs
   i. Shredders: Shred organic matter, increasing the surface area and making the food available to more microorganisms. These include earthworms and arthropods.
   ii. Grazers: Feed on bacteria and fungi, stimulating and controlling the growth of those populations. Grazers include protozoa, nematodes, and microarthropods.
   iii. Higher-level predators: Consume other heterotrophs, like grazers and shredders, helping control the lower trophic-level predator populations

6. Unique food web for each ecosystem, determined by:
   i. Climate
   ii. Soil/parent material
   iii. Vegetation
   iv. Land management practices
E. Soil Biota

1. Community characteristics
   a) High diversity of organisms in soil can rival that of coral reef ecosystems
   b) High abundance of organisms, on the order of hundreds of millions to billions of microbes in 1 g of soil
   c) High biomass of organisms, e.g., from hundreds to thousands of pounds of microbes per acre of soil

2. Habitats
   a) Habitats within soil ecosystems are unevenly distributed
   b) Habitats are concentrated at organic matter sites
      i. Root zone (rhizosphere)
         • Succession of organisms as root grows
         • Some root exudates (molecules released into the soil by the roots, including sugars and amino acids) may stimulate microorganisms and thus increase labile SOM
      ii. Litter (dead organic matter on the soil surface)
      iii. Surfaces of soil aggregates
      iv. Incorporated organic matter

3. Functional classification
   a) Microorganisms
      i. Colonial growth forms (cells about 1/25,000 inch wide)
         • Bacteria, archaea, and yeast
         • Adapted to high surface area:volume environments
         • Colonize surfaces, crevices, pores
         • Teaspoon of soil contains 100 million to 1 billion bacteria
         • Biomass equivalent to 2 cows per acre
         • Functional roles include: N fixers, nitrifiers, denitrifiers, decomposers (the byproducts of which help in the formation of soil aggregates), pathogens
      ii. Mycelial growth forms (hyphae length ranges from a few cells to many yards)
         • Fungi actinomycetes, and oomycetes
         • Penetrate organic matter
         • Translocate nutrients
         • Functional roles include: Decomposers, mutualists, pathogens, predators (e.g., nematode-trapping fungi)
      iii. Algae
         • Dominated by “blue-green” algae (Cyanophyta) and eukaryotic algae
         • Present where sunlight is available near soil surface, active when there is moisture available, too
         • Help bind soil particles together, reducing erosion potential (biological soil crusts)
         • Increase water retention capacity of soil through exudates
         • Often 1000 to 10,000 per g of soil
         • Functional roles: Primary producers (photosynthesizers), N fixers
   b) Microfauna
      Note: This section and the macrofauna section below are based on information from the European Atlas of Soil Biodiversity; see Resources for details
      i. Protozoans (1/5000 inch to 1/50 inch wide)
• Small animals (acellular) living in water films
• Encystment (hibernating in a cyst): Distinctive response to drying out
• Inhabit transitory environments, so reproduce rapidly
  – Colpoda divide once or twice per day at 120°C
• Several distinct types
  – Ciliates have fringe of small hairs used for locomotion
  – Amoebae have an amorphous body shape
  – Flagellates have a whip-like tail for locomotion
• Functional roles: Predators (e.g., of bacteria, other microorganisms), decomposers
  (feed on detritus)

ii. Nematodes (1/500 inch in diameter, 1/20 inch in length)
• Global distribution
• Soil abundance = million/m²
• Outer cuticle protects; resistant to toxins
• Functional roles include: Microbivores, omnivores, predators, some parasites (10%)
• Abundant at sites with high OM concentration

iii. Rotifers (1/50 to 1/120 inch long)
• Multicellular, though still microscopic
• Live in water films on moist soils; one of most abundant taxa in the top layer of soil
  or litter (32,000 to 2 million per m²)
• Can undergo anhydrobiosis (survive drying down by forming a cyst)
• Primary feeding strategies: Grazing the bacterial film on SOM or other particles or
  filter feeding on bacteria, yeast, and algae in the soil water
• Functional roles: Herbivores (e.g., on algae), decomposers (feed on detritus)

iv. Tardigrades (1/25 inch)
• Multicellular, though still microscopic
• Terrestrial tardigrades live on moss or lichen
• Can undergo anhydrobiosis (survive drying down by forming a tun)
• Primary feeding strategies: Some use stylets to pierce moss, algae, protozoans,
  rotifers or nematodes and suck out fluids; others consume whole microfauna
• Functional roles: Herbivores, predators

v. Functional roles of microfauna do not include shredding of organic matter into
  smaller pieces

C) Mesofauna
i. Potworms (Enchytraeida, 1/50 to 2 inches long)
• Small annelids (related to earthworms
• Tolerate pH < 4
• Thousands/m² in high organic matter soil
• No burrows
• Feed on fungal hyphae, microorganisms, feces

ii. Collembolans (springtails, 1/100 to 4 inches long)
• Small arthropods (related to insects); with mites, the most numerous soil
  arthropods
• Live in soil and leaf litter
• Hundreds to thousands per handful of soil high in SOM
• Feed on fungal hyphae, bacteria, detritus
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iii. Mites (acari, 1/125 to 1/30 inches long)
   - Small arachnids (related to spiders); with collembolans, the most numerous soil arthropods (1000 to 10,000 per m²)
   - Global distribution
   - Live in soil and in habitats with high quantities of OM
   - Primarily predators, feeding on collembola, nematodes, insect larvae

iv. Insect larvae
   - Fly (Diptera) larvae are probably the most important
   - In home compost systems, black soldier fly larvae (in the family Stratiomyidae, order Diptera) can play a key role in consuming organic matter, on par with earthworms
   - Diverse functional roles include: Predators, parasites, herbivores, decomposers (feeding on detritus)

v. Symphyla (1/125 to 1/30 inches)
   - Small soil-dwelling myriapods, related to millipedes and centipedes
   - Primarily eat decaying vegetation and microorganisms, but also seeds, roots, and root hairs in agroecosystems, thus damaging crops when they do
   - Up to 20,000 per m²

vi. Overall, mesofauna regulate microfauna (and other mesofauna) by grazing

vii. Minor shredding of organic matter

viii. Total of 500 to 200,000 per square meter, far less abundant and with lower biomass than microfauna

d) Macrofauna

i. Earthworms (1/3 to 45 inches long)
   - 3 ecological types: Anecic—large, live in permanent burrows in the soil, feed on litter from the surface mixed with ingested soil; endogeic—small, live in temporary burrows in the soil, feed on rich soils to obtain nutrients from organic matter; epigeic—small, live at the soil surface in litter, feed on litter there
   - Obtain nutrition from partially decomposed organic matter and part from microbes living on the organic residues they ingest
   - Stimulate microbial activity through effects on SOM, microbial inoculation onto substrates, soil structure, etc.
   - Mix and aggregate soil
   - Increase water infiltration
   - Provide channels for root penetration deep into soil
   - Bury and shred organic matter
   - Abundance decreases after disturbance (tillage, chemicals)

ii. Myriapods
   - Millipedes (Diplopoda, 1/12 to 11 inches long) and centipedes (Chilopoda, 1/8 to 11 inches long)
   - Millipedes live in litter and upper layers of soil; some are shredders that feed on organic matter, others are predators on arthropods or earthworms, others pierce and suck plant cells. More common in soils high in calcium carbonate (e.g., from limestone); 15 to 800 per m².
   - Large species of centipedes live in litter or close to the soil surface, while small and narrow species of centipedes live in deeper soil layers. They are primarily generalist predators consuming insect adults and larvae, collembolans, mites, nematodes, potworms and earthworms, and occasionally leaf litter; 20-300 per m².
iii. Isopods (woodlice, 1/15 to 2 inches long)
   - Crustaceans, related to lobsters and crabs
   - Live in leaf litter, in vegetation, under stones
   - Generally are decomposers, feeding on dead organic material, but sometimes are predators of bacteria, fungivores, or herbivores

