Introduction to Soil Chemistry

Analysis and Instrumentation

ALFRED R. CONKLIN, Jr.

A JOHN WILEY & SONS, INC., PUBLICATION

Introduction to Soil Chemistry

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This book is dedicated to my
wife Petra
son Russ
and
daughter Petal

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PREFACE

The author is both a soil scientist and a chemist. He has taught courses in all areas of chemistry and soil science and has analyzed soil for organic and inorganic compounds in both soil solids and extracts, using various methods and instruments, for 37 years. *Introduction to Soil Chemistry: Analysis and Instrumentation* is the result of these 37 years of experience that have taken place in two distinct climatic zones in the Philippines, four countries in Africa, and one in Central America. In the United States it includes analysis of soils from all sections of the country.

This book is intended as a reference for chemists and environmentalists who find that they need to analyze soil, interpret soil analysis, or develop analytical or instrumental analysis for soil. Soil scientists will also find it valuable when confronted by soil analyses that are not correct or appear to be incorrect or when an analysis does not work at all.

There are two themes in this work: (1) that all soil is complex and (2) that all soil contains water. The complexity of soil cannot be overemphasized. It contains inorganic and organic atoms, ions, and molecules in the solid, liquid, and gaseous phases. All these phases are both in quasi-equilibrium with each other and constantly changing. This means that the analysis of soil is subject to complex interferences that are not commonly encountered in standard analytical problems. The overlap of emission or absorption bands in spectroscopic analysis is only one example of the types of interferences likely to be encountered.

Soil is the most complicated of materials and is essential to life. It may be thought of as the loose material covering the dry surface of the earth, but it is much more than that. To become soil, this material must be acted on by the soil forming factors: time, biota, topography, climate, and parent material. These factors produce a series of horizons in the soil that make it distinct from simply ground-up rock. Simply observing a dark-colored surface layer overlying a reddish lower layer shows that changes in the original parent material have taken place. The many organisms growing in and on soil, including large, small, and microscopic plants, animals, and microorganisms also differentiate soil from ground-up rock.

There are other less obvious physical changes constantly taking place in soil. Soil temperature changes dramatically from day to night, week to week, and season to season. Even in climates where the air temperature is relatively constant, soil temperatures can vary by 20 degrees or more from day to night.

Moisture levels can change from saturation to air dry, which can be as low as 1% moisture on a dry-weight basis. These changes have dramatic effects on the chemical reactions in the soil. Changes in soil water content alter the concentration of soil constituents and thus also their reaction rates.

Not only are soil's physical and observable characteristics different from those of ground-up rock; so also are its chemical characteristics. Soil is a mixture of inorganic and organic solids, liquids, and gases. In these phases can be found inorganic and organic molecules, cations, and anions. Inorganic and organic components can be preset as simple or complex ions. Crystalline material occur having different combinations of components from, for example, 1:1 and 2:1 clay minerals, leading to different structures with different physical and chemical characteristics with different surface functionalities and chemical reactivities.

Organic components range from the simple gaseous compounds, such as methane, to very complex materials, such as humus. Included in this mix are gases, liquids, and solids, and hydrophobic and hydrophilic molecules and ions. All organic functional groups are included in soil organic matter, and it is common to find polyfunctional organic molecules as well as simple and complex biochemicals. Humus is an example of a complex molecule that contains many different functional groups. Polyfunctional organic molecules and biochemicals coordinate and chelate with inorganic materials in soils, particularly metals.

The fact that soil always contains water, or more precisely an aqueous solution, is extremely important to keep in mind when carrying out an analytical procedure involving soil because water can adversely affect analytical procedures and instrumentation. This can result in an over- or underdetermination of the concentrations of components of interest. Deactivation of chromatographic adsorbants and columns and the destruction of sampling tools such as salt windows used in infrared spectroscopy are examples of the deleterious effects of water. This can also result in absorbence or overlap of essential analytical bands in various regions of the spectrum.

All of these physical and chemical characteristics have a pronounced effect on soil and its analysis. The intention here is to first investigate some of the most important soil characteristics that impact its analysis and instrumentation applied to analysis of the soil as well as its extracts, and to elucidate those interferences that may be most troubling to the analysis.

Chapters conclude with a bibliography, followed by a list of references. The bibliography lists general sources for the material covered in the chapter, while the references give some specific examples illustrating the application to soil. These lists are intended to provide the reader with additional resources and examples of how the material covered in the chapter is actually used in soil analysis and research. They are also a source of standard methods and procedures of soil analysis and provide the reader with many analytical procedures along with pitfalls and interferences that may be encountered in the particular analysis being discussed.

The Internet references given have been checked and were found accurate at the time of writing. However, Internet addresses are subject to change. If you are unable to find an address, try accessing the parent organization and looking for the desired information through its home page. For instance, if the Internet address is a USDA (United States Department of Agriculture) site, you can access the USDA Web site and find the needed information from there.

The author wishes to thank D. Meinholtz, J. Bigham, N. Smeck, H. Skipper, B. Ramos, and T. Villamayer for their help in reviewing and preparing this manuscript.

Alfred R. Conklin, Jr.

CHAPTER

1

SOIL BASICS I
MACROSCALE FEATURES

Soil is essential to life. All life supporting components derive, either directly or indirectly, from the soil. Plants growing in soil are directly used for food or are fed to animals, which are then used for food. These same plants take in carbon dioxide produced by animals and give off oxygen. Soil and the plants it supports moderate the amount of liquid and gaseous water in the environment by serving as a reservoir controlling its movement. Elements essential to life, even life in water, are released from soil solids and recycled by soil chemical and biologically mediated reactions.

Thus, although, as will be seen, soil is extremely complex, an understanding of its physical characteristics and the chemistry occurring in it are important in its analysis and the instruments used in this analysis.

Soil is vastly more complex than simply ground-up rock. It contains solid inorganic and organic components in various stages of decomposition and disintegration, an aqueous solution of elements, inorganic and organic ions and molecules, and a gaseous phase containing nitrogen, oxygen, carbon dioxide, water vapor, argon, and methane plus other gases. In addition, it contains a large and varied population of macro-, meso-, and microscale animals, plants, and microorganisms. If any of these components is missing, it is not soil!

The solid portion of soil is composed of inorganic sand, silt, clay, and organic matter, which interact to produce the large soil features[1] (i.e., peds, profiles, pedons, landscapes). These features, not considering rock, are discussed in this chapter. In Chapter 2, components smaller than sand, which soil scientists define as those inorganic particles smaller than 2.00 mm in diameter, are discussed. Geologic features and gravel, stones, rock, and other substances are not discussed.

Large soil components consisting of sand, silt, clay, and organic matter are peds, profiles, pedons, and landscapes. Peds are formed by the aggregation of sand, silt, and clay particles to form larger (secondary) soil structures that result from the action of the soil forming factors (see Figures 1.1–1.5). Profiles develop in the loose material on the earth's surface and are composed of

[1] Many soils contain gravel, stones, and rock; however, these components, because of their low reactivity, will not be considered in this book.

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

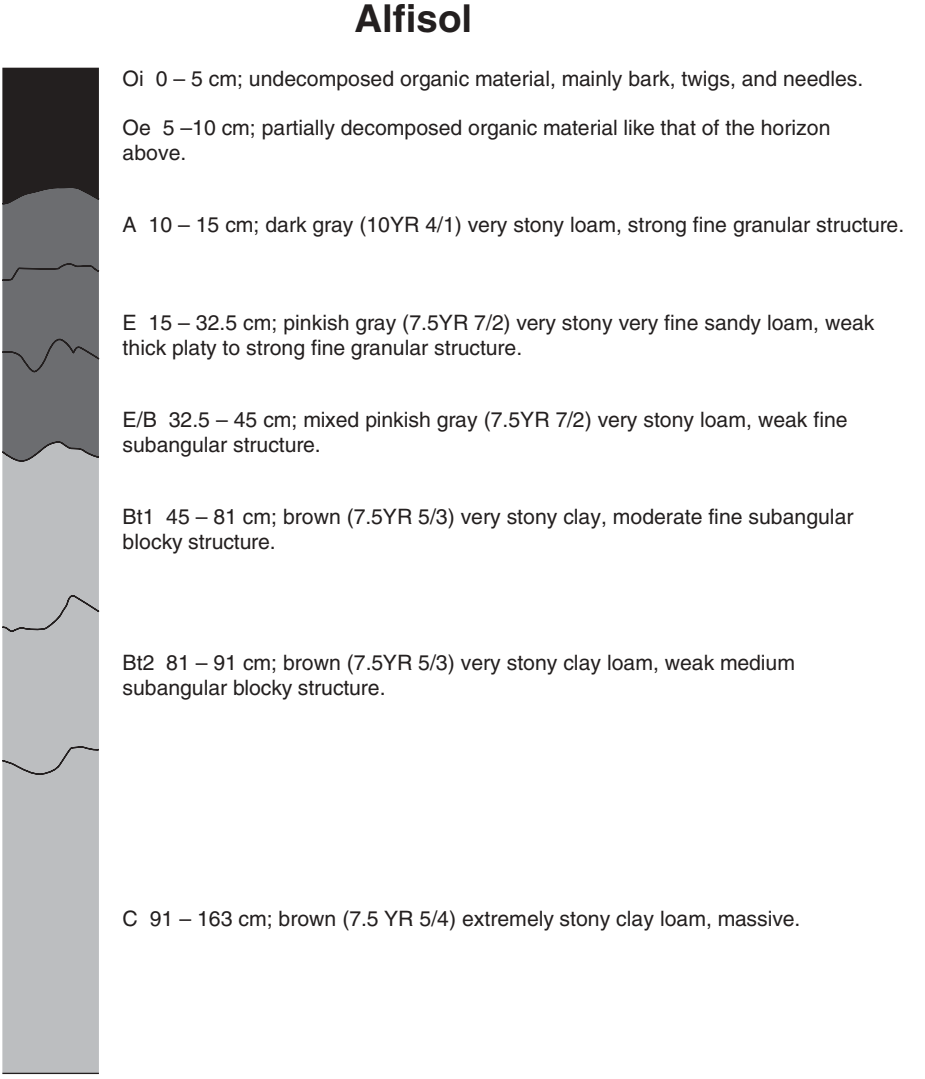


Figure 1.1. An example of an Alfisol; this is the Seitz soil series, which is the state soil of Colorado (see Ref. 2).

horizons of varying texture, structure, color, bulk density, and other properties. Typically they are, as the name implies, horizontal, and are of varying thickness. A pedon is the smallest unit that can be considered "a soil" and consists of all horizons extending from the soil surface to the underlying geologic strata. An area consisting of similar pedons is called a *polypedon*.

Soil features, texture, peds, profiles, and other properties and materials go together in different ways to form different soils. Soil scientists in the United

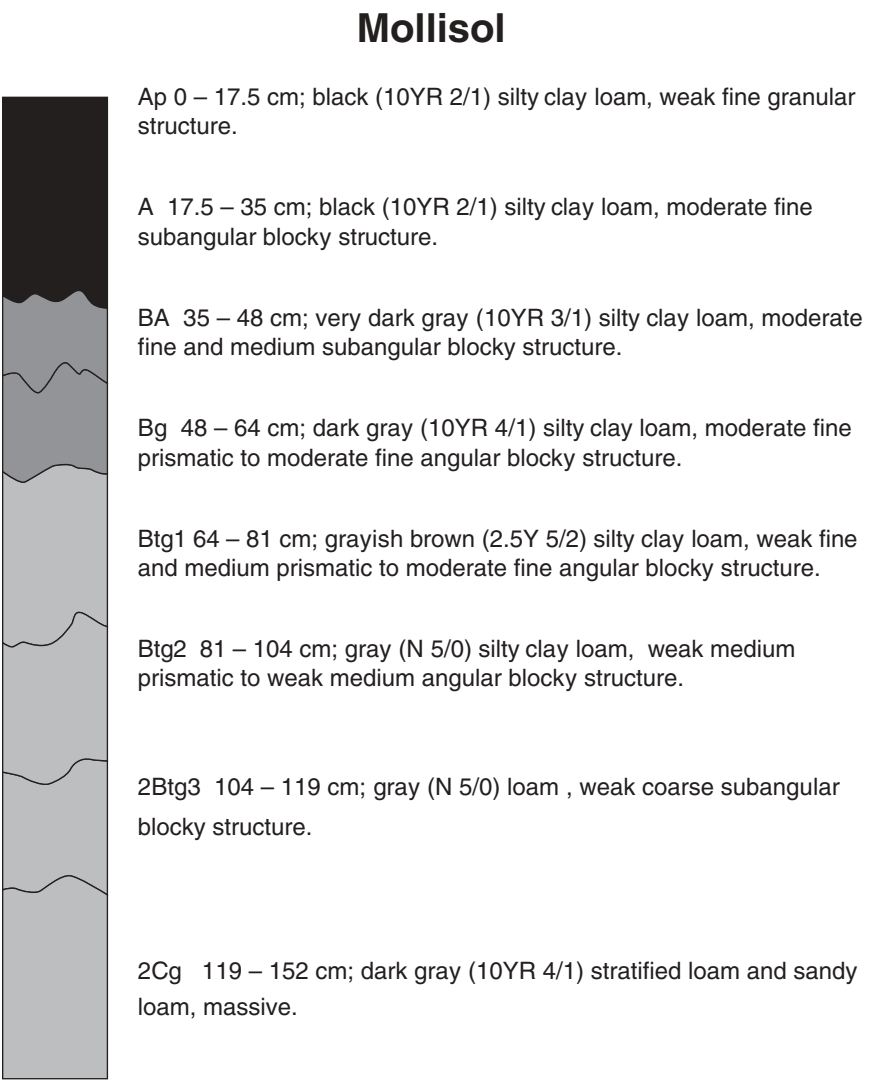
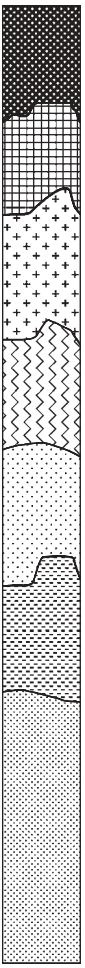


Figure 1.2. An example of a Mollisol; this is the Drummer soil series, which is the state soil of Illinois (see Ref. 2).