iv. Mollusks (snails and slugs, ¼ inch to 10 inches)
   - Live in damp soil conditions (although snails can hibernate for up to 4 years in dry conditions)
   - Most active at night or on cloudy, foggy days
   - Small component of soil fauna biomass but can be of high agronomic and ecological significance (especially when populations near ½ million per acre)
   - Primarily herbivores, especially of succulent foliage such as seedlings and fruit near the ground, but also detritivores

v. Insects
   - Many insects live in or on the soil as larvae or adults, and thus fill many functional roles in the soil food web. Two examples include:
     - Ants: Ant diversity can be very high, with tens to hundreds of species in a few acres. Ants fulfill multiple trophic roles, e.g., herbivores, predators, scavengers, parasites.
     - Carabid beetles: Both larvae and adults may live in the soil. May be predators, e.g., feeding on snails or collembola, fungivores, frugivores (eating seeds), or herbivores.

vi. Macrofauna shred and incorporate plant remains (may become pests by feeding on living plants if insufficient organic residues present)

vii. Also alter the soil structure, e.g., by burrowing, mixing, defecating, and helping form soil aggregates

e) Megafauna
   - Large invertebrates, vertebrates, including moles, mice, rabbits, gophers, snakes, and lizards
   - Primary ecosystem engineers of the soil: Important for moving and turning soil, contributing to nutrient cycling, aeration, and drainage
   - Fill a range of functional roles: Herbivores and predators of invertebrates and small vertebrates

F. Rhizosphere Ecology

1. Definitions
   a) Rhizosphere (R): The narrow zone of soil subject to the influence of living roots, as manifested by the leakage or exudation of substances that promote or inhibit microbial activity
   b) Rhizoplane (r): The actual root surface, which provides a highly favorable nutrient base for many species of bacteria, archaea and fungi
   c) Edaphosphere (S): Soil beyond root influence
   d) Rhizosphere Effect: Soil microorganisms are stimulated by the roots
      i. RsS ratio generally greater than 1 (i.e., more biota in R than in S)
   e) Rhizosphere succession: The sequence of changes in the composition and densities of soil microbes and fauna in the area surrounding a growing root (see below)
2. Roots
   a) Root environment
      i. Determined by above-ground processes (products of photosynthesis are
         translocated to roots)
      ii. Exudates (see below), sloughed hairs, and epidermal (root’s surface) cells
          feed soil organisms in R and r
      iii. Plant roots also can release bicarbonate (HCO$_{3}^{-}$), which raises the soil pH. This can
          make some cations (e.g., Fe$^{3+}$, Ca$^{2+}$, Mg$^{2+}$, and K$^{+}$) unavailable to plants. Irrigation
          water may also contain bicarbonate and affect soil pH and availability of some
          nutrients.
      iv. Oxygen decreases, CO$_{2}$ increases in root zone over time due to plant and R organism
          respiration
   b) Root form
      i. Fibrous roots
         • Most monocots (e.g., grasses, corn)
         • Primary root replaced by series of adventitious roots
      ii. Tap roots
         • Most monocots and gymnosperms
         • Tap root persists and forms many lateral branches
      iii. Root depth
         • Species specific, influenced by environmental conditions
   c) Root structure
      i. Root cap
         • Live cells produced by meristem
         • Protects root, like a bud scale
         • Constantly replaced (5–6 day turn over)
         • Responds to gravity
      ii. Meristematic zone: 2 mm (.08 inch) zone where most cell division happens
      iii. Zone of elongation: Rapid growth, cells from meristem
      iv. Mucilage
         • Covers root from tip to beginning of root hair zone
         • Secreted by root cap and epidermal cells
         • Possible functions: Lubricates and protects root as it grows through the soil, helps
           with nutrient uptake, prevents drying, fills spaces between root and soil and helps
           bind soil aggregates, food for microbes, including beneficial microbes
      v. Root hair (differentiation) zone
         • Are lateral outgrowths of single epidermal cells; microscopic
         • Root hairs have life span of days to weeks; rye plants can produce over 100 million
           per day
         • Do not become large structural roots, though help anchor the plant in the soil
         • Key role is improving nutrient absorption by increasing surface area for nutrient
           and water uptake. Root hairs make up the majority of root surface area.
         • Food sources that support rhizosphere microbes, contribute significant amounts
           of soil organic matter
      vi. Lateral roots
         • Originate from the vascular bundle inside the root cortex
         • Cortex and epidermis are ruptured by new lateral root
         • Bacteria colonize these emergence sites
vii. Vascular bundle
   • Xylem and phloem in the root cortex
   • Connects roots to the rest of the plant, including for transport of photosynthetic products (sugars) to the roots and of water and nutrients from the soil up to the aboveground portion of the plant
   • Foliar sprays may move into roots (depends on molecular weight)
   • Herbicides, antibiotics may also move into roots
   • Streptomycin moved from Coleus leaves to roots in 24 hrs; bacteria in the rhizosphere were suppressed by the streptomycin

d) Root nutrition
   i. Maximum nutrient uptake occurs behind meristem (in the elongation and root hair zones)
   ii. Water and nutrients are withdrawn from narrow band around roots
   iii. Replenished from surrounding soil by mass flow (the movement of nutrients with the overall flow of water to plant roots); all ions in solution move towards root during mass flow
   iv. If mass flow is slower than uptake, a depletion zone is created around the root, resulting in lack of some nutrients
   v. If uptake is slower than mass flow for a particular ion (or even nonexistent if the ion is not used by the plant) certain ions may accumulate around the root

e) Root exudates
   i. Amounts
      • 20–50% more C enters the soil from exudates, sloughed cells, and root hairs than is present as fibrous roots at end of growing season = substantial contribution to SOM
      • Amount of exudates increased by:
         – Wetting, after a drying spell
         – Physical or chemical injury (e.g., mowing, grazing of perennial grass cover crop)
         – Abrasion, phytotoxic residues, osmotic stress
      • Amount of exudate varies with plant species and age, as well as the soil environment
   ii. Types
      • Carbohydrates and amino acids: Most-researched components of exudates
         – 10 sugars, glucose and fructose most common
         – 25 amino acids
      • Also organic acids, fatty acids, sterols, enzymes, volatile compounds, and growth factors
      • Type of exudate varies with plant species, age, soil environment
      • Difficult to separate plant and microbe sources
   iii. Exudates released from meristem zone
      • Nematodes and zoospores congregate there