States classify soils into 12 orders. The orders are differentiated by their characteristics which are a result of the soil forming factors: climate, parent material, topography, biota, and time, which all interact during soil formation. Climate, moisture, and temperature, determine the biota, which can survive in the locality. However, topography and time will also determine the vegetation, as well as its age and whether it can survive in a certain locality. The soil parent

Ultisol



Ap 0 – 12.5 cm; very dark grayish brown (10YR 3/2) fine sandy loam, weak fine granular structure.

E 12.5 – 22.5 cm; yellowish brown (10YR 5/4) fine sandy loam, weak fine granular structure.

BE 22.5 – 35 cm; strong brown (7.5YR 5/6) fine sandy loam, weak medium subangular blocky structure.

Bt1 35 – 55 cm; yellowish red (5YR 4/6) sandy clay loam; weak medium subangular blocky structure.

Bt2 55 –104 cm; red (2.5YR 4/8) sandy clay loam, weak medium subangular blocky structure.

Bt3 104 – 173 cm; red (2.5YR 4/6) sandy clay loam, weak medium subangular blocky structure.

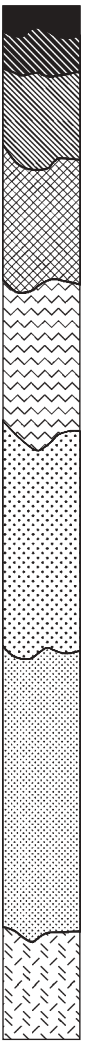
Bt4 173 – 188 cm; red (2.5YR 4/8) sand clay loam, weak coarse subangular blocky structure.

Figure 1.3. An example of an Ultisol; this is the Bama soil series, which is the state soil of Alabama (from Ref. 2).

material will be acted on by the other factors but will in turn provide the support and minerals needed for plant growth and other activity.

Consideration of the larger components of soil might seem like a strange place to start a discussion of soil chemistry, analysis, and instrumental methods. However, these larger structures can and do affect the chemistry of a soil. For instance, in many cases the larger features control the movement of air and water in soil. Sandy textured soils will have higher infiltration and percolation rates than will clayey soils. The same can be said for soils with good, strong

Spodosol



Oi 0 – 2.5 cm; undecomposed forest litter.

A 0 – 2.5 cm; black (7.5YR 2.5/1) sand, weak fine granular structure.

E 2.5 – 10 cm; brown (7.5YR 5/2) sand, weak fine granular structure.

Bhs 10 – 15 cm; dark reddish brown (5YR 3/3) sand, weak fine granular structure.

Bs1 15 – 53 cm; dark brown (7.5YR 3/4) sand, weak fine granular structure.

Bs2 53 – 89 cm; strong brown (7.5YR 4/6) sand; weak fine granular structure.

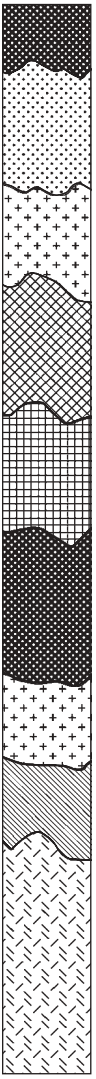
BC 89 – 127 cm; yellow brown (10YR 5/6) sand; weak fine granular structure.

C 127 – 203 cm; light yellowish brown (10YR 6/4) sand, single grain.

Figure 1.4. An example of a Spodosol; this is the Kalkaska soil series, which is the state soil of Michigan (single-grain structure in horizon C means that all the sand particles act independently of each other) (from Ref. 2).

structure versus soils with poor or weak structure. As water moves into soil, it displaces air, and as it moves out, air replaces it. Thus soil with poor or slow infiltration and percolation will generally have lower oxygen content. This directly affects soil aeration and thus its oxidation–reduction status.

Aridisol



An 0 – 2.5 cm; light yellowish brown (10YR 6/4) fine sandy loam, moderate thin platy structure.

BAn 2.5 – 12.5 cm; pale brown (10YR 6/3) fine sandy loam, strong medium and coarse angular blocky structure.

Btknz1 12.5 – 20 cm; light yellowish brown (10YR 6/4) sandy clay loam, moderate medium prismatic structure.

Btknz2 20 – 27.5 cm; light yellowish brown (10YR 6/4) sandy clay loam, moderate medium prismatic structure.

Btknz3 27.5 – 45 cm; strong brown (7.5YR 5/6) clay loam, strong medium prismatic structure.

Btknz4 45 – 81 cm; strong brown (7.5YR 5/6) clay loam, strong medium prismatic structure.

Btknz5 81 – 99 cm; brown (7.5YR 5/4) clay loam, moderate medium and coarse angular blocky structure.

2BCnzl 99 – 117 cm; light yellowish brown (10YR 6/4) sandy clay loam, moderate medium subangular blocky structure.

2BCknz2 117 – 152 cm; light brown (7.5YR 6/4) sandy clay loam, weak medium subangular blocky structure.

Figure 1.5. An example of an Aridisol; this is the Casa Grande soil series, which is the state soil of Arizona (from Ref. 2).

Infiltration and percolation rates also determine which salts have been leached out of the soil. For instance, high infiltration and percolation rates leach calcium and magnesium out of soil and they become acidic. Where calcium and magnesium are not leached out of soil, the soils are neutral

or basic. Thus, the type and amount of salts present will affect a soil's pH, which will in turn affect the solubility and availability of both natural and contaminating inorganic and organic compounds.

The species of components present will also be affected by oxidation–reduction and pH. For example, iron is primarily in the Fe^{3+} or the Fe^{2+} state depending on the oxidation–reduction potential of the soil. Speciation, which depends in part on the oxygen status of soil, is of environmental concern because some species are more soluble and thus more biologically available than others. The occurrence of a specific species is related to the chemistry occurring in a soil, which is related to its features. Thus, large features must be considered when developing analytical and instrumental methods.

1.1. HORIZONATION

The most striking large feature of most soils is the occurrence of distinct horizons. For the analytical chemist, three soil horizonation conditions exist. High-rainfall areas typically have tree or tall grass vegetation and extensive horizon development. Low-rainfall and desert areas with sparse and desert vegetation with little horizon development. Areas with rainfall between these extremes will have variable vegetation and horizonation. It is not possible to draw sharp boundaries between these areas because local conditions such as frequency, time of year, and intensity of the rainfall will dramatically affect the climate in transition areas. Each of these situations presents unique challenges to the analytical chemist.

1.1.1. Horizon Development under Moist Conditions

When water falls on soil, parent material salts are dissolved. These salts are leached into and eventually out of the soil. Plants grow, produce biomass, exude organic matter (including acids), die, and thus add organic matter to both the soil surface and subsurface. Silica and alumina, which are relatively immobile, slowly dissolve and are eluted into the lower areas of a developing soil. Organic matter is decomposed, mixed by macro- and microorganisms with and leached into the soil. As this process continues, horizons develop in the soil parent material and eventually a recognizable soil profile is produced (see Figures 1.1–1.5). The depth of the soil and the number and types of horizons that develop depend on the soil forming factors, the most active of which are climate and biota, although parent material, as mentioned above, is also very important [1,2].

Soil parent material is not always derived from the underlying rock. In some cases rock is covered by other geologic materials deposited by ice (glacial till), water (alluvium), wind (loess), gravity (colluvium), or a combination of these transporting agents. A more complete list of transporting agents and the geo-

Table 1.1. Soil Parent Material Transporting Agents, Including Name of Material and Geomorphic Features They Form[a]

Agent	Name Applied to Material	Geomorphic Features
Gravity	Colluvium	Toe slope of hills
Air	Loess	Loess and loess cap
	Dune sand	Sand dunes
	Volcanic ash	Volcanic ash layers
Water	Alluvium	Floodplains
		Deltas and alluvial fans
	Lacustrine	Lake sediments
	Outwash	Terraces, outwash planes, kames, and esker
Ice	Glacial till	Till plains
Ocean water	Marine sediments	Costal plains

[a] This is not an exhaustive list.

morphic features they form is given in Table 1.1. Once deposited, these materials become the parent material from which soil will develop.

It is logical to assume that the surface horizons in a soil will be made up of material derived from the parent material and plants growing on the soil. In most, if not all, cases this is not true. Many soils have continuous additions of both inorganic and organic material deposited from both water and wind. In Ohio (USA) many of the soils develop from an underlying glacial till and an overlying silt loess, wind-transported silt, cap. In other areas occasional or regular flooding may deposit material on the soil surface, which then becomes part of the *soil surface horizons*. Even in areas where there would appear to be little wind- or water-transported material, small amounts of both inorganic and organic compounds will be added to soil from the atmosphere.

The first horizon develops on the surface of the soil and is called the A horizon. Because it has organic matter deposited on and in it and is the first to have salts dissolved and eluviated, it is higher in organic matter and lower in salts than the lower horizons. Clay is eluviated out of this horizon and is deposited lower in the profile. It also has finer granular and crumb structure. This is the horizon, which is plowed in preparation for planting crops[2] and is the one most commonly sampled for analysis.

The A horizons are slightly different from other horizons because they are plowed and planted. In many cases a soil may have an Ap horizon, indicating that it is or has been plowed. The plowing need not have been done recently for the horizon to be described as Ap. Even a soil so newly developing that it has no horizons will have an Ap horizon designated if it is plowed.

[2] Soils may be worked in a number of different ways other than plowing. In addition, they may not be worked at all when no till planting in used. In this case soil disturbance is minimal.

Table 1.2. Major–Minor Horizon Designation in Soil Profiles[a]

Designations	Characteristic
	Major
O	Organic horizon may be partially or totally decomposed organic matter
A	Topmost inorganic horizon characterized by relatively high organic matter content
E	Interior horizon characterized by eluviation
B	Subsoil horizon showing evidence of weathering or illuviation
C	Lowest horizon in geologic material underlying the soil showing little evidence of soil formation
	Minor (*Subdesignations*)
p	Plowed—applied only to topmost or A horizon
t	Accumulation of illuvial clay
a	Highly decomposed organic matter
h	Illuvial accumulation of organic matter
s	Illuvial accumulation of oxides of iron and aluminum
i	Slightly decomposed organic matter
e	Intermediately decomposed organic matter
g	Gleying
k	Accumulation of carbonates (lime)
n	Accumulation of sodium ions
z	Accumulation of soluble salts

[a] Partial listing.

The small letter p, which stands for plow, is not used with any other horizon designation.

With the passage of time a full set of horizons will develop in a regolith or parent material. Major horizon designations and some of their distinguishing characteristics are given in Table 1.2. Each horizon may be subdivided on the basis of other distinguishing characteristics. The O, or organic, horizons can be subdivided into those where the organic matter is just beginning, Oi, is well along, Oe, or highly decomposed, Oa. Examples of these horizons can be seen in Figure 1.1. Likewise, the B horizons can be characterized by increased clay content, Bt, or reducing conditions, Bg [the meaning of the upper- and lowercase letters are described in Table 1.2]. When two horizons are similar but still distinguishably different, a number following the major horizon designation indicates this. For example, a soil may have several Bt horizons designated by Bt1, Bt2, Bt3. A number in front of a horizon designation indicates that it is formed or is forming from a different parent material. Examples of these different horizons can be seen in Figures 1.1 and 1.2.

Figure 1.1 also has two other distinctive horizons. The E horizon, which stands for *eluviated*, is characterized by depletion of clay and is lighter in color and coarser in texture than the over- or underlying horizons. The second dis-

tinctive horizon is the E/B. This designation indicates a *transition horizon*, which contains characteristics of both the overlying E, and underlying B horizons. There are other designations of transition horizons such as AB. Whichever letter comes first indicates that that horizon characteristic dominates the transition horizon.

The small (lowercase) letter subordinate horizon designators are used wherever appropriate to designate significant differences or distinctions in a horizon (see Table 1.2). A common designation is the small letter t, which stands for the German word *ton*, which means clay. This is a B horizon, where there is a significant increase in clay when compared to the overlying horizons. Another designation, shown in Figure 1.2, is the small letter g, which stands for *gleying*, also called *mottling*. This is a situation where varying colors develop under reducing conditions, which result when the soil is saturated with water for a significant period of time. In many cases the colors are varying shades of red or rust-colored spots. However, sometimes grays and even blues can occur depending on the soil.

In Figure 1.4 the profile description shows an E horizon and a B horizon with small letter hs designation and a B horizon below this with a small letter s. The E horizon is light in color and, if light enough, might also be called an *albic horizon*. The small letter h indicates an accumulation of highly decomposed illuvial organic matter, and the small letter s refers to the accumulation of oxides of illuvial iron and aluminum.

Many other designators are used for various horizons and horizon conditions. A complete list of these will not be given here but can be found in any introductory soils text or book on soil classification.

In naming soils, diagnostic horizons are used. These terms are different from those used in a profile description such as those given in Figures 1.1–1.5. However, these diagnostic horizons are indicated in the name given to the soil and are not necessarily designations that the researcher doing soil chemistry, developing analytical or instrumental procedures for soil, is likely to have occasion to see or use. If the need arises, they are easily found in the literature (see Bibliography).

1.1.2. Horizon Development under Low-Rainfall and Desert Conditions

Under low-rainfall and desert conditions the lack of rainfall results in very different soil conditions. Figure 1.5 shows the profile description of an Aridisol (an arid, desert-region soil) (see Section 1.4 for explanation of the Naming System). All lower horizons contain a high content of sodium ions and most also contain other soluble salts of various kinds.

In low-rainfall regions salts are not readily washed out of soil and may even build up on the soil surface. This happens when rain dissolves salts in the soil surface horizon and leaches them a little way into the soil. Subsequent evaporation of water from the soil surface causes water and dissolved salts to move to and be deposited on the soil surface. No salts or only the most soluble salts are leached out of the soil, in low-rainfall regions, and the pH is typically basic.

As would be expected, only limited horizon development will occur and frequently the horizons formed will be thin. However, it is possible to find soils with significant horizon development in desert regions. In some cases this soil developed when the particular area received more rainfall.

Salts, in addition to causing the soil to be basic, will, in some cases, have deleterious effects on analytical procedures. Significant error can occur if a potassium-selective electrode is used to determine potassium in a high-sodium soil. As discussed in Chapters 5 and 8, other salts could cause inaccurate results to be obtained when an atomic absorption analysis of a soil extract is carried out.

1.1.3. Horizon Development in Areas between High- and Low-Rainfall Conditions

In areas between high- and low-rainfall horizonation may be well or poorly developed, soil pH may be either acidic or basic, and there may or may not be salt buildup in the soil. In these areas the analyst must be aware of this potential variation and have the reagents necessary for all of these eventualities.

1.2. PEDS

Profile descriptions also detail the structure found in that horizon and indicate its strength. Figure 1.6 shows a soil profile with an indication of the location of the various structure types. Looking at Figures 1.1–1.5, an example of each major structure type is indicated. In most cases granular and crumb structure is expected to occur only in the top 25 cm of soil. Platy structure can be found in any horizon although it is frequently found in E horizons or in the transition zones between the A and lower horizons. Traffic, farm or other, will promote the formation of platy structure in the A horizon and often results in an angular, blocky structure at the base of the A horizon. In C horizons, platy structure is a remnant of the original character of the parent material from which the soil is forming. Subangular and angular blocky structure is typically found in the upper part of the B horizons and the prismatic structure, in the lower part. However, blocky and prismatic structure can be found in any part of the B horizons.

Peds are formed by natural aggregation of sand silt and clay particles. Although a soil's texture is defined by the relative proportions of sand, silt, and clay, these components almost never act independently of each other. The binding agents that hold peds together are clay, organic matter, particularly microbial gums, and various inorganic ions, particularly calcium, magnesium, iron, and aluminum. Planes of weakness occur between peds and are extremely important in determining a soil's characteristics because these are areas where air, water, and roots penetrate soil easily. When observing soil peds, roots are seen growing in the voids between structures (roots can be seen on the second

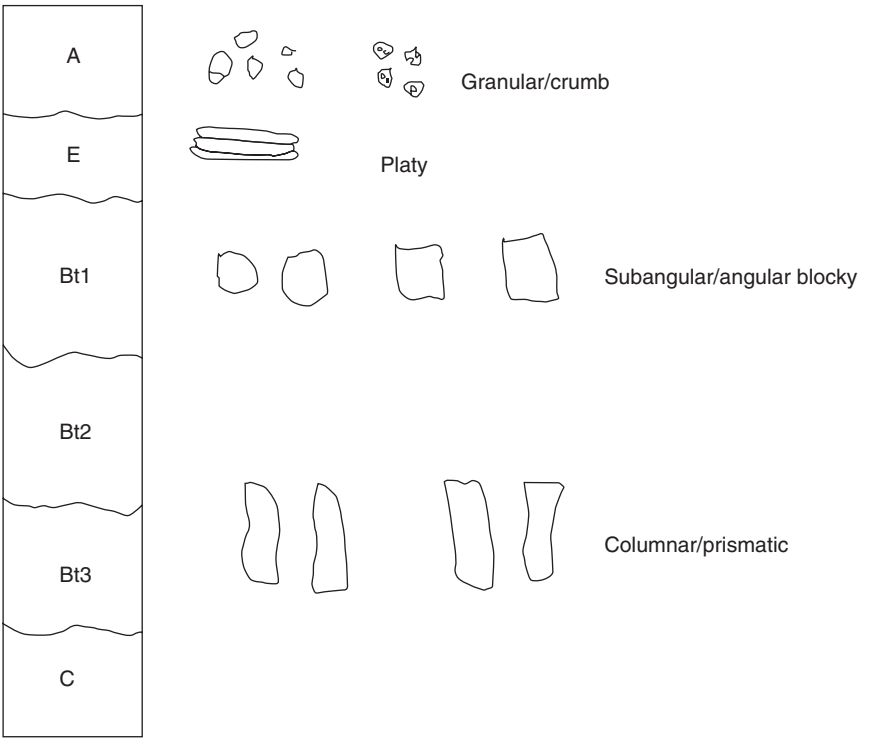


Figure 1.6. Soil structure and its most common location in a soil profile. Platy structure can be found in any horizon. Subangular and angular blocky structure can be found both higher and lower in the profile than indicated.

from the bottom peds in Figure 1.7). Because of their effect on air, water, and roots, peds help in determining a soil's chemistry.

Figure 1.7 shows actual peds isolated from a soil profile. Generally speaking, structure, except for platy peds, is considered to be a positive component in soil. Platy structure, however, can retard the movement of air and water down through the soil profile and so is considered a negative component. Because it restricts air and water movement, it may cause areas of water saturation and thus reducing conditions.

Because large voids between peds allow ready movement of water down through the soil profile, ped surfaces often become coated with material carried in the water. The most common coating is clay and is called "clay skins." If there is rubbing between the peds, the skins will have a smeared surface and will often be shiny. Under such conditions the surfaces are said to be "slicken sides." Less common are surface coatings of silt and organic matter carried by leaching into the lower horizons. Silt coatings are common in horizons from which clay particles have been translocated to lower horizons.

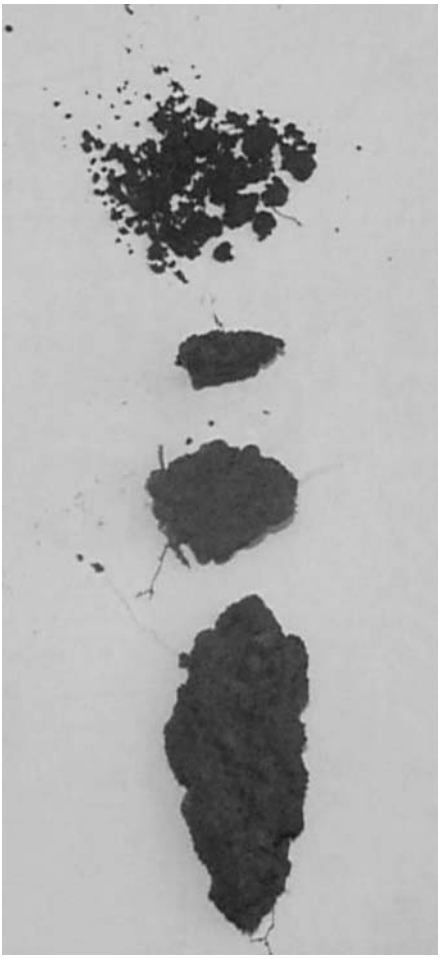


Figure 1.7. Peds isolated form soil. From top to bottom spheroidal, platelike, blocklike, and prismlike.

Organic matter coatings are common in acid sandy soils, especially those called *Spodosols*.

Coating of primary and secondary soil structures is an extremely important phenomenon for the analysis of soil. Without coating the chemical activity of soil primary and secondary particles could be determined by knowledge of their structure and makeup. With coating the chemistry becomes vastly more complex. This complexity will be discussed in later chapters.

For the soil separates (sand, silt, clay) to become aggregated, they must come close together. The forces that cause particles to come together are the weight of overlying soil, plant roots moving through the soil, heating and

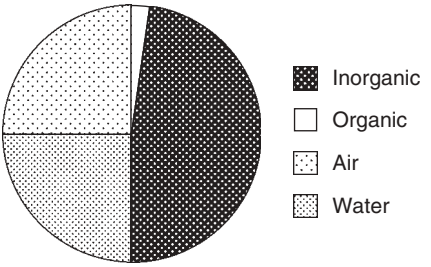


Figure 1.8. Idealized general composition of a soil sample.

cooling, freezing and thawing, and wetting and drying. It might also be argued that cultivation forces particles together and thus tends to improve structure. However, it is equally true that cultivation breaks down structure. Because of these two competing effects, the net effect of cultivation on soil structure can be either positive or negative depending on the condition of the soil, particularly wetness, when cultivated.

Ideally a well-aerated soil is considered to be half-solid and half-void space. The void space is half-filled with air and half with water. This idealized condition is illustrated in Figure 1.8. Such a soil is under general oxidizing conditions, oxidation of all components, particularly organic matter, is expected. In real soil samples the total amount of void space depends on soil texture and structure, and the amount of airspace is inversely related to the water content. When the void space becomes filled with water, the soil becomes reducing. This takes a little time because dissolved and trapped oxygen must be used up before full reducing conditions are reached. Reducing conditions are accompanied by the production of methane and, if sulfur is present, hydrogen and other sulfides.

At this point oxidation and reduction in soil seem simple. When the soil is not saturated with water, it is oxidizing and when saturated, it is reducing. However, even under oxidizing conditions reduced compounds and species are produced. How can methane be produced even in a soil under oxidizing conditions? The answer is that, in addition to simple pores, which are large in diameter and open at both ends, there are pores that have restricted openings, have only one opening, or have only one restricted opening as illustrated in Figure 1.9.

Where the pores have only one opening or restricted openings, they do not drain and the interiors are reducing even when the soil in well aerated. The reducing conditions lead to the production of species such as methane and Fe^{2+}. Although methane and iron are two of the most easily found and identified species, other reduced species, both organic and inorganic, are commonly present.

Two conditions exist where the soil separates are not aggregated to form secondary particles or peds. One is where the individual separates act inde-

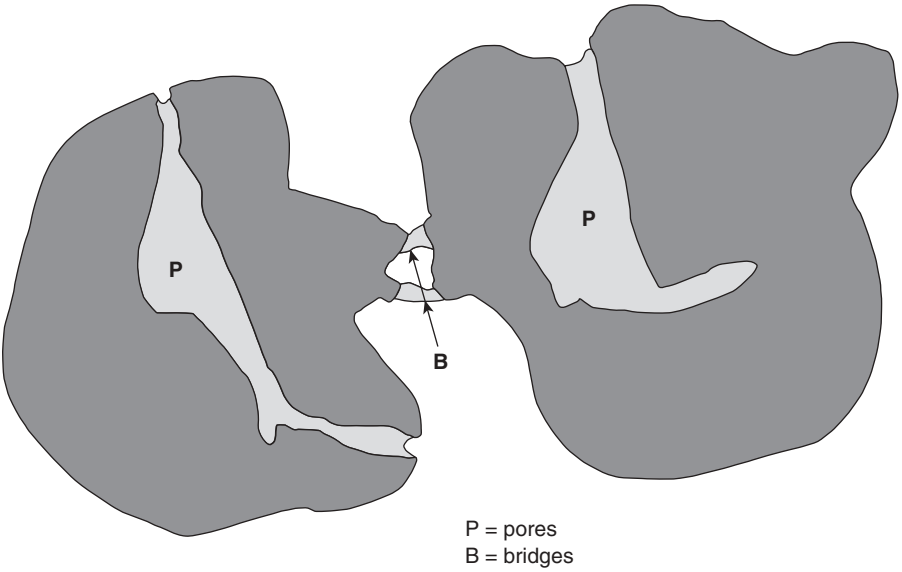


Figure 1.9. Two soil peds showing a pore with restricted openings on the left and a pore with only one opening on the right. In between are bridges holding the two peds together.

pendently from each other. This condition is called *single-grained*, like beach sand, and occurs in coarse sandy soil. The second condition is where there are no lines of weakness between units and the soil is said to be *massive*; that is, it acts as one massive block. Soil becomes massive if it is worked when it is too wet, forming hard clods that, when dry, form smaller irregular clods when broken. Neither of these conditions is common in soil, although the massive condition occurs in the soil parent material or rigolith before soil formation begins.