f) Management effects on rhizosphere
   i. Synthetic fertilizers
      • Sometimes no effect
      • Sometimes increase R:S indirectly through stimulation of plant growth
   ii. Organic manures
      • Same indirect positive effect on R:S
3. Soil organisms
   a) Bacteria and archaea
      i. Most responsive to plant exudates
      ii. 2 to 20 fold increase in bacterial populations in R vs. S
      iii. Pseudomonas most consistently abundant in rhizosphere
      iv. Also Rhizobium (some are used in DNA transfer as part of genetic engineering) and Achromobacter
      v. Azotobacter, non-symbiotic nitrogen fixer
         • If inoculated on seed can persist in rhizosphere
      vi. Rhizobium, Nitrosomonas, and Nitrobacter, all important to the nitrogen cycle (see Figure 2.10 in Unit 2.2), common in R
   b) Fungi
      i. Average increase of 10 to 20 fold in R of crop plants from S
      ii. Fusarium is a dominant genera of R fungi
      iii. Mycorrhizae can provide physical and chemical suppression of pathogens
   c) Protozoans
      i. Mainly bacteria grazers, so some increase is expected in R
      ii. Example: In a wheat field, bacteria R:S was 23:1, protozoan R:S was 2:1
      iii. Some large amoebae may provide biocontrol of some fungi
   d) Nematodes
      i. Root substances stimulate egg hatching of some parasitic nematodes
      ii. Host and non-host plants may stimulate hatching of nematodes, e.g., some crucifers and chenopods stimulate Heterodera hatching, but don't support root invasion by larvae. Some plants will cause eggs of parasitic nematodes to hatch, but then are not susceptible to attack by the parasite. Therefore the plant stays healthy, and the nematodes fail to thrive.
      iii. Nematodes tend to congregate around elongation zone of roots
      iv. Degree of nematode attraction is proportional to root growth rate
      v. Some root exudates repel nematodes (e.g., isothiocyanates in mustard)
   e) Microarthropods
      i. Some grazers consistently more abundant around roots
   f) Rhizosphere succession
      i. Root tip releases labile carbon
      ii. Labile carbon stimulates rapid increase of microbes and thus nutrient immobilization in R
      iii. Grazers increase, tracking the microbe population increases
      iv. Water and carbon in root hair zone decrease
      v. Microbes eventually decrease; grazers cause net mineralization and release of nutrients from SOM
      vi. Later, grazers encyst or migrate
G. **Management Effects on Soil Ecosystems**

1. No-tillage or reduced-tillage cropping systems
   a) Organic litter is retained on the soil surface
   b) Physical disturbance is minimized
   c) Surface soil stays cooler and moister
   d) More surface organic matter available as food substrate
   e) Ratio of fungi to bacteria increases over time
   f) Earthworms and arthropods become more plentiful
   g) Effects on nutrient cycling: May increase total soil N, improve N use efficiency of plants, but may increase N₂O emissions
   h) Effects on soil physical properties: May increase SOM and aggregation

2. Rotations
   a) Monocultures and clean cultivation
      i. Create little habitat for soil organisms, leading to less abundant and diverse soil ecosystems
      ii. Consistent plant hosts may serve to develop populations of pathogenic organisms, causing pest problems and crop losses that facilitate the need for pesticide use.
   b) Complex rotations
      i. Result in greater variety of microbial food sources (roots, root exudates, and residues)
      ii. Increase diversity of soil organisms, leading to increased competition for resources, as well as predation of pathogens and pests
      iii. Interrupt plant-host pest cycles
   c) Multiculture or polyculture
      i. Growing more than one crop in one field
      ii. More closely mimics natural ecosystem
      iii. Likely to support even greater diversity of soil organisms, especially invertebrates
      iv. Also interrupts plant-host pest cycles

3. Biocides (insecticides, herbicides, fungicides)
   a) Effects vary depending on:
      i. Type of chemical
      ii. Species of soil organism in question
      iii. Concentration and other exposure factors
   b) High levels of pesticide use generally reduce food web complexity
      i. Methyl bromide and other fumigants are extreme examples, resulting in temporary soil sterilization
      ii. Eliminate most organisms
      iii. Some bacteria quickly return
      iv. Other organisms only slowly return
   c) Biocides and predator-release phenomenon
      i. In cases where biocides selectively eliminate predators, lower trophic levels may become more abundant
      ii. Destabilizing effect on food webs
         • Overgrazing on food sources results in depletion of food sources
         • Population explosion, followed by crash
necessarily compatible with crop needs. May result in leaching of water-soluble nutrients, especially forms of N.

d) Earthworms
   i. Most strongly affected (negatively) by fungicides and fumigants
   ii. Herbicides
      • Don’t seem to be directly toxic to earthworms
      • Indirect negative effect through elimination of vegetation

4. Food web structures
   a) Fungi/bacteria ratio
   b) Dominant microbe influences other trophic levels

5. Interaction with fertility needs (also see Unit 1.1)
   a) Measures of available nitrogen
      i. Conventional cropping systems
         • Most N provided by additions of fertilizer
         • Measurements of nitrate reflect accurately (but highly temporally) what is available to plants
         • Key management decisions are when to apply fertilizer
      ii. Cropping systems based on organic matter management
         • Soil food web becomes primary source of N derived from organic matter inputs
         • Soil analysis in efficiently managed farming systems may indicate “inadequate” levels of N at any given time because much of soil N is immobilized
         • Cumulative release of mineral N over growing season may match amounts seen in conventional system
         • Managing the timing of mineralization (through tillage, OM quality [e.g., C:N ratio], incorporation of high-OM nutrient amendments, irrigation) by soil food web becomes more critical
         • If managed well, less risk of nutrient loss through leaching or volatilization
Demonstration 1: Organic Matter Decomposition in Litter Bags

for the instructor

OVERVIEW
To demonstrate the capacity of different soils to decompose organic matter, this exercise requires you to bury cellulose disks (Whatman filter paper) in a variety of locations. This should be done at least two weeks prior to the class to allow decomposition to proceed before the disks are retrieved on the day of the class. To accelerate decomposition, filter paper disks can be dipped in a bucket of water with some fish emulsion added just before burial.

MATERIALS NEEDED
- Whatman filter paper discs
- Plastic mesh bags
- Flags to mark burial sites
- Flat shovel
- Litter Bag Data Sheet (see Appendix 2)
- Pencils

† For plastic mesh bags, you may use pond and pool netting obtained from a local feed and seed supply. It is a 3/8-inch polypropylene mesh. Cut the mesh into 6-inch x 12-inch pieces, fold in half, then fold the edges over and staple the edges shut. Other sources are the mesh bags that bulbs are sold in, garlic or onion bags, or the mesh bags that imported rice noodles are packed in. The smaller the mesh size, the smaller the organisms that will be excluded from the bag. This feature can be exploited by comparing decomposition rates of organic matter buried in bags of different mesh sizes. Organic matter in bags with very fine mesh will be decomposed primarily by microflora and microfauna. Organic matter in larger mesh bags will also be decomposed by larger fauna.

PREPARATION
1. Place litter bags in soil at least two weeks prior to class. Place them vertically in soil and all at the same depth. For a 10 cm disc, 0 to 10 cm is a convenient depth.

2. Flag each site, and make a note of the burial locations. A minimum of 3 bags should be placed in each habitat. Possible habitats include raised garden beds, cultivated fields, fallow fields, orchards, compost piles, vermicompost bins, weedy borders, and on the soil surface (not buried).
PROCEDURE

1. After two weeks, bring students out to the sites and ask them to observe the biotic, abiotic, and human management elements of the soil habitat that each bag was in, noting features such as relative soil moisture, presence or evidence (e.g., burrows or tunnels) of any soil organisms, vegetative cover and shading, and prior cultivation.

2. Students or the instructor can unbury the bags. This should be done very gently, as the paper is likely to be very fragile. (If too rapid decomposition makes this demonstration difficult, an alternative material to use is a 50/50 cotton/polyester fabric. Even if the cotton is entirely degraded, the polyester matrix will remain intact. Strips would have to be weighed before and after burial to determine mass loss.)

3. Gently brush soil from discs. Ask students to visually estimate the percentage of the disc remaining. You may wish to provide a sheet showing examples of visual estimates of percentages, e.g., to help standardize results.

4. Record results and calculate the average percentage of the disc remaining for each habitat selected. A sample form is provided (see Appendix 2, Litter Bag Data Sheet) for recording data. Appendix 3 provides an example of a filled-out data sheet.