1.3. SOIL COLOR

In Figures 1.1–1.5 the horizon descriptions give a color in both words and in Munsell color book designations. The Munsell system describes color in terms of hue: the basic color, value (lightness), and chroma or the purity of the color. The hue is the first number followed by a color designation. In Figure 1.1 the A horizon is 10YR (where YR indicates a yellow-red color); in Figure 1.2 the Btg1 horizon has a color designated as 2.5Y (where Y indicates yellow). In the Munsell system the value for white is assigned the number 10 and black, the number 0. The chroma or purity increases from left (1) to right (8). However, not all colors in the Munsell system are used to describe soil colors.

The typical Munsell color chart for soils covers only those colors described as being yellow or red or some combination of the two. This is not to say that

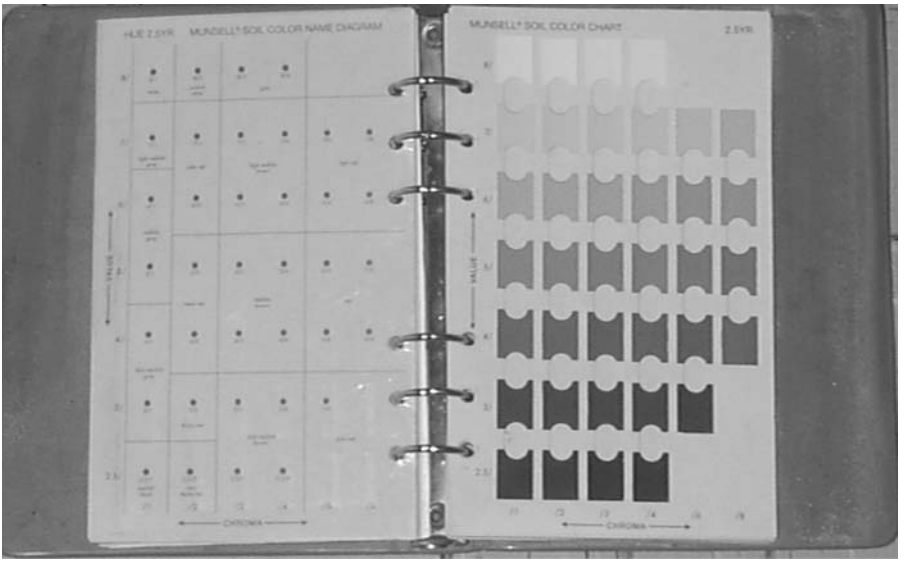


Figure 1.10. The 2.5YR (yellow-red) page of a Munsell color book. The value is from white at the top to black at the bottom. Chroma becomes higher from left to right.

other colors do not occur in soil; they do. It is common to find gray soils in which no hue occurs, and in this case only the value and chroma are indicated as shown in Figure 1.2, horizon Btg2. Under highly reducing conditions blues will sometimes be found. Most books used by soil scientists include a gley page for such soils. A picture of a page in a Munsell color book commonly used by soil scientists is shown in Figure 1.10.

Soil color deserves special attention. When rock is ground up, it produces a gray powder. Gray occurs in E and gleyed horizons under acid and reducing conditions, and thus this color provides important information when analyzing such as soil. The normal soil colors red and black are derived from iron and organic matter, respectively. Iron is in the form of oxides and hydroxy oxides (Fe_2O_3, FeOOH, etc.). Black and brown colors are normally derived from highly decomposed organic matter and humus.

As a soil develops, decomposition of organic matter (OM) occurs, producing humus, which is black; and iron is released from minerals by weathering, which yields various reds and yellows; both mechanisms yield soil coloring agents. Under oxidizing conditions, where soil is not saturated with water, the iron will be oxidized and thus in the ferric state [Fe(III)]. When the iron and organic matter are deposited on the surfaces of sand, silt, clay, and peds, they develop a coat that gives them a surface color. However, soil color is not only a surface characteristic but extends through the soil matrix. Under oxidizing conditions soil has a reddish color. The chroma of this color depends to some extent on the amount of and the particular iron oxide present.

Under most conditions little organic matter is eluviated down through the soil. However, many soils have dark or black thick upper horizons, which are high in organic matter. These horizons are found in soils developing under high water conditions and grass vegetation where the grass and associated roots die each year and contribute organic matter to the profile to a depth of $\geqslant$0.5 m depending on the type of grass and other environmental conditions.

When soil is saturated with water, the soil environment becomes reducing, and under these conditions iron is reduced to the ferrous [Fe(II)] state. The soil color becomes lighter and more yellow. Under reducing conditions soil also develops variations in color called *mottling* or *gleying*. Thus any soil horizon description, which includes a "g" designation, indicates that the soil is under reducing conditions for a significant period of time during the year. It might be expected that mottling or gleying would occur only in the lower horizons; however, it can occur anywhere in a soil profile, even quite near the surface.

Iron in the ferrous state is more soluble than iron in the ferric state; indeed, in the ferric state it is insoluble in most soil conditions . Under reducing conditions ferrous iron may be leached out of soil, leaving it gray in color. This is the origin of the term "gleying".

Some types of vegetation and environmental conditions result in acid producing litter on the soil surface. Under these conditions organic matter decomposition products (humic, fluvic acids, etc.) can eluviate and be deposited deeper in the soil. In Figure 1.4 the litter in the Oi horizon produces acid, which allows the illuviation of aluminum, iron, and organic matter decomposition products into the B horizons to form the Bhs horizon. The leaching of aluminum, iron, and organic matter out of an area of the soil profile results in the horizon becoming light gray or white, giving rise to the potential development of an albic horizon.

To some extent the description of a soil profile gives an indication of some of the chemistry and chemical conditions occurring in that profile. This in turn provides the researcher and analyst with information about the types of compounds and species likely to be found and the conditions necessary to isolate them [3].

1.4. SOIL NAMING

Various different soil naming systems are used throughout the world. In all of these systems the horizons and their subdesignations vary somewhat according to the different classification systems. In the United States the United States Department of Agriculture (USDA) has developed the USDA Soil Taxonomy system (simply referred to as *Soil Taxonomy*), which recognizes 12 soil orders. The United Nations, through its Food and Agriculture Organization and United Nations Educational, Scientific and Cultural Organization (FAO-UNESCO) and the International Society of Soil Science, has a system that includes 26 soil groupings. There are many other systems, including those

developed by Canada, France, the Former Soviet Union, China, Australia, and other countries. Each recognizes a different number of major soil groups or soil orders.

Two basic concepts are used in developing the naming of different soils. First is the idea that a soil's characteristics will determine which group it falls into and its name. Another idea is that soils will be related to a reference soil. In both cases the natural soil horizons are used in the soil description. In Soil Taxonomy horizons used for naming the soil are called *diagnostic horizons*. In the reference soils the horizons are called *reference horizons*. The general concepts are similar in all systems, and so are many names for soil characteristics. The names are often descriptive in that they give an idea of the characteristic of the soil.

In the FAO soil system and Soil Taxonomy several soils have the same or similar names or names that can easily be seen as being related to each other. Examples would be Histosols and Vertisols, which carry the same name in both systems. Andosols and Andisols only differ by only one letter. Ferralsols and Oxisols should be understood as being similar if one knows a little about highly weathered soils. If one is working on an international scale or wishes to work globally, it is important to know that there are different ways of naming soils and that familiarity with the characteristics of various soil types is necessary. This is particularly important if one is developing a soil testing method or instrumental method, which might be either inadvertently or intentionally used on an international scale.

1.5. THE LANDSCAPE

In a landscape, the scientist differentiates one soil from another by the soils' physical, including horizonation, and chemical characteristics. The smallest unit that can be considered a soil, called a *pedon*, has an area of 1–10 m^2 and is normally 1.5–2 m deep. If an area contains contiguous pedons of similar characteristics, it is said to be a *polypedon* (multipedon) region.

In the USDA Soil Taxonomy and in field reports of soils the description given is that of a pedon—considered the "typical" pedon. In the first case the field description is that of a pedon, and when mapping soils the soil mapping unit strives to capture the distribution of similar polypedons. A soil mapping unit applied to the field may contain "inclusions" of dissimilar ploypedons. Both the Soil Taxonomy and the soil mapping unit nomenclature contain the terminology likely to be associated with soil samples that the soil chemist, environmental analyst, or instrument procedure developer will encounter. Knowing where the soil comes from, its Soil Taxonomy designation, and field description give the soil researcher and analyst a great deal of information about the chemical characteristics of the soil. In addition, reporting the soil name and its analysis together provide the reader with information that is invaluable in applying the results to that particular soil as well as other environmental conditions.

A landscape will contain many different soils, which can be expected to change with position in the landscape, vegetation, slope, climate, and parent material. It is also to be expected that the soil will change only over longer periods of time, namely, decades and centuries. However, groupings of soils can be and are made for all soils on this planet, and all soil types can be found on all continents except Antartica. Ultisols, Mollisols, Alfisols, Spodosols, and other soil types occur in North and South America, Asia, Europe, and Africa, Australia, and New Zealand. Thus researchers and analysts all over the world need to know about soils, soil types, and their nomenclature.

1.6. RELATIONSHIP OF LARGE FEATURES TO SOIL CHEMISTRY, ANALYSIS, AND INSTRUMENTATION

Each topic discussed above provides a great deal of information about the soil and its chemistry. This information is invaluable to the soil chemist, the person developing a soil analytical procedure, or a person wishing to make an instrumental analysis of some soil characteristic or component. The discussion below should not be taken as exhaustive, but only as providing examples of the types of information available and why knowledge of the large features of soil are important in understanding its chemistry and analysis. The following chapters will refer back to these characteristics where appropriate.

1.6.1. Soil Types

What do the names Alfisol and Mollisol tell us about the chemistry of these soils? Alfisols develop under humid climates and thus are acidic and have a medium base saturation. All the easily leached salts have been removed. However, basic parent materials often underlie them. Mollisols develop under lower rainfall conditions than do Alfisols and so have higher pH levels and higher concentrations of more easily leached salts, and—although they contain significant levels of calcium—they are still often slightly acidic. Mollisols have higher organic matter throughout the upper portions of the soil profile than do Alfisols. Both soils have well-developed B and Bt horizons.

Knowing which of these types of soils is present tells us much about its chemistry, the likely pH or pH range of the soil, and its salt content The occurrence of clay, especially in the Bt horizon, will affect the availability and solubility of both inorganic and organic components. For instance plant available phosphorus is decreased by low (acid; $pH < 7$) and high (basic or alkaline; $pH > 7$) soil pH. Likewise, the type of clay present will dramatically affect the extractability of a soil component or contaminant. Clay type and its chemistry will be discussed in Chapter 2.

Ultisols and Spodosols develop under different conditions but have some important similarities. Ultisols are the ultimate in soil development. Salts have been leached out; however, they are generally considered soils having

maximum clay formation. These soils are typically very acidic (here "very" is used relative to other soils), and there is the occurrence of aluminum as Al^{3+}. This is of note because aluminum in this form is toxic to plants. Although there is a Bt horizon, the clay is the simplest and least active of the clays (see Chapter 2). Spodosols develop in coarse-textured soil under trees, the dentris of which provides acid to water leaching through the soil profile. Its B horizon contains illuvial accumulation of highly decomposed organic matter and oxides of iron and aluminum. In this case the B horizon as shown in Figure 1.4 does not have an increase in clay.

The pH and clay content of these soils are extremely important in understanding their chemistry. The lack of salts, low pH, and dominance of low-activity clays will greatly affect the retention and extraction of components from both Ultisols and Spodosols.

In the Aridisol both the occurrence of clay in the lower horizon and the occurrence of high pH and salt contents will greatly affect the retention of components. For these soils, analytical procedures must be potentially impervious to high pH levels and salt content, or steps must be taken to remove salts or change the pH before analysis.