PREPARATION TIME

1 hour to make 24 bags, 1 hour to bury 24 bags (allow additional time for gathering materials)

DEMONSTRATION TIME

1.5 hours

DISCUSSION QUESTIONS

1. After retrieving the litter bags, ask students to offer hypotheses about why the disks decompose more rapidly in some habitats than others.

2. What environmental factors might have influenced the results?

3. What management factors might have influenced the results?

4. Can you see any signs of biological activity on the disks (e.g., fungal mycelia, soil animals, invertebrate feces, comminution)?

5. What do the results suggest about nutrient cycling rates in the soils tested?

6. Can these observations for cellulose decomposition rates be extrapolated to other types of organic matter?

7. What are the limitations of this method?

VARIATIONS

If possible, pair the litter bag demonstration with other methods of assessing biological activity, such as:

- Carbon dioxide evolution (see Demonstration 2, Soil Respiration)

- Earthworm density (see Demonstration 3, Earthworm Populations)

- Tullgren funnel extractions of microarthropods (see Demonstration 4, Soil Arthropods)

- Microbial biomass measurements (this generally requires more extensive lab work, but you might check with local agricultural or ecological researchers to see if anyone doing similar work could accommodate a few samples and help your students analyze the results)
Demonstration 1: Organic Matter Decomposition in Litter Bags

**INTRODUCTION**

The decomposition of organic matter is an important soil process for organically managed farms and gardens. Organic matter includes a vast array of compounds that are biologically decomposed at various rates, depending on the compounds’ physical and chemical complexity. Physical factors such as temperature and moisture as well as biological factors such as activity of soil organisms heavily influence decomposition rates, and are all influenced by management practices.

We can use discs of filter paper to represent uniform pieces of cellulose-rich organic matter. If discs are placed in the soil for a set period and then retrieved, we can begin to understand the capacity of various soils to decompose cellulose. By placing the discs in plastic mesh bags prior to putting them in the soil, we make it easier to retrieve the discs intact. Decomposition can be estimated by a visual estimate of percentage surface area remaining. A more quantitative method is to weigh the discs prior to putting them in the field, then collecting them, rinsing them, drying them (e.g., in a drying oven) and reweighing them to estimate mass loss.

**MATERIALS NEEDED**

- Whatman filter paper discs
- Plastic mesh bags
- Stake wire marking flags
- Flat shovel
- Litter Bag Data Sheet (see Appendix 2)
- Pencils

**PREPARATION**

1. Litter bags, each consisting of a filter paper disc placed inside a plastic mesh bag, were placed in soil at least two weeks prior to this class. They were placed vertically in the soil, all at the same depth. For a 10 cm disc, 0 to 10 cm is a convenient depth, but your instructor will tell you the actual depth used.

   Depth of litter bags: ________________.

**PROCEDURE**

1. With your instructor, visit each site where the litter bags have been buried. At each site, observe the biotic, abiotic, and human management elements of the soil habitat that each bag was in.

   - What is the relative soil moisture at each site? Is it damp, dry, or in between?
   - Do you see any presence or evidence of any soil organisms? Are there burrows or tunnels, and if so, are they large or small? What kind of organisms might be using them?
   - Is there vegetative cover of the soil? If so, what kind (grasses, broad-leaf plants, woody shrubs, weeds, crops) and what size (overhead, several feet, close to the ground)? Do the plants shade the soil?
   - Is there evidence of prior or current cultivation?

2. Unbury the litter bags. This should be done very gently, as the paper is likely to be very fragile.

3. Gently brush soil from discs. Visually estimate the percentage of the disc remaining.
4. Record results and calculate the average percentage of the disc remaining for each habitat, using the data sheet supplied.

DISCUSSION QUESTIONS

1. After retrieving the litter bags, ask students to offer hypotheses about why the disks decompose more rapidly in some habitats than others:
   a. What environmental factors might have influenced the results?
   b. What management factors might have influenced the results?
   c. Can you see any signs of biological activity on the disks (e.g., fungal mycelia, soil animals, invertebrate feces)?

2. What do the results suggest about nutrient cycling rates in the soils tested?

3. Can these observations for cellulose decomposition rates be extrapolated to other types of organic matter?

4. What are the limitations of this method?
Demonstration 2: Soil Respiration

for the instructor

OVERVIEW

Soil microbes breathe in oxygen and breathe out carbon dioxide; using Draeger gas detection tubes to measure the carbon dioxide output gives an indication of the relative activity of the microbes. The brief instructions below point the instructor to comprehensive directions in the Soil Quality Test Kit Guide published by the NRCS: www.nrcs.usda.gov/wps/portal/nrcs/detail/soils/health/assessment/?cid=nrcs142p2_053873. Follow the link to Soil Respiration Test for directions and photos.

MATERIALS

See Soil Respiration Test in the NRCS Soil Quality Test Kit Guide for the full list. Among more common items such as a soil thermometer and stopwatch, you will also need to construct 6-inch diameter rings with fitted lids that have holes with stoppers, allowing equipment such as soil thermometers and Draeger tubes to be inserted. The Draeger tubes will need to be specially ordered, e.g., from a scientific supply company (see Sources of Supplies at the end of this outline). Students may record measurements in the Soil Respiration Data Sheet in Appendix 4.

PREPARATION

Here, too, see the Soil Respiration Test (see directions in the Soil Quality Test Kit Guide: www.nrcs.usda.gov/wps/portal/nrcs/detail/soils/health/assessment/?cid=nrcs142p2_053873. Follow the link to Soil Respiration Test for directions and photos for the complete preparation needed for the demonstration. In addition to gathering and constructing materials, the site will need to be brought to proper soil moisture. From the test guide: “Microbial activity is greatest when the soil is moist (at or near field capacity). If the soil is dry, a second respiration measurement should be made at a minimum of six hours (preferably 16 to 24 hours later) after the infiltration test or wetting of the soil. If the soil is saturated, soil respiration is inhibited, and this test should not be run.” To save time during the demonstration, rings can be placed and soils wetted the previous day. It may be useful to combine the litter bag (demonstration 1) and soil respiration measurements, allowing students to compare results from two different methods of measuring soil biological activity.

Locate best sites to use before the demonstration. As with the litter bag demonstration, select a variety of habitats to test, such as raised garden beds, cultivated fields, fallow fields, orchards, compost piles, vermicompost bins, and weedy borders.

PROCEDURE

Divide class in teams of two or more, and assign each team to one sample site. Demonstrate the technique first using equipment prepared at different stages, à la Julia Child. Use one ring to show how rings should be placed and head-space measurements taken. Have a second ring already
placed and capped so you can demonstrate how
to collect a CO\textsubscript{2} sample. Then send teams out to
do their own sampling, using the Soil Respiration Data Sheet (Appendix 4) to record their measurements.

See the Soil Respiration Test (online) for details on how to perform the tests, including preparing the sample area, inserting the rings in the soil, preparing the rings for measurement, taking the measurements, and using the Draeger tubes.

**CALCULATIONS**

Soil Respiration (lb CO\textsubscript{2} - C/acre/day) =

PF \times TF \times (\%CO\textsubscript{2} - 0.035) \times 22.91 \times H

PF = pressure factor = 1

TF = temperature factor = (soil temperature in Celsius + 273) / 273

H = inside height of ring = 5.08 cm (2 inches)

If a laptop is available in the field, students can enter the data into a spreadsheet and do these calculations. Calculators could also be used with printed spreadsheets in the field.