1.6.2. Soil Color

The color of soil gives an indication of its oxidation–reduction conditions and the amount of organic matter present. Well-aerated soils will be observed under oxidizing conditions, and iron will be in the Fe^{3+} state, less soluble and thus less available for chemical reaction. Under water-saturated conditions, soil will be under reducing conditions as indicated by increased yellow coloring, gleying, and mottling. Iron will be in the Fe^{2+} state, which is more soluble and thus more available for chemical reaction. Under these conditions reduced species such as methane, CH_4, hydrogen, and other sulfides will be found.

Under saturated or very wet conditions soils tend to have increased amounts of organic matter. This results in dark colors and dramatically changes the chemical characteristics of a soil. Organic matter increases a soil's sorptive and cation exchange capacities and thus alters the movement and extraction of components present. Organic matter increases ped formation and stability, thus increasing both aeration and percolation, but under saturated conditions reduction reactions prevail (see Figure 1.9).

While color is often discussed as being closely related to iron and its oxidation state, which in fact it is, it is also related to all other soil components. When soil color indicates oxidizing conditions, all multiple oxidative state capable cations are expected to be in their highly oxidized states, and when soil color indicates reducing conditions, low oxidative states will occur.

1.6.3. Soil Structure

Increasing soil structure results in increases in both small and large pores, which means improved water and oxygen movement in and out of soil and

thus increased oxidative conditions. The use of adjectives such as "strong" in describing soil structure such as in the soil profile descriptions in Figures 1.1 and 1.5 indicates ped formation that is strong enough to resist breakdown under wet conditions, thus leading to pores that drain well even when wet. Strong structure favors oxidation of soil constituents as long as the soil does not have a high water table.

1.7. CONCLUSIONS

Soils develop by the action of the soil forming factors on soil parent materials, including material transported by different agents. The result of these soil forming factors is the formation of soil horizons, different colors, and peds. Each of these factors has a pronounced effect on a soil's chemistry. Knowledge of the soil type and profile description can provide the soil chemist, analyst, or researcher with valuable information about the characteristics of soil relevant to the development of extraction, analytical, and instrumental analytical procedures. It also is the place to start when investigating the failure of a procedure.

PROBLEMS

1.1. Describe how horizons form in soil. How would knowing the horizon help a soil chemist understand differences in extractability of a soil component?

1.2. List three major horizon designations and three subordination designations and explain what information they reveal about the horizon.

1.3. What common soil characteristics are indicated by soil color? Explain how a soil's color is described.

1.4. Name two major different types of soil structure. How does soil structure relate to a soil's oxidative or reductive conditions?

1.5. Which major soil types have high salt and pH levels? Which have low levels?

1.6. Describe the effect of small pores on the chemistry of a soil and what types of compounds are likely to be found.

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CHAPTER

2

SOIL BASICS II

MICROSCOPIC TO ATOMIC ORBITAL DESCRIPTION OF SOIL CHEMICAL CHARACTERISTICS

In this chapter soil components will first be considered as individual independent, noninteracting entities. Then the interaction between the various components in soil will be discussed. However, it is essential to know and remember that components in soil never act independently of each other. In addition, surfaces always have a coating of some type that is not continuous, varies in thickness, and sometimes exposes the underlying surface. Sometimes this first coating will have another, different, coating on top of it.

Before an understanding of the interactions between soil components and surfaces is possible, it is essential to know the composition of uncoated soil components. Once this is known, it is then possible to discern the interactions and bonding patterns of these components with and without coatings.

The solid portion of soil is made up of sand, silt, clays, and organic matter. Elements and inorganic and organic molecules and ions are also present. The soil solution is a combination of elements and inorganic and organic ions and molecules. The gaseous portion contains gases commonly found in the atmosphere. However, the concentrations of these gases are very different in soil air than in the atmosphere. All components are subject to partitioning between these three phases.

The chemistry of soil is contained in the chemistry of these phases, the elements present, their bonding, and the atomic and molecular orbitals available for bonding and reaction. For the solid phase the chemistry will depend on the amount and type of surface available for reaction. In the liquid phase solubility will be the most important characteristic determining the chemistry occurring. In the gaseous phase gas solubility and the likelihood that the component can be in the gaseous form (i.e., vapor pressure) will control reactivity.

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

SOIL COMPONENTS INDEPENDENT

2.1. SOIL SOLIDS

Soil scientists define sand particles 2 mm in diameter as the largest "regular" component of soil. Many soils contain gravel, rocks, and other materials, that are larger than 2 mm in diameter but because of their relatively large size have limited surface area and provide little in the way of chemical reactivity in soil. For this reason these particles will not be considered. Particles smaller than 2 mm are divided into groups in numerous ways by different disciplines and researchers; however, for our purposes it will suffice to view them as belonging to three groups: sand, silt, and clay.

2.1.1. Sand

The sand fraction of soil is composed of particles 2–0.02 mm in diameter.[1] It is derived from the physical breakdown of rock. Wave action in oceans and lakes, tumbling in a river, or grinding in a glacier are common ways sand is produced. Heating and cooling, wetting and drying, and freezing and thawing are also common physical ways that sand-size particles are produced. Plant roots growing through cracks in rocks also break them down into smaller particles that may eventually become sand size. Many soil scientists find it convenient to subdivide sand in to fine, medium, and coarse or even more groups. The surface area of each group is different; however, the surfaces and the chemical reactions occurring on them are not.

In terms of chemistry, all sand is assumed to be silica, the empirical formula of which is SiO_2 or silicon dioxide and can be considered the anhydride[2] of silicic acid, H_4SiO_4. However, in the environment in general and soil specifically, silica is polymerized to form sheets and three-dimensional structures such as sand particles. Here each silicon atom is tetrahedrally surrounded by four oxygen atoms (see Figure 2.1) and each oxygen also has two lone pairs of electrons. All surfaces of silica structures will have electrons that attract any positive or partially positive species near the surface. Thus water, through its partially positive hydrogens, and any positive species it contains will be attracted to the surfaces of silica. Figure 2.1 also shows a water molecule illustrating the partially positive hydrogens and partially negative oxygens and the lone pair of electrons on the oxygens attached to silicon.

At broken edges it is possible to find oxygen atoms that are attached to silicon but have no silicon bonding partner. These are often associated with hydrogen forming hydroxy (—OH) groups. The bonds and orbitals of silicon,

[1] This is the international Soil Science Society definition. The USDA defines sand as ranging from 2 to 0.05 mm in diameter.

[2] An anhydride is a compound that has lost the elements of water.

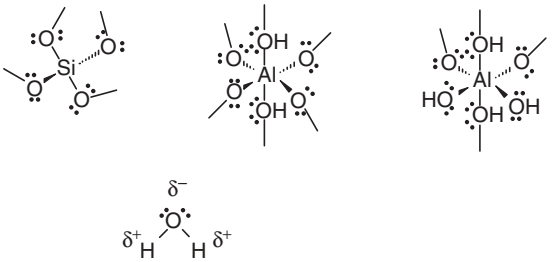

Figure 2.1. A silicon tetrahedron (left), an aluminum octahedron as a central layer in a 2:1 clay and an aluminum octrahedron as a surface layer in a 1:1 clay (right). Both the oxygen and OH groups are bonded to other silicon and aluminum atoms in the clay (bonds are not intended to be shown at the correct angels). Below is a water molecule showing partially positive hydrogens and partially negative oxygens. Also shown are the two lone pairs of electrons on all the oxygens.

oxygen and hydroxy groups bonded to silicon determines the chemical reactivity of freshly formed surfaces of this soil fraction.

Quantifying silica interactions with its surroundings is difficult. First, the surfaces are not regular, and thus it is impossible to calculate their area. Surface areas must be measured, and although surface area measurement is not difficult, it is time-consuming and open to inaccuracies. Second, as noted above, the surfaces are irregularly covered and it is impossible to know the extent, type, and thickness of materials covering all the surfaces. However, silica bonds and electron pairs are important in any chemical analysis, analytical procedure or instrumental procedure applied to soil.

2.1.2. Silt

The silt fraction is particles 0.02–0.002 mm in diameter. This fraction or separate is produced by the same physical processes as described above for the formation of sand. Silt is more finely divided silica, but the surfaces are basically the same as those of sand (i.e., silicon), and oxygen lone pairs of electrons and hydroxy groups control its chemistry. Because the particles are smaller, they have a larger surface area, that is, more surface per unit mass. This results in the availability of a greater number of bonds for chemical reactions. However, again, although the amount of surface area can be measured, the availability of silicon, oxygen lone pairs of electrons, and hydroxy groups for chemical reaction cannot be known exactly [1].

2.1.3. Clay

The next smaller separate is actually a group of particles of differing types collectively called *clay* and are particles measuring less than 0.002 mm in diameter. They are significantly different from sand and silt separates both physically

and chemically. Physically they are mostly colloidal in size and thus remain suspended in water and have large surface areas (unit mass basis). There are a large and varied number of clays that differ in terms of both the arrangement of their components and in the components they contain. Here we will consider four common types of clays found in soil that are models for all soil clays. These clays can be grouped into two classes: (1) clays composed of layers of alumina octahedral sheets and silica tetrahedral sheets and (2) the amorphous clays.

There are three potential sources of soil clays:

1. Clay that is present in rock and is released when rock breaks down as a result of physical processes. These clay structures are subject to change once released from rock.
2. Clays released by chemical breakdown of rock, which can lead not only to the release but also the formation of clays.
3. Clays formed in soil by chemical reactions occurring after rock decomposition.

Soil components silica and alumina are solubilized, in low concentration, and can react, or crystallize, to form new clays. In addition, clays from any source change over time and become simpler and simpler. Silica is more soluble than alumina and so the silica : alumina ratio decreases over time. Eventually this leads to deposits of alumina that are used to as an aluminum ore for the production of aluminum metal. Although these reactions are considered to be very slow on a human timescale, they do occur.

Clays composed of layers are called *layered silicates*. The most common sheets are of silicon tetrahedra and aluminum octahedral (see Figure 2.2). Three common representative clays in soil are the 1 : 1 kaolinite, 2 : 1 fine-grained micas, and 2 : 1 smectites; that is, kaolinites have one sheet of silicon tetrahedra and one sheet of aluminum octahedra. The fine-grained mica and smectites have two sheets of silicon tetrahedral and one sheet of aluminum octahedra. These latter clays have a sandwichlike arrangement with the aluminia octahedral sandwiched between two silica tetrahedral layers.

In terms of soil development and the development of soil horizons, the smectites and fine-grained micas are found in younger, less weathered soils. Kaolinite is found in highly weathered soils. Considering a time sequence, at the beginning of formation soil will contain more complex clays that weather to simpler forms over time. However, it is convenient to start with a description of the simpler layer silicate clays and then describe the more complex clays.

An additional important characteristic of clays is their surfaces, which are distinguished as being either external or internal. Internal surfaces occur, for example, in nonexpanding 2 : 1 clays, such as the fine grained micas, and are generally not available for adsorption, chemical, or exchange reactions.

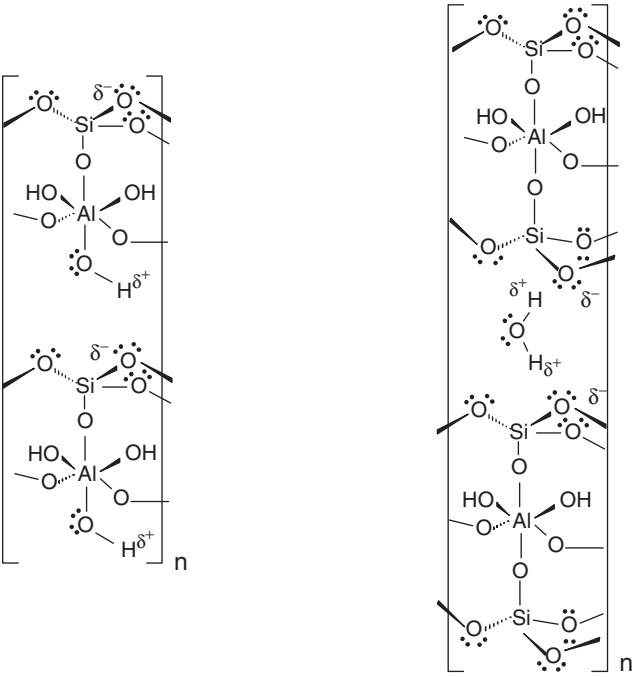

Figure 2.2. The left structure represents kaolinite, a 1:1; and the right, a 2:1 clay mineral. These representations are intended to show surface groups, surface pairs of electrons, unsatisfied bonds, and associations between clay particles. Note that clay structures are three-dimensional and these representations are not intended to accurately represent either the three-dimensional nature or the actual bond lengths; also, the brackets are not intended to represent crystal unit cells.

2.1.3.1 1:1 Clay—Kaolinite

The 1:1 kaolinite clay is depicted in Figure 2.2. One surface is composed of oxygens with lone pairs of electrons. These electrons are in *p* orbitals and thus extend away from the oxygens into space. The oxygens are partially negative as indicated by the symbol δ^- because they are more electronegative than the surrounding atoms, namely, Si. The other surface is composed of —OH groups where the hydrogens are partially positive as indicated by the symbol δ^+. The electrons in the sigma bond between the oxygen and hydrogen are drawn closer to the oxygen because it is more electronegative, thus leaving the hydrogen slightly positive.

The consequence of these partial charges it that one surface of kaolinite is compatible and attractive to the other surface. This results in increased stability of kaolinite and the formation of relatively stable structures. Some kaolinite particles can even be larger than the 0.002 mm upper limit for clay! Both surfaces also attract and hold water through these partial charges. The adsorptive activity of kaolinite is associated with its surface electrons and partially

positive surface hydrogens, and thus the two-face kaolinite can attract anions, cations, water, and electrophilic and nucleophilic compounds.

When a particle of kaolinite is broken or at the edges of the particles, bonds will be unsatisfied. This leaves negative charges, which attract cations. Thus kaolinite has cation exchange capacity. Typically these charges are satisfied by metal cations, particularly Ca^{2+}, Mg^{2+}, K^{+}, and Na^{+}. Other cations can be attracted to these sites including positively charged organic molecules such as quaternary amines and ammonium. Because the crystals are relatively large and the charges develop only at the edges, the cation exchange of kaolinite is small compared to that of other clays.

2.1.3.2 2:1 Clays—Fine-Grained Micas and Smectites

The 2:1 clays are smaller and much more complex than 1:1 clays. Because they are smaller, they have a larger surface area and more edges. This results in both increased cation exchange and adsorption. However, the adsorption will be different from that occurring in kaolinite because the surfaces of the particles are the same. In this clay cation exchange is not limited by edge effects because of a phenomenon called *isomorphous substitution*, which results in increased cation exchange, changes in the shape of the particles, and changes in the way they interact with water and cations.

A first approximation to the makeup of 2:1 clays can be seen in Figure 2.3. The particles consist of a sandwich of aluminum octahedral between two sheets of silicon tetrahedral. One result of this arrangement is that both surfaces are made up of only oxygen. This means that both surfaces are slightly negative and thus two particles repel each other if there is no positive or partially positive species, such as the hydrogens on water molecules, between them. In Figure 2.3 water is shown between the clay particles, which allow them to come closer together, and in reality water molecules are always present between the clay layers. As with all soil solutions, this water contains cations, further enabling the particles to come together.

Another characteristic of 2:1 clays is isomorphous substitution, where *iso* means same and *morphous* means shape. During the formation of clay, cations other than aluminum and silicon become incorporated into the structure. In order for this to work and still ensure stable clay, the cation must be of about the same size as either aluminum or silicon; hence the term *isomorphous*. There are a limited number of cations that satisfy this requirement. For silicon, aluminum as Al^{3+} and iron as Fe^{3+} will fit without causing too much distortion of the clay structure. For aluminum, iron as Fe^{3+}, magnesium as Mg^{2+}, zinc as Zn^{2+}, and iron as Fe^{2+} will fit without causing too much structural distortion (see Figure 2.3).

The two 2:1 clay types are distinguished by the type of substitution in the layers. Nonexpanding fine grained mica-type clay has isomorphous substitution of Al^{3+} in the silicon tetrahedral sheet. Thus, in a tetrahedral sheet of fine mica, aluminum may be substituted for a silicon atom. The expanding clays

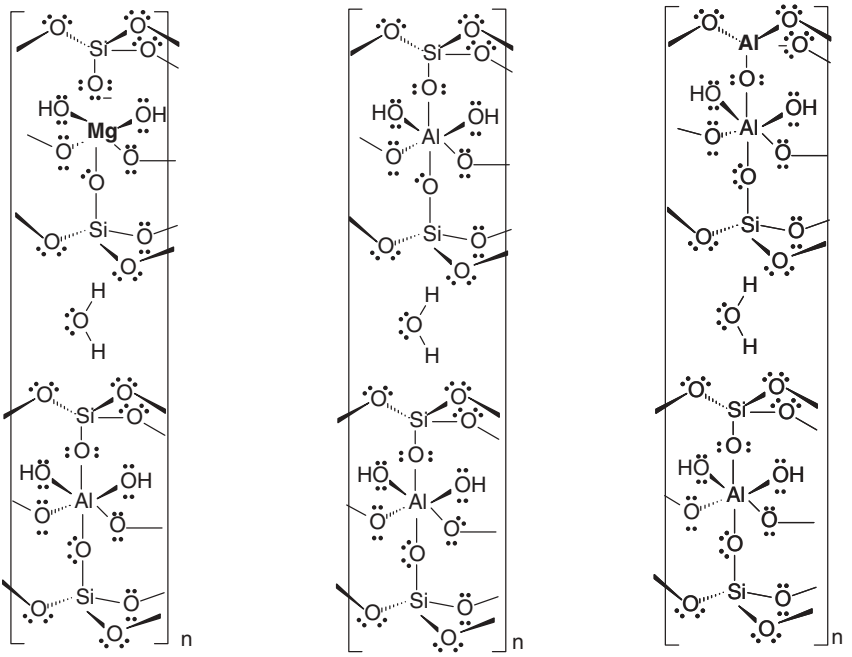

Figure 2.3. Two types of isomorphous substitution. The middle structure is a two-dimensional representations of clay without isomorphous substitution. On the left is isomorphous substitution of Mg for Al in the aluminum octahedral sheet. On the right is isomorphous Al substitution for Si in the silicon tetrahedral sheet. Clays are three-dimensional, and —OH on the surface may be protonated or deprotonated depending on the pH of the surrounding soil solution. There will be additional water molecules and ions between many clay structures. Note that clay structures are three-dimensional and these representations are not intended to accurately represent the three-dimensional nature or the actual bond lengths; also, the brackets are not intended to represent crystal unit cells.

have substitution in the aluminum octahedral sheet. For instance, an octahedral sheet might have substitution of magnesium for aluminum. These two substitutions were chosen to illustrate that with substitution some bonds in the clay structure will go unsatisfied. This means that some bonding electrons will not be shared between two atoms, resulting in the clay having a negative charge that is satisfied by the same cations discussed for kaolinite (Section 2.1.3.1). This results in cation exchange capacity greater than that ascribed to edge effects alone.

The Fine Grained Micas. The fine grained micas are distinguished by having 2:1 structure and are nonexpanding when the water content of their surroundings changes. Isomorphous substitution is in the silica tetrahedral micas, and causes a change in the shape of the crystal. Thus this portion of

the surface has a defect in it, which is the correct size to allow either or both ammonium as NH_4^+ or potassium as K^+ to fit between the layers.[3] Also, the unsatisfied bonds resulting from isomorphous substitution are close to the surface and thus the charge is closer to that of the cations found on the surface, resulting in a relatively strong attraction for both cations and water. These two phenomena result in trapping of both water and cations between crystals.

Water and cations trapped between layers are held so strongly that they do not exchange with water and cations in the surrounding environment and thus are not biologically available. For instance, ammonia trapped between the clay layers is not a source of nitrogen for plants and will not be oxidized by bacteria to nitrite and nitrate and thus will not be a source of nitrate or nitrite pollution.

Soil containing large amounts of fine grained mica clay can result in surprising analytical results. An analytical procedure that breaks apart the fine mica, such as strong-acid digestion, will show that the soil contains a large amount of ammonia or potassium, which can cause concern if it is not known whether the NH_4^+ and K^+ are unavailable and stable under normal environmental conditions.

The Smectite Clays. The smectite-type clays are distinctive in that they expand and cause significant destruction to synthetic (human-made) structures. In this type of 2:1 clay iomorphous substitution occurs in the aluminum sheet. If there is substitution of a lower-oxidation-state metal such as magnesium, there will be an unsatisfied pair of bonding electrons in the interior of the crystal and there will be no noticeable change in the surface. Because the charge is in the interior of the crystal, its attraction for cations is diminished by distance. Thus smectite crystals are not held strongly together by cations and are able to incorporate more water and ions between layers when the environment is wet and less when it is dry.

The amount of swelling depends on the cations and the number of waters (i.e., water molecules) of hydration that they have. Cations with higher numbers of hydration waters cause a higher degree of swelling. Lithium thus causes more swelling than does sodium, and so on. The greater the swelling, the easier it is for water and other ions to move in and out between the clay layers, making them both environmentally and biologically available.

The amount of swelling and contraction in smectites is quite dramatic. Typically soils containing large amounts of this type of clay will develop cracks 30 cm wide at the surface and >100 cm deep, and these cracks will allow surface material to fall into them during dry periods. This characteristic is so unique that these types of soils are given their own name. They are called *Vertisols*,

[3] Although it seems strange, NH_4^+ and K^+ ions are approximately the same size.

the name taken from the concept that material from the surface falls to the bottom of the cracks, resulting in inversion of the soil.

In the field when soil containing 2:1 clays become wet, they swell shut and water movement through them is extremely slow. In a soil profile wetting and swelling of this type of clay will prevent downward movement of water and associated contaminants. For this reason swelling clays are used to seal both landfills and ponds to prevent leaching or leaking.

The crystals of 2:1 swelling clays are typically smaller than either kaolinite or fine grained mica and thus have higher adsorption and cation exchange capacities. However, surface adsorption will be of the same chemistry as that in the fine micas.

2.1.3.3 Amorphous Clays

In addition to the crystalline clays described above, there are some materials that act like clays but do not have a crystalline structure. Amorphous clays do not have a definite X-ray diffraction pattern and are differentiated from the crystalline clays on this basis. They are composed of mixtures of aluminum, silicon, and other oxides and generally have high sorptive and cation exchange capacities. Few soils contain large amounts of amorphous clays [2].

2.1.4. Soil Texture

Sand, silt, and clay are the three components of soil texture. Various relative compositions, expressed on a percentage bases, are used to give soils a textural name such as sandy loam, or loamy clay. For soils containing significant amounts of silt, the term *loam* is used, although with high levels (>88% silt, <20% sand, and 12% clay), the soil would be called *silt*. Thus, a soil containing high amounts of sand would be sand, sandy loam, loamy sand, or sandy clay, and clay soils would be designated as clay, clay loam sandy clay, and silty soils as silt loam, silty clay loam, and so on. The textural name of a soil is established by determining the soil's relative percentage of sand, silt, and clay and then finding this percentage on a textural triangle, included in all standard texts on soils (see Reference 1 or 14), to find the name. The textural name of a soil often accompanies the name, (e.g., Milton silt loam) or can be obtained from a soil scientist familiar with an area's soils.

Sandy soils are easiest to extract and analyze, while clay soils are the hardest. Drying, crushing, and sieving will aid in extraction and analysis, although it may not be necessary to crush very sandy soils. Clayey soils may retain small amounts of contamination even after extensive extraction. In all cases extraction and analysis procedures must be robust enough to handle all textures containing all clay types [3].

SOIL COMPONENTS INTERACTING

2.2. BONDING CONSIDERATIONS

As noted in Chapter 1, sand, silt, clay, and organic matter do not act independently of each other in soil. Thus one or several types of chemical bonds or interactions—ionic, polar covalent, covalent, hydrogen, polar–polar interactions, and Van der Waals attractions—will be important in holding soil components together. The whole area of chemical bonding is extremely complex, and thus, in addition to specific bonding considerations there are also more general ways of investigating the interaction between components in soil. These involve, for instance, graphing the adsorption of organic compounds to soil constituents at increasing levels of organic compound. The shape of the graph is used as an indication of the type(s) of interaction(s) between constituents at various concentrations.

2.2.1. Orbital Overlap

Interaction between silicon or aluminum surfaces or edges and surrounding components will entail overlap of surface orbitals and available orbitals in the approaching species forming covalent or polar covalent bonds. The strength of the interaction will depend on the strength of overlap of the available orbitals. Bonding energies of orbitals are well known, and so the strength of the bonds can be either directly known or estimated. Orbital overlap is not the only factor affecting the interaction or strength of bonding. In addition to energy, reaction path, steric, and rate factors will play a role in any attraction.

Orbitals can also be considered from a molecular orbital standpoint. Each reacting species must have molecular orbitals available, and be of the correct symmetry such as to allow for bonding. These will be called the "frontier orbitals," composed of the highest occupied (HOMO) and the lowest unoccupied (LUMO) molecular orbitals. In addition to their involvement in bonding between species, these orbitals are of considerable interest in that they are largely responsible for many of the chemical and spectroscopic characteristics of molecules and species and are thus important in analytical procedures and spectroscopic methods of analysis [4–6].

2.2.2. Ionic Bonding

Ionic bonding is generally regarded as the predominant type of bonding between the ions that make up salts or the compounds formed between metals and nonmetals. The basic concept is always illustrated as a compound such as sodium chloride and explained by saying that sodium donates its outer most electron to chlorine such that both have a noble gas electron configuration.

The two oppositely charged species then attract each other, forming a compound. Although the seminal characteristic of compounds held together by ionic bonds is that they dissolve in water, giving a solution containing ions, it is essential to keep in mind that many ionic compounds are insoluble. The solubility of these compounds depends on the relative strength of solvation and bonding energy.