**PREPARATION TIME**

1–2 hours (varies depending on what materials are available)

**DEMONSTRATION TIME**

1–1.5 hours

**DISCUSSION QUESTIONS**

1. Compare soil respiration results for different sites. How may management practices on the different sites have influenced results?

2. If measurements were made before and after wetting soil, compare before and after results. How does soil moisture influence biological activity?

3. Would it be possible to estimate all carbon imports and exports to a soil ecosystem? What information would you need to start to make such an estimate?

**SOURCES OF SUPPLIES**

Draeger tubes, latex tubing, hypodermic needles:

Fisher Scientific, Pittsburgh, PA

www.fishersci.com

(800) 766-7000

Draeger tubes:

Scientific Industries
2207 Blue Bell Ave. Boulder, CO 80302

(303) 443-7087
Demonstration 2: Soil Respiration

step-by-step instructions for students

INTRODUCTION

Soil is alive, teeming with organisms that are eating, growing, breathing, and reproducing. Many of these organisms, from microorganisms such as bacteria and archaea, to macroorganisms such as earthworms and insects, and even plant roots, take in oxygen (O\textsubscript{2}) and release carbon dioxide (CO\textsubscript{2}). The release of CO\textsubscript{2} from the soil is called soil respiration and is a key component of healthy agroecosystems.

Soil respiration can be limited by soil moisture, temperature, and oxygen availability. Optimal respiration rates usually occur around 60% of water-filled pore space, with lower rates when the soil is either dry or saturated with water. Biological activity doubles for every 18°F rise in temperature until the optimal temperature is reached, although this optimum level varies for different organisms. Activity then declines as temperature rises above optimum. The most efficient soil organic matter decomposers are aerobic, so soil respiration rates are highest where there is high O\textsubscript{2} availability, such as in well-aggregated soil with many macropores, and decline when O\textsubscript{2} concentrations are low, as in soils that are saturated with water. Note that soil respiration is highly variable both spatially and seasonally, especially as soil moisture, temperature, and oxygen availability change, so it’s important to keep these factors in mind when interpreting your results.

Soil respiration also depends on the availability of decomposable organic substrates, that is, all the bits of organic matter of various sizes that are food for micro- and macroorganisms. Additions of organic materials will generally increase soil respiration. Organic materials with low carbon to nitrogen (C:N) ratios (e.g., manure, leguminous cover crops) are easily decomposed, so the addition of these materials to soil will increase soil respiration quickly. Materials with high C:N ratios (e.g., compost, sawdust) decompose more slowly but provide a more stable, long-term supply of organic material than legumes and manure. C:N that is too high has drawbacks: Soil microbes will compete with crop plants for the limited nitrogen supply when soil is amended with products having C:N ratios higher than 25:1.

The history of the sampling site is also important. Tillage or cultivation loosens the soil and creates better O\textsubscript{2} accessibility, increasing decomposition of organic matter and respiration rates. However, high respiration rates without adequate replenishing of organic materials can result in net loss of soil carbon. Use of agricultural chemicals that directly kill or otherwise impair soil microorganisms, such as fungicides and nematocides, on the site is also important to consider. Although these pesticides target pathogenic organisms, they may also impair the beneficial organisms and temporarily decrease soil respiration.
Management factors influencing soil respiration

INCREASES SOIL RESPIRATION
- Adding organic amendments, such as cover crops, composts (including composted manure), and crop residues
- Irrigating to proper moisture content
- Tillage

DECREASES SOIL RESPIRATION
- Removing or burning crop residues
- Continuous tillage without organic matter replacement
- Chemical pesticides (e.g., fungicides and nematocides)

MATERIALS
Soil Respiration Test in the NRCS Soil Quality Test Kit Guide Procedure

Follow the instructions from the Soil Respiration Test, as provided by your instructor.

CALCULATIONS:
Soil Respiration (lb CO₂-C/acre/day) =
PF × TF × (%CO₂ - 0.035) × 22.91 × H

PF = pressure factor = 1
TF = temperature factor = (soil temperature in Celsius + 273) / 273
H = inside height of ring = 5.08 cm (2 inches) if not measured

Interpretation of soil respiration values
In general, a higher respiration rate indicates better soil quality. A high soil respiration rate, indicative of high biological activity, can be a good sign of rapid decomposition of organic residues into nutrients available for plant growth. A low respiration rate, when soil temperature and moisture are favorable for biological activity, would indicate too little organic matter input (i.e., the soil organisms have too little organic matter to consume). Some general guidelines to interpreting respiration values are presented in Table 2.13. These are only guidelines and should not be applied to every soil type and management situation.

<table>
<thead>
<tr>
<th>TABLE 2.13</th>
<th>GENERAL SOIL RESPIRATION CLASS RATINGS AND SOIL CONDITION AT OPTIMUM SOIL TEMPERATURE AND MOISTURE CONDITIONS, PRIMARILY FOR AGRICULTURAL LAND USES  (Woods End Research, 1997)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SOIL RESPIRATION (lbs. CO₂-C/ac/day)</td>
<td>CLASS</td>
</tr>
<tr>
<td>0</td>
<td>No soil activity</td>
</tr>
<tr>
<td>&lt; 9.5</td>
<td>Very low soil activity</td>
</tr>
<tr>
<td>9.5 – 16</td>
<td>Moderately low soil activity</td>
</tr>
<tr>
<td>16 – 32</td>
<td>Medium soil activity</td>
</tr>
<tr>
<td>32 – 64</td>
<td>Ideal soil activity</td>
</tr>
<tr>
<td>&gt; 64</td>
<td>Unusually high soil activity</td>
</tr>
</tbody>
</table>
These guidelines are rules of thumb, but the soil respiration rate must be interpreted within the context of other indicators. For example, if the soil has very low nitrate concentrations and high respiration rates, there may be high nitrogen immobilization (when microbes bind up nitrogen in organic forms, so it's not available to other organisms, such as plants); this can result from adding soil amendments that have high C:N ratios.

Similarly, as mentioned in the introduction, high respiration rates without adequate input of organic matter can indicate too much decomposition, leading to a decrease in the stable component of soil organic matter. This then decreases the key properties of soil organic matter, such as aggregation, cation exchange, and water holding capacity, that make it such an important part of soil health. High respiration rates can happen immediately following a tillage operation, due to exposure of organic matter to organisms and oxygen, as well as after rainfall. The increase in soil respiration is affected by the length of time the soil is dry before the rainfall event.

Spatial differences, even on a small scale, are helpful to consider in interpreting soil respiration rates. Under dry conditions, soil respiration tends to be higher in the crop row than between the rows, due to respiration from the crop roots. This difference disappears during wet conditions, when pore space is filled and oxygen availability drops in both microhabitats. However, when the soil between rows has been compacted (e.g., by wheels) and the soil is wet, soil respiration tends to be lower than in the row, because of lower soil porosity under compaction.

**DISCUSSION QUESTIONS**

1. Compare soil respiration results for different sites. How may management practices on the different sites influence results?
2. If measurements were made before and after wetting soil, compare the before and after results. How does soil moisture influence biological activity?
3. Would it be possible to estimate all carbon imports and exports to a soil ecosystem? What information would you need to start to make such an estimate?
Demonstration 3: Assessing Earthworm Populations as Indicators of Soil Quality
for the instructor

OVERVIEW

This demonstration introduces students to techniques for assessing earthworm populations as indicators of soil quality.

You have a choice of two methods for this demonstration. The shovel-count method will be more tedious for the students because they will have to sort through the soil and remove all earthworms. The vermifuge method may take a little more effort at first to gather the materials needed, but it will make the students’ work easier.