Some ionic compounds contain a combination of bonds. For instance, in polyatomic ions such as ammonium (NH_4^+), the hydrogens are bonded to the nitrogen by polar covalent bonds. The ionic bond is thus between this covalently bonded moiety and another ion.

Ionic bonds are typical of inorganic compounds, and thus the mineral or inorganic components of soil often contain ionic bonds and are soluble in water. This means two things: (1) the soil solution should always be expected to contain salts and their corresponding ions and (2) the inorganic components of soil should be expected to dissolve in the soil solution, some at a very slow rate, resulting in their ions being present in low concentrations.

The third aspect of this is that analysis of inorganic or ionic compounds must take into account not only their solubility in the soil solution but also the possibility that they may be present as exchangeable ions. Extraction procedures designed for inorganic components thus need to take these two characteristics into account [7].

2.2.3. Ion Exchange Interactions

The soil colloids, both inorganic (i.e., clay) and organic (i.e., humus), contain charges that are balanced by cations and anions associated with the charged sites. Most soil clays and humus contain a predominance of negative charges and thus act as cation exchangers. Some clays also have significant numbers of positive charges and will act as anion exchangers. Thus soil can have both *cation exchange capacity* (CEC) and *anion exchange capacity* (AEC).

When considering cation or anion exchange capacity the pH of the soil solution is extremely important. There will be competition for binding sites between H_3O^+ and other cations in the soil solution. In addition, some surface atoms may become protonated, thus decreasing the available negative sites on these surfaces. Therefore, the observed CEC will be less at high proton concentrations, that is, at low or acid pH levels, and higher at basic pH levels. Thus the CEC of the soil at the pH being used for extraction is the important value, not a CEC determined at a higher or lower pH.

Exchangeable cations must be removed from the exchange sites to be detected and quantified. To accomplish this, the soil sample is extracted with a solution containing a cation having multiple charges or present at high concentration. At the same concentration, cations having more positive charges will replace those on exchange sites having smaller charge. This condition can be overcome if the less charged cation is in high concentration when even a cation with one positive charge can replace a cation with multiple charges.

Soils with anion exchange capacity can be expected to exchange anions in the same way. However, in many soil anions are present as oxyanions, which often react with soil components and thus do not and cannot act as exchangeable anions. Phosphate anions are excellent examples of this type of interaction [8].

2.2.4. Hydrogen Bonding

Hydrogen bonding is typified by the attraction of a partially positive hydrogen, attached to a partially negative oxygen, which is attracted to a partially negative oxygen on another molecule. A common example is the hydrogen bonding in water, where the hydrogen of one water molecule is attracted to the oxygen in another water molecule. Whenever hydrogen is bonded to a significantly more electronegative element, it will have a partial positive charge. It can then be attracted to lone pairs of electrons on other elements in other molecules and thus produce an interaction that, although not classically a hydrogen bond, is very similar to hydrogen bonding. Nitrogen and phosphorus atoms would all fall into the category of electronegative atoms having lone pairs of electrons.

Although hydrogen bonding is considered to be a much weaker interaction than covalent or ionic bonding, it is nevertheless a relatively strong interaction. When molecules have multiple sites for hydrogen bonding, there can be significant strength in the association; for instance, paper is held together by hydrogen bonding [9,10].

2.2.5. Polar–Polar Interactions

Whereas hydrogen bonding can be considered as a type of polar–polar interactions, I define polar-polar interactions as the intermolecular attraction of polar groups, which does not involve hydrogen. An example would the attraction between two propanone (acetone) molecules, where a partially positive carbon of the carbonyl group in one molecule is attracted to the partially negative carbonyl oxygen of another molecule. This will be a weaker interaction than hydrogen bonding but a stronger interaction than Van der Waals.

2.2.6. Van der Waals Interactions

Van der Waals attractions are described as the development of instantaneous polar regions in one molecule that induce the development of polar regions in another molecule and result in an attraction between the molecules. These are considered to be instantaneous or short-lived polar regions that are in a constant state of flux. The most common example is the attraction between hydrocarbon molecules where this is the only discernable attracting force between molecules.

Because different components are held to soil components by different types of bonding and attractions, the interaction can be relatively strong or weak. Thus extraction procedures must be capable of extracting the desired component when it is held by different forms of bonding to different components.

2.2.7. Other Ways of Investigating Bonding

Bonding and other interactions between the components in soil (i.e., the clays) and organic components can be investigated by conducting adsorption experiments. An organic molecule is added to a suspension of clay and the amount adsorbed after a fixed amount of time is determined. The amount adsorbed is plotted against the amount added to produce adsorption isotherms. The shape of the graph is then used to indicate the type of interaction between the molecule and the clay. With this type of investigation, various types of adsorption phenomona can be distinguished.

Two of the most common ways of handling such data is to try to fit the data to either a Langmuir or a Freundlich type of equation, or alternatively to simply determine which of these two equations best describes the data obtained. Although some useful information can be obtained about the interactions between the components being studied, neither provides specific information about the type of bonding in terms of orbitals, or interactions such as those discussed in the previous sections. Spectroscopy, as discussed in Chapter 7, is typically the method used to determine bonding details [11,12].

SOIL COMPONENTS IN COMBINATION

2.3. SURFACE FEATURES

Both sand and silt surfaces are dominated by oxygen and its lone pairs of electrons in *p* orbitals. In some instances broken surfaces may also have silicon-hybridized sp^3 orbitals[4] available for bonding. Comparison of sand, silt, and clay reveals the surface area of sand and silt to be low and the interaction between surface bonding orbitals and components in the surrounding medium relatively weak.

As a first approximation, the surfaces of the clays can be grouped into three types: (1) surfaces consisting exclusively of oxygens with their lone pairs or electrons, in *p* orbitals, extending at an angle away from the surface into the surrounding medium; (2) surfaces containing —OH groups with the partially positive hydrogens extending into the surrounding medium—because of the

[4] This would be a hybridized orbital formed by the hybridization of one *s* and three *p* orbitals as opposed to 2*s*,2*p* hybridization in carbon.

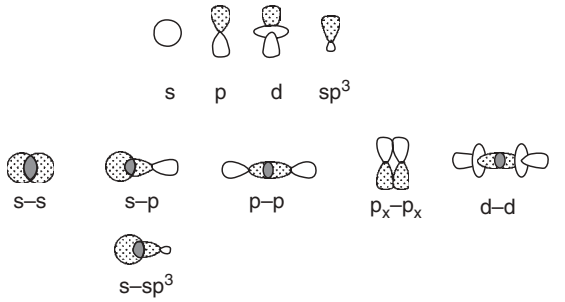

Figure 2.4. Common representations of the *s*, *p*, *d*, and atomic orbitals; sp^3-hybridized orbitals and some representations of how they overlap to form bonds between atoms are also shown.

bonding angle between oxygen and hydrogen, the partially negative oxygen will also be exposed to the medium; and (3) surfaces with broken edges, which can present a number of different orbitals depending on where the break occurs.

It can be imagined that the bonds at edges can be broken at any given location, that is, with an oxygen, hydroxy, silicon, or aluminum exposed. In this case it could further be imagined that *s*-, *p*-, and sp^3-hybridized orbitals would be on the surface. This will lead to complex bonding and reactivity, resulting in bonds of varying strengths and interactions and of varying types. The issue is then how these surfaces will interact with components commonly found in the soil solution.

Molecular orbital depictions of the orbitals described above are given in Figure 2.4. To the newcomer, these types of diagrams can be confusing. The orbitals and their shapes are calculated. The calculations result in the orbitals having charge signs, plus (+) and minus (–), and will often be represented in this fashion. It is common to think of negative and positive signs as representing negative and positive charges with negative and positives attracting each other and two negatives or two positives repelling each other. However, the signs (+, –) in this case do not represent charges. In molecular orbital diagrams the overlap of two atomic orbitals having the same sign denotes a positive interaction leading to bonding, that is, holding the two atoms together. This can also be indicated by two orbitals having the same shading as shown in Figure 2.4.

When the two orbitals have different signs, they do not overlap (nor do they cancel each other out) but result in the formation of antibonding orbitals. In this case the electrons are not shared between two atoms and do not hold the atoms together. For each bonding molecular orbital there is an antibonding orbital. Antibonding orbitals have higher energy than do bonding orbitals.

The overlap of *p* and *d* orbitals seen in Figure 2.4 can be of two types. They overlap can be *end-on-end* as depicted in the *p–p* and *d–d* representation, or

side-by-side, as depicted in the p_x–p_x representation. This is possible because the *p* and *d* orbitals are directed in space along the *x*, *y*, and *z* axes.

In some atoms the *p* and *s* orbitals are mixed together to form several equivalent orbitals. The most common example is carbon, where four orbitals are formed by mixing one *s* orbital with three *p* orbitals to give four equivalent orbitals designated as sp^3 orbitals.

Silicon as SiO_2, or its polymeric form $[SiO_4]_n$, has only two types of orbitals available for bonding. In the following scheme, sp^3 bonding occurs in SiO_4 and $[SiO_4]_n$; however they are tetrahedral, not planar as shown:

(2.1)

In SiO_2 the Si and O atoms have sp^2-hybridized orbitals[5] forming sigma bonds and *p* orbitals forming π bonds. The oxygen also has *p* orbitals containing lone pairs of electrons. Polymeric silicon has sp^3-hybridized obitals available for bonding, while oxygen still has lone pairs of electrons in *p* orbitals. These then are the orbitals available for bonding to elements or compounds that come into contact with the particle's surface. This type of interaction could be of either end-on-end or π type (i.e., side-by-side). In all cases steric hindrances may limit the type of interaction occurring.

Clays contain aluminum oxides in addition to silica as SiO_2 and its polymeric forms. Again there are the *p* orbitals of oxygen and *sp*-hybridized orbitals from aluminum, which may result in end-on-end or side-by-side bonding with the same restrictions encountered with silicon.

For removal of compounds bonded to silicon, aluminum or oxygen bonded to silicon or aluminum *p*- and *sp*-hybridized orbitals must be broken and new bonds formed. This will require energy and the correct orientation of attacking groups along with an effective attacking species. In addition, the rate of species removal will depend on the reaction path and the steric factors involved. All of these put together will determine the overall rate of the reaction and the time needed for extraction.

2.4. ENERGY CONSIDERATIONS

The first question in terms of the stability or strength of any bonding interaction is energy. A general equation for energy is $\Delta H = H_i - H_e$. This equation

[5] sp^2-hybridized orbitals are formed by mixing one *s* orbital with two *p* orbitals to produce three sp^2 orbitals.

simply states that the change in enthalpy is the difference between the energy at the beginning H_i and at the end of the reaction H_e. If a reaction is endothermic it will be at a higher energy level at the end that at the beginning. The reverse is true for an exothermic reaction. It is expected that the exothermic reaction leads to the more stable product and thus is more likely to be formed.

In addition to enthalpy, there is also the consideration of entropy or randomness. Reactions generally tend to go to a more random state. It would seem that any reaction involving the formation of a highly ordered crystal would be going to a state of decreased randomness. Thus there would be an increase in entropy, which would not favor the reaction. However, if the overall entropy of the system increases, then the reaction is favored.

The total energy of a system involves both enthalpy and entropy. Thus, whichever causes the greater change during a reaction will be the one controlling the reaction and determining whether it is exothermic, endothermic, spontaneous, or not spontaneous [13].

2.5. REACTION PATHS

Reaction paths involve following the energy path during a reaction. Typically reactants have a starting energy (R–R, Figure 2.5) which increases through either some transition state or reaction intermediate or both, leading to an ending energy. In Figure 2.5 the transition states (indicated by TS) are hypothetical species or structures that are in transition and cannot be isolated and identified. As indicated by the slight energy trough, the reactive intermediates are stable enough to be isolated and studied. Often reactive intermediates are ions such as carbanions.

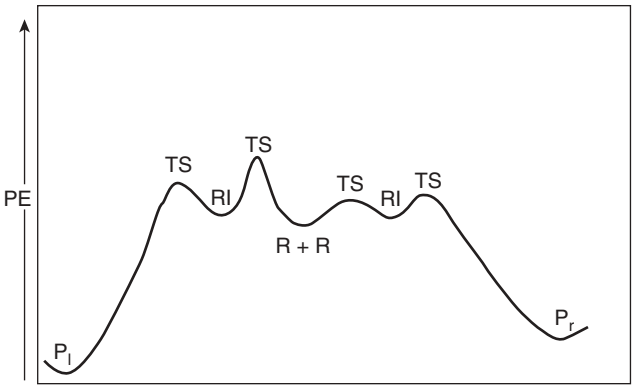

Figure 2.5. A potential energy (PE) diagram for a reaction that can occur in two different ways producing two different products (P): one kinetically and the other thermodynamically controlled (R—reactant; TS—transition state; RI—reactive intermediate; P—product).