MATERIALS

SHOVEL-COUNT METHOD
- Shovels
- Earthworm Data Sheet (Appendix 5)
- Pencils

VERMIFUGE METHOD
- Sample rings†
- Clippers
- Watering can
- Scoop
- Stirring rod
- Fresh water
- Jars
- Earthworm Data Sheet (Appendix 5)
- Pencils
- Ground yellow mustard seed (available in bulk from health food stores or from herb companies)‡

† Sample rings define the sample area and prevent vermifuge from escaping sample area. A simple design is to cut the top 8–12 inches from a 5-gallon drum and weld on a piece of metal pipe that overhangs each side by 6 inches to use as a handle. The ring is pressed into the soil to 2–3 inches depth, and vermifuge is added within the sample ring. Sample rings can also be fashioned from sheet metal, housing duct pipes, or large clean paint cans with the bottom cut off.

‡ 60 ml (volume) or 32 grams of yellow mustard powder to 4.5 liters of tap water = 13 ml/1 liter or 7g/liter. 4.5 liters of vermifuge is the amount required per sample area in this demonstration.
PREPARATION

SHOVEL-COUNT METHOD
For the shovel-count method, very little preparation is required. Identify sample areas, try to collect a similar soil volume at each location, and record results.

VERMIFUGE METHOD
The vermifuge method requires more preparation. Sample rings must be obtained or made. Other materials must be gathered. To minimize the amount of time needed for the demonstration, sample rings can be set out the day before. Ideally, a minimum of 4 can be set out per habitat. Select areas with contrasting management regimes. Possible habitats include orchard, row crop, fallow, and uncultivated field soils.

To begin the demonstration, gather group at one sample ring to explain technique. Divide class evenly among the number of sample rings and have each “ring-team” collect their sample. Have one person in each team do a shovel-count at each site for comparison. Collect results and derive an average abundance per habitat. Observe species differences and discuss results.

PROCEDURE
1. Select sample area.
2. Place sample rings on the surface of the site and push them several inches into the soil.
3. Carefully clip vegetation and removed all litter from inside sample area.
4. Slowly sprinkle 4.5 liters of vermifuge into each sample area, distributing it evenly over the entire surface.
5. After all of the vermifuge solution infiltrates the soil, wait 10 minutes, and make a second vermifuge application (4.5 liters).
6. Collect all earthworms that surface inside the sample area.
7. After 10 minutes elapse since infiltration of the second vermifuge application, use a hand spade to dig through the surface layer of soil (~5 cm deep) and collected any more earthworms found there.
8. Rinse earthworms in water, drain, and store in containers inside an insulated cooler with ice packs (unless samples are to be counted in the field and returned to the sample area).
9. An alternate method that does not require a sample ring can be found in the USDA Soil Quality Test Kit Guide, which is available on the internet (see Resources section).

PREPARATION TIME
For the shovel-count method, 0.5 hour is all that is needed. For the vermifuge method, several hours or more may needed to gather materials.

DEMONSTRATION TIME
1.5–2 hours

DISCUSSION QUESTIONS
1. Most earthworm species found in farmed soils in the U.S. were not present in those soils 400 years ago. Where do you think they came from?
2. Compare your findings from different habitats. Which habitats had the most earthworms per sample area? Which had the highest diversity (greatest number of species)? Why?
3. Determine what ecological types of earthworm were present in each sample area (see Table 2.13, page 2-114). How do you think these results were influenced by soil management practices in those areas. Consider factors such as amount and type of soil disturbance, organic matter inputs, presence of surface organic layer, etc.
4. How do these findings relate to agricultural productivity and sustainability?
5. If you were in charge of management decisions for the farm soils that were sampled, would you alter any practices based on this information? Why?
Demonstration 3: Assessing Earthworm Populations as Indicators of Soil Quality

*step-by-step instructions for students*

**INTRODUCTION**

*Earthworms are representative of the many organisms that make up soil food webs, and their abundance can be an indicator of soil biological activity.*

There are a number of ways to estimate how many earthworms are living in a particular field. Perhaps the simplest is the shovel-count: turn over a shovel-full of soil and count the worms present. Dig down 8 inches to a foot, and count every earthworm you can find in the shovel-full. Do this in half-a-dozen or more spots in each soil type on your land and come up with an average for each. If you find 5 to 10 worms per shovel-full, that represents a fairly healthy earthworm community. If this is done at about the same time each year the results will give some indication of how management practices are affecting earthworm populations.

Keep in mind that earthworm populations are very patchily distributed, and their location and abundance are heavily influenced by soil moisture, temperature, organic matter, time of year, and probably several other variables such as barometric pressure. For these reasons, a sufficient number of samples must be collected in order to accurately characterize earthworm populations in a particular field. Using more standardized sampling methods may also help.

Another method for sampling earthworms uses a vermifuge, or chemical irritant, which causes the earthworms to burrow to the soil surface, where they can be collected by hand. For many years the standard vermifuge has been a very dilute solution of formalin (about 8 ml formalin in 4.5 liters of water). However, recent studies have shown that mustard powder in water can be equally as effective.

Those interested in developing an even greater depth of understanding about earthworm ecology and how it interacts with farming may want to do more than just count numbers of earthworms present. Earthworms can be classified according to some simple physical characteristics that are directly related to their ecological roles in soil. Table 2.14 (next page) highlights the three types of earthworms.

Try using Table 2.14 to determine if you have more than one type of earthworm in your samples. Most California farm soils have endogeic earthworms, but epigeic and anecic species are rare. Epigeic species are more likely to be found in fields that have a permanent organic mulch on the surface. They may be added along with composts, but are not likely to thrive in the absence of an organic cover. Anecic species are desirable because of the work they do incorporating organic matter into the soil, mixing surface and deeper soil horizons, and creating deep channels for aeration, infiltration, and easy root penetration. Anecic earthworms could be introduced by direct inoculation, but transferring blocks of soil (one cubic foot each) from an area with a large earthworm population into a farm soil might work better.

Another idea is to set aside a small portion of a farm to be managed as an earthworm reservoir. If needed, the soil could be limed to bring it near pH 7, fertilized, irrigated regularly, and a cover crop established and cut periodically to provide an organic mulch as food and cover. A population of an anecic species could be introduced into this area and built up. Nightcrawlers can be purchased from bait dealers, who generally get them from nightcrawler harvesters in the Pacific Northwest.

From this reservoir, blocks could periodically be taken and introduced into the field. This might be done each year in the fall when earthworm activity is increasing. Remember to provide an organic mulch. The rate of spread would vary with species and conditions in the field. *Lumbricus terrestris*, the nightcrawler, is capable of traveling at least 19 meters (62 feet) on the soil surface in the course of one evening foray.
MATERIALS NEEDED
Assemble materials as per instructor’s outline

SHOVEL-COUNT METHOD
1. For the shovel-count method, very little preparation is required. Identify sample areas, try to collect a similar soil volume at each location, and record results.

VERMIFUGE METHOD
1. Select sample area
2. Place sample rings on the surface of the site and push them several inches into the soil.
3. Carefully clip vegetation and removed all litter from inside sample area.
4. Slowly sprinkle 4.5 liters of vermifuge into each sample area, distributing it evenly over the entire surface.
5. After all of the vermifuge solution infiltrates the soil, wait 10 minutes, and make a second vermifuge application (4.5 liters).
6. Collect all earthworms that surface inside the sample area.
7. After 10 minutes elapse since infiltration of the second vermifuge application, use a hand spade to dig through the surface layer of soil (~5 cm deep) and collected any more earthworms found there.
8. Rinse earthworms in water, drain, and store in containers inside an insulated cooler with ice packs (unless samples are to be counted in the field and returned to the sample area).
9. An alternate method that does not require a sample ring can be found in the USDA Soil Quality Test Kit Guide, which is available in the internet (see Resources section).

<table>
<thead>
<tr>
<th>TABLE 2.14</th>
<th>THREE DIFFERENT TYPES OF EARTHWORMS</th>
</tr>
</thead>
<tbody>
<tr>
<td>GROUP</td>
<td>WHAT THEY LOOK LIKE</td>
</tr>
<tr>
<td>Epigeic</td>
<td>small; dark red or brown; fast growing move quickly</td>
</tr>
<tr>
<td>Endogeic</td>
<td>small to medium; light or no pigmentation; slower moving continuous burrows in soil; often found in root ball; generally feed and defecate below ground mixture of buried organic matter and mineral soil, decaying roots</td>
</tr>
<tr>
<td>Anecic</td>
<td>large and very muscular; wedge-shaped tail; color on front end, less on tail end; slow growing build permanent, vertical burrows that are very deep; raised midden of castings and residue marks burrow entrance</td>
</tr>
</tbody>
</table>

A visual guide to these three types of earthworms can be found here:
www.nrri.umn.edu/worms/identification/ecology_groups.html
Demonstration 4: Soil Arthropods
for the instructor

OVERVIEW
This demonstration introduces students to techniques for sampling soil arthropods and familiarizes them with their functional roles.

For this short demonstration, both of these exercises provide a hands-on, show-and-tell of soil arthropods. You should have identification keys available, and some familiarity with what kinds of animals students are likely to find.

A pitfall trap is buried so the top sits flush with the soil surface, allowing surface-dwelling arthropods to fall in. Preserving liquid can be used in the bottom of the trap to keep the arthropods there for easy removal and identification. A Tullgren funnel uses light to dry out a soil or compost sample, driving out organisms so they can be collected and identified.

MATERIALS

PITFALL TRAPPING
• Cups (e.g., 500 or 1000 ml plastic drink cups; 1 per trap if one-time installation, or 2 per trap if you wish to repeat the trapping)
• Trowel
• (Optional) Preserving agent, e.g., ethanol or propylene glycol, especially for demonstrations that will run several days or more from start to finish. You need enough for 5-10 cm in the bottom of each trap.

TULLGREN FUNNELS
• Funnels (1 for each Tullgren funnel set-up)
• Light source (4 to 40 watt —7 watt “Christmas” style lights work well; each funnel needs its own light source)
• Aluminum foil (if light source does not have a shade for focusing the heat on the sample)
• Screen (1 piece per funnel)
• Jars (1 per funnel)
• Ethanol or propylene glycol. You need enough for 10–20 cm in the jar below the funnel.
† Steep-sided funnels with no seams work well; inverted soda bottles work, and inverted polypropylene Erlenmeyer flasks with bottoms removed are excellent. Use 500 ml flasks for 5 x 5 cm soil cores, and 2000 ml flasks for compost or litter samples.
‡ Examples for screen material are fine hardware cloth or plastic needlework backing—it should be fine enough to allow small organisms to pass through (e.g., less than 1-cm openings). The screen should be trimmed to fit across the mouth of or in the middle the funnel, as its role is to hold the sample in place in the funnel while it dries out.

FOR BOTH METHODS
• Dissection microscopes or hand lenses
• Identification guide, e.g.,
  www.cals.ncsu.edu/course/ent525/soil/soilpix/index.html
• Soil Arthropod Data Sheet (Appendix 6)
PREPARATION

PITFALL TRAPPING

Select sampling areas in different habitats. Try for a minimum of 3 or 4 samples per habitat. Traps can be set 24 to 48 hours in advance of the demonstration. Traps can also be collected before the demonstration if time is at a minimum, although it will help students contextualize their results if they can see where and how the traps were set.

At each sampling site, bury a cup so that the top edge is flush with the soil surface; to help prevent the cup from filling with soil, you can bury two cups together, one inside the other, and then remove the top cup to have a clear working cup below. If you are using two cups for your trap, set the lower one down enough so the top cup is flush with the soil surface. The top of the cup may be left open, covered with hardware cloth, or covered with a board, leaving enough room between the board and pitfall trap for free access by surface roaming creatures (e.g., by propping the board up with a stone under each side).

If collected frequently, pitfalls may be left empty so that live specimens are obtained. You may also obtain specimens by adding 5–10 cm of soapy water in the bottom of the trap, or by using a preservative such as 70% ethanol (rubbing alcohol mixed 7:3 with water) or propylene glycol (“non-toxic” anti-freeze). Preservatives help ensure the organisms do not eat one another; propylene glycol has the added benefit of not evaporating.

TULLGREN FUNNELS

Collect samples from various habitats. You can use soil cores (approx. 5 x 5 cm²), decomposing leaf litter, or compost.

Set up one Tullgren funnel for each sample. Place a piece of screen across the mouth of or part-way down a funnel. Carefully place the sample on the screen. If too much sample material falls through the funnel, add more screens, or a piece of coarse cheesecloth below the funnel. Place a wide-mouth jar with 10–20 cm of 70% ethanol or of propylene glycol under the funnel—this is where the organisms will be collected. Place the light source above the funnel, with the light above, but not touching, the sample. Do not shake or disturb funnels, keeping the sample jars as free of soil as possible. Let samples stand in funnels with the lights on for 5–7 days.

Samples can be collected and extracted in advance of the demonstration, although as with pitfall traps, it will help students contextualize their results if they can see how the samples were collected and how the Tullgren funnels work.

PROCEDURE

Observe the collected arthropods under magnification, either with dissecting scopes or hand lenses. If live collections are made from the pitfall traps, students can observe behavioral adaptations of the animals (e.g., springing springtails, fast-moving predators like centipedes and mesostigmatid mites, and slower-moving fungal grazers like oribatid mites and millipedes). Have simple keys available for help with identification. For a quantification exercise, have students count species or functional groups and calculate a diversity index, e.g., the Shannon index (see here: www.tiem.utk.edu/~gross/bioed/bealsmodules/shannonDI.html) or Simpson’s index, to compare habitats.

PREPARATION TIME

2 hours to 7 days, depending on which exercises are followed (less time generally for the pitfall trap, more for the Tullgren funnel) and what materials are available or need to be obtained.

DEMONSTRATION TIME

From 0.5 hour for a brief show-and-tell, where students observe samples previously collected, to 1 to 2 hours if students are involved in collecting samples, observing, and quantifying.

DISCUSSION QUESTIONS

1. Can you guess which animals might be predators? Which ones might be grazers? What about their shape indicates their functional group, that is, how they feed?

2. What effects do each habitat have on the soil organisms found there? Think about the sizes of creatures, diversity, food-web interactions, and pigmentation.

3. Which habitats had the greatest abundance? Which had the greatest diversity? Why?

4. What effects do you think different soil management practices have on soil arthropods? Besides the various effects of organic matter inputs, think about the influence of physical disturbance.
Assessment Questions

1) What is soil?

2) What forms of life exist in soil ecosystems?

3) How would you define a “healthy” agricultural soil?

4) What is a soil food web?

5) What might be some negative effects of the long-term practice of monoculture cropping and the use of synthetic chemical fertilizer and pest control agents on the soil ecosystem?
Assessment Questions Key

1) What is soil?
   • An ecological system consisting of inorganic minerals (sand, silt, clay, and nutrients), pore spaces, water, gases, organic matter, living organisms, and plants.

2) What forms of life exist in soil ecosystems?
   • Bacteria, fungi, actinomycetes, millipedes, isopods, mollusks, insects, insect larvae, worms and many small vertebrate animals such as gophers, ground squirrels, moles, etc.

3) How would you define a “healthy” agricultural soil?
   • A soil with a set of desirable physical and chemical properties which has the capacity to sustain biological productivity, maintain environmental quality, and promote plant, animal, and human health. This would include many of the following general characteristics:
     a) adequate rooting depth for the crop(s) to be grown
     b) a 3–5% organic matter content
     c) maintains stable soil aggregates
     d) allows for rapid water infiltration without soil erosion
     e) a low bulk density (good structure with minimal compaction)
     f) pH between 6 and 7
     g) an extractable nutrient profile within the optimal range of physiological tolerance for the crops to be grown
     h) good water holding capacity and well-drained
     i) high soil biological diversity and activity (soil respiration)
     j) adequate supplies of labile organic matter with potentially mineralizable nitrogen
     k) seasonal soil temperatures from 60–85°F

4) What is a soil food web?
   • The entire assemblage of soil organisms (producers, consumers and decomposers) in a soil ecosystem interacting among and between trophic levels

5) What might be some negative effects of the long-term practice of monoculture cropping and the use of synthetic chemical fertilizer and pest control agents on the soil ecosystem?
   • Loss of SOM, reduction in soil aggregation, reduction in nutrient- and water-holding capacity, reduction in soil biological diversity and activity, increased pest and disease incidence
Resources

PRINT RESOURCES


The best textbook introduction to the subject that I know of. Gives an overview of the basics, and attempts to consider the applications.


A weighty tome, with chapters including taxonomic keys and basic biology/ecology on virtually all organisms found in soils.


Soil quality is the current buzzword in soil science circles. This volume explores the application of the idea to sustainable environmental management.


More hands-on and less academic than the above works, this book is aimed at plant growers and has lots of practical information.


An overview of US soils, from soil biota to bankrupt farmers, done in classic NG style, with lots of great photos and drawings.


Provides a brief overview of the most commonly used conventional agricultural practices and the environmental and agroecological consequences of their use.


WEB-BASED RESOURCES

Appropriate Technology Transfer for Rural Areas (ATTRA)

atra.ncat.org/publication.html#soils

Colorado State University Extension, Garden Notes: The Living Soil

www.ext.colostate.edu/mg/gardennotes/212.html

Introduces various types of beneficial soil organisms and their roles, as well as how to encourage beneficial organisms by creating a favorable soil environment.

European Atlas of Soil Biodiversity

eusoils.jrc.ec.europa.eu/library/maps/Biodiversity_Atlas/

Information-rich resource on soil organisms offers a comprehensive guide to soil biology, soil ecosystem functions, and the ecosystem services that soil organisms provide (e.g., nutrient cycling).

Food and Agriculture Organization (FAO) of the United Nations


Discusses the effect of different agricultural practices on soil organisms. Includes information on how to improve soil biodiversity through soil management, sustainable agriculture, and agroecological farming options.

www.fao.org/docrep/009/a0100e/a0100e0d.htm

Describes the categories and characteristics of soil organisms, including beneficial and harmful organisms in agricultural soils. Includes a
discussion of the effects of organic matter on soil properties.

Great Lakes Worm Watch!
www.nrri.umn.edu/worms/identification/ecology_groups.html

Excellent description of earthworm ecological groups

Nature Education, The Rhizosphere – Roots, Soil and Everything in Between
www.nature.com/scitable/knowledge/library/the-rhizosphere-roots-soil-and-67500617

Comprehensive description of the rhizosphere, including an excellent description and graphics of legume-rhizobia symbiosis, mycorrhizal fungi and nutrient acquisition, and root system architecture. Includes a useful glossary

USDA, Natural Resources Conservation Service

Resources and publications on soil health, including information sheets and technical notes on soil organic matter, soil erosion, soil biodiversity, and soil quality evaluation

Thorough discussion of the soil food web, written by Elaine Ingham.

SOIL QUALITY

Appropriate Technology Transfer for Rural Areas (ATTRA)
www.attra.org/attra-pub/soil-lab.html#Soil%20Health

Illinois Soil Quality Initiative (ISQI)
www.aces.uiuc.edu/~asap/resources/isqi/soil-health.html

Life in the Soil
www.crslm.waite.adelaide.edu.au

Soil and Health Library
www.soilandhealth.org/index.html

Soil Biological Communities
www.blm.gov/nstc/soil/index.html

Soil Ecology Society
vax.wcsu.edu/ses/ses.html

Soil Quality Institute—NRCS

The Soil Foodweb: Its Importance in Ecosystem Health, Elaine Ingham
www.rain.org/~sals/ingham.html

University of California Sustainable Agriculture Research and Education Program (UC SAREP)
www.sarep.ucdavis.edu/soil/websites.htm

SOURCES OF SUPPLIES

Fisher Scientific, Pittsburgh, PA
www.fishersci.com
(800) 766-7000

Scientific Industries
2207 Blue Bell Ave. Boulder, CO 80302
(303) 443-7087
### Appendix 1: Major Organic Components of Typical Decomposer Food Sources

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Appendix 2: Litter Bag Data Sheet

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<th>% REMAINING (VISUAL ASSESSMENT)</th>
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<th>OTHER OBSERVATIONS</th>
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## Appendix 3: Litter Bag Data Sheet Example

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Soil Respiration Data Sheet

**Soil Respiration (at Initial Field Water Content)**

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<tr>
<th>Date</th>
<th>Sample Height</th>
<th>Sample Ring Start</th>
<th>Sample Ring End</th>
<th>Soil Temperature ºC</th>
<th>% CO₂ (n=5) lbs C/A/day*</th>
<th>% CO₂ (n=1) lbs C/A/day*</th>
<th>Soil Draeger Tube Start</th>
<th>Soil Draeger Tube End</th>
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**Soil Respiration (at least 6 hours after irrigation or soil wetting)**

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<tr>
<th>Date</th>
<th>Sample Height</th>
<th>Sample Ring Start</th>
<th>Sample Ring End</th>
<th>Soil Temperature ºC</th>
<th>% CO₂ (n=5) lbs C/A/day*</th>
<th>% CO₂ (n=1) lbs C/A/day*</th>
<th>Soil Draeger Tube Start</th>
<th>Soil Draeger Tube End</th>
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Notes:

- **Conversion:** Degrees Celsius = 5/9 x (Degrees Fahrenheit - 32)
- **H = 5.08 cm (ft not measured)**

Note: This adjustment is necessary at elevations > 3,000 ft.; otherwise Pf = 1

Pf = Pressure Factor = raw barometric pressure in inches Hg/29.9 inches Hg

Soil respiration = Pf x ((Soil Temp ºC + 273)/273) x (CO₂% - 0.035) x 22.91 x Ring Ht = lbs CO₂/C/Aday

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Notes:

- * = Soil respiration = Pf x ((Soil Temp + 273)/273) x (CO₂% - 0.035) x 22.91 x Ring Ht = lbs CO₂/C/Aday
## Appendix 5: Earthworm Data Sheet

**DATE:**

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<th>EPIGEIC EARTHWORMS</th>
<th>ENDOGEIC EARTHWORMS</th>
<th>ANECIC EARTHWORMS</th>
<th>TOTAL EARTHWORMS</th>
<th>EARTHWORMS PER SQ METER</th>
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**NOTES:**

Epigeic: Small; dark red or brown color; fast growing; move quickly

Endogeic: Small to medium; light or no pigmentation; slower moving

Anecic: Large and very muscular; wedge-shaped tail; color on front end, less on tail end; slow growing
Appendix 6: Arthropod Data Sheet

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