In Figure 2.5 the path to the right involves lower energy transition states and reactive intermediates, and so the reaction will initially follow this path, although the product (P_r) is at an energy higher than the one obtained by following the left path (P_l). The formation of this product (P_r) will be controlled by kinetics, will be produced first, and will initially be at the highest concentration. After some time, reaction will take place and the product on the right (P_r) will rereact to form the more stable, thermodynamic, product on the left (P_l). This means that on standing, the product on the right will decrease to a low concentration while the one on the left will dominate the product mix.

In soil it is very common to find that a chemical added to soil becomes increasingly difficult to remove with time. The mechanism described above is one that would account for this observation. A compound added to soil crystallizes to a less stable crystal structure that is easily dissolved and extracted from soil. With time its crystalline structure rearranges to a less soluble, lower energy, and thus less easily extractable form [14].

2.6. STERIC FACTORS

Large bulky groups will slow or prevent nearby reactive sites from being attacked. They may also cause a reactive species to assume a shape that is not accessible to attack. The following two common bulky groups, which have a high degree of steric hindrance, are used extensively in organic synthesis:

t-butyl 2,2-dimethylpropyl group (2.2)

The position of such groups can affect the reactivity, access to reactive sites, and the conformation of the species to which they are attached. Thus, in considering the extractability of a component, its form, reactivity, and the occurrence of steric hindrances must all be considered [15].

2.7. RATE FACTORS

The rate of a reaction can also be an important factor in both the amount and longevity of a component in soil. A very common and interesting example is the microbial oxidation of ammonia first to nitrite and then to nitrate:

$$NH_4 \xrightarrow{\textit{Nitrosomonas}\ \text{spp.}} NO_2^- \xrightarrow{\textit{Nitrobacter}\ \text{spp.}} NO_3^- \qquad (2.3)$$

$$NH_4^+ + 1.5O_2 \xrightarrow{\textit{Nitrosomonas}\ \text{spp.}} NO_2^- + H_2O + 2H^+ \qquad 275\ \text{kJ} \qquad (a)$$

$$NO_2^- + 0.5O_2 \xrightarrow{\textit{Nitrobacter}\ \text{spp.}} NO_3^- + \qquad 76\ \text{kJ} \qquad (b)$$

$$\text{Difference} \qquad 3.62\times$$

When observing the oxidation of ammonia to nitrite in soil, it is found to be a slower reaction than is the oxidation of nitrite to nitrate. When the energy available from each of these reactions is considered [see (2.3)], it is obvious that this observation is directly related to the amount of energy available.

In this case nitrite is not expected to occur or build up to appreciable levels in the environment. *Nitrobacter* species must use approximately 3.6 times as much nitrogen in terms of nitrogen atoms to obtain the same amount of energy as *Nitrosomonas* spp. Thus it can be expected to take up nitrite at a higher rate to compete. This type of energy calculation is simply done by factoring in the amount or energy required for bond breaking and the amount released in bond making. Alternatively, the energy can be measured directly by calorimetry.

2.8. ALL FACTORS TOGETHER

In a purely chemical approach to how reactions take place, all of these factors come together; specifically, the rate is related to total energy used and released, the energy of activation required, steric effects, and the types of bonds being broken and formed. For this reason it is most common to measure the energy and rate quantities directly for the conditions of the reaction. Because of the complexity of soil, it is even more important to measure these directly.

2.9. MICELLES

In Chapter 1 it is observed that sand, silt and clay do not act independently of each other. In a similar fashion clay particles alone do not act independently of each other; rather, they form groups of particles called *micelles*. The model for a micelle is a group of long-chain fatty acid salts in water. The hydrophobic ends are associated with the ionic "salt" end exposed to water. An idealized micelle with some associated water is shown in Figure 2.6. The structure is often described as being a ball-like. This ideal is hard to visualize when looking at the typical shape and size of a clay particle. However, it is possible to envision individual clay crystals associated with each other through hydro-

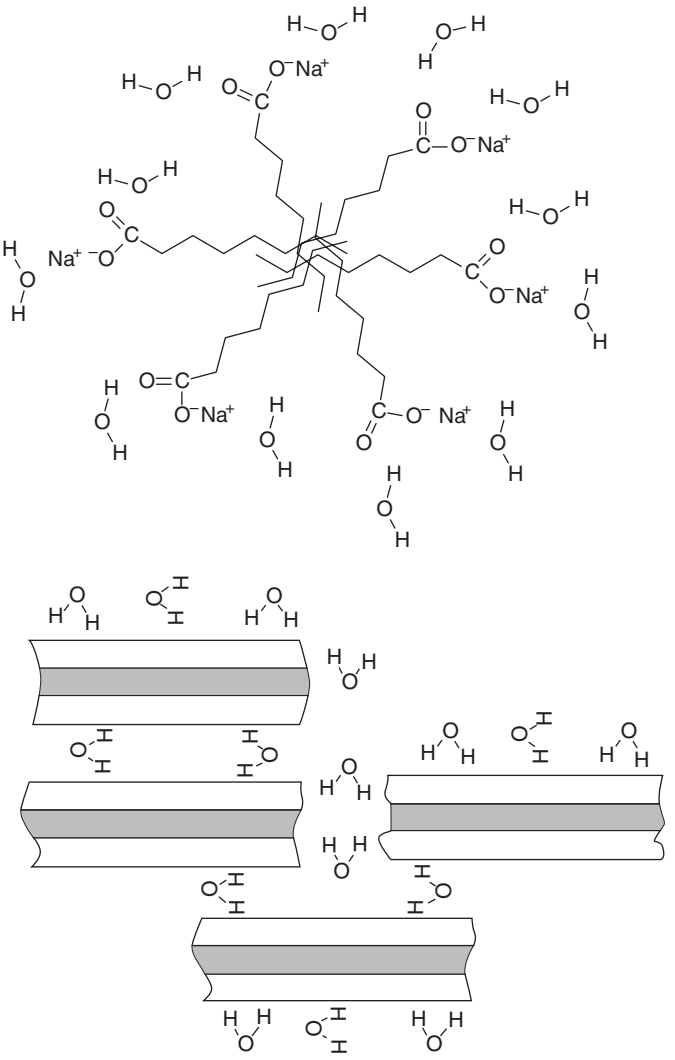

Figure 2.6. An upper micelle of sodium octanoate in water lower micelle of 2:1 clay (center gray layer is an aluminum octrahedrial sheet; the upper and lower ones are silicon tetrahedrial sheets).

gen bonding and ion–charge interactions to form small conglomerations of clay particles that will act like micelles also as shown in Figure 2.6 [16].

2.10. COATED SURFACES

The discussion to this point assumes that all the surfaces of all the components in soil are clean and available for reaction. This never happens. All surfaces are covered by coatings of various types; the number, types, and amounts of

possible coatings are quite varied. However, three of the most important coatings with which one should be familiar to understand the chemistry of soil are water, iron oxides, and organic compounds. All soil surfaces will be "contaminated" with a combination of these three compounds plus others, such as manganese oxides and elemental carbon, if they are present. Thus the orbitals, bonding, energy, and other characteristics of these surface coatings will also come into play when considering their reactivity and extraction of components from them [17–19].

2.11. CONCLUSIONS

Soil inorganic solids are composed of particles of decreasing size from sand to clay. The clay fraction is further divided into principally 1:1 and 2:1 clays. The 1:1 clays are typified by kaolinite, which, compared to other clays, exhibits lower activity. The 2:1 clays are typified by fine grained micas that are not expanding and smectites that are expanding. Clays have high sorptive capacity and are one source of cation exchange in soils. Bonding of soil components to each other and to surfaces involves all the standard types of bonding, namely, ionic, polar covalent, covalent, hydrogen, polar–polar, and Van der Waals. Reactions thus involve *s*, *p*, *d*, and *sp* orbital overlap and ionic and partial charges such as those involved in hydrogen bonding. These bonding considerations also involve the common surface features such as surface oxygens and hydroxy groups along with exposed aluminum and silicon. Extraction of components from soil therefore involves all types of bonding along with energy, reaction path, steric, and rate factors. Also involved will be micelle formation along with coatings on all soil surfaces.

PROBLEMS

2.1. Identify the three primary particles in soil and describe the chemical differences between them.

2.2. Identify the three major types of clays in soils and explain how they differ chemically.

2.3. Describe the different types of bonding and their primary occurrences in soil.

2.4. Describe the surface features, with particular reference to orbital availability, involved in surface binding of components in soil.

2.5. In terms of bonding energy, the bonds formed in an exothermic reactions must be lower than those in the reactants. How is this known?

2.6. Explain how both reaction mechanisms and energy considerations contribute to the abundance of the species of compounds found in soils.

2.7. Explain how the rate of a particular reaction may affect the amount of a particular component in soil.

2.8. Diagram the expected structure of an organic and an inorganic (clay) micelles in soil.

2.9. In general terms, describe the condition of the surfaces of all soil components.

2.10. Describe the expected differences between sand and silt surfaces and clay surfaces.

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CHAPTER

3

SOIL BASICS III

THE BIOLOGICAL AND ORGANIC COMPONENTS IN SOIL

It might be assumed that soil is basically a dead, inert, nonliving material, which it is not. A more accurate view is that soil is living or alive. It has living organisms, which appear to be mostly plants, in and in intimate contact with it. Plants have two parts, which interact with soil the tops and the roots. Both are important, although roots have a more intimate and immediate effect on the biological, physical, and chemical properties of soil. Many other types of organisms (i.e., microorganisms and animals), both large and small, live in soil. Microorganisms come in a wide variety of sizes, shapes, and abilities in terms of the types of environments they live in and the reactions they carry out. It is common knowledge that worms and prairie dogs live in soil, but these serve only as representatives of the variety of animals present.

Animals and plants, especially roots and microorganisms, provide biochemical, bioorganic, and organic compounds in soil. These may be in the form of cellular components, such as cell walls and membranes, enzymes, and complex and simple organic compounds. Decomposition of complex cellular material and biochemicals leads to the formation of simpler intermediate bioorganic and organic compounds. Aerobic decomposition of any organic matter in soil eventually leads to the production of carbon dioxide and water, anaerobic decomposition leads to the same products plus methane, and both result in the synthesis of humus, which is a dark-colored, high-molecular-weight, and highly complex material. The overall reaction of organic matter in soil is illustrated in Figure 3.1.

Organic compounds interact with each other and with the inorganic, macro, micro, and colloidal components in soil. Complex and simple organic molecules form complexes with inorganic cations and anions and with colloidal carbon and clay in the soil solution. Some enzyme–soil complexes may have increased catalytic activity, while others have decreased activity or are completely inactive. Also, some combinations and complexes may render organic matter resistant to decomposition. Eventually organic matter and complexes may become stabilized such that they have long residence times in soil.

All of these factors are active in all soil at all times and may affect any analysis. Simpler organic and inorganic molecules, compounds, and ions may

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

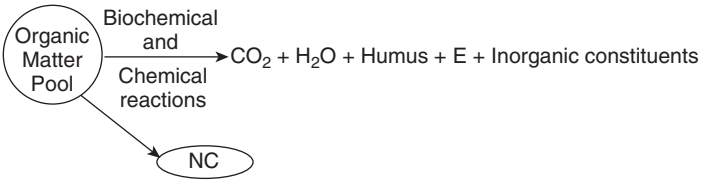


Figure 3.1. Organic matter (OM) breakdown in soil under aerobic conditions. These reactions lead to the formation of humus and are carried out to release energy (*E*), which is used by microorganism. Heterotrophic microorganism use OM to construct new cells (NCs). They also lead to a greater randomness in the system.

be associated with more complex organic materials, with humus, or with inorganic carbon and clay as part of the soil matrix. These interactions can mask the occurrence and concentration of both organic and inorganic components, obscuring analysis and confusing analytical results.

SOIL BIOTA

3.1. ANIMALS

Animals living in soil range from as large as ground hogs to as small as the smallest insect and arthropod. However, for our purposes, when animals become so small that a microscope is needed to see them, they are classified as microorganisms. It is assumed that the main additions to soil from animals are urine and feces. These are indeed common additions; however, all animals add hair, skin and saliva, as well as the dead and decaying bodies of the organisms themselves. In addition to biochemicals and bioorganic and organic molecules, animals cause both large and small changes in the physical characteristics of soil that can change its chemistry and the results of chemical and instrumental analysis.

The effects of animals on the physical and chemical characteristics of soil tend to be locally distributed in the sense that the holes are dug and urine and feces and other waste products are deposited in localized areas. Animal holes can lead to the movement of surface soil lower in the soil profile such that A horizon material can be found in what would be expected to be a B horizon. This occurs most often with the larger burrowing animals. With smaller animals, material from lower levels in a soil may be brought to the surface such as in the case of ants and termites. An ant colony and a grub found about 9 cm deep in soil is shown in Figure 3.2. In other cases, such as with worms, soil may be intimately mixed with organic matter but not moved long distances. There are many different types of worms in soil, such as nematodes, but only a few cause this important type of change.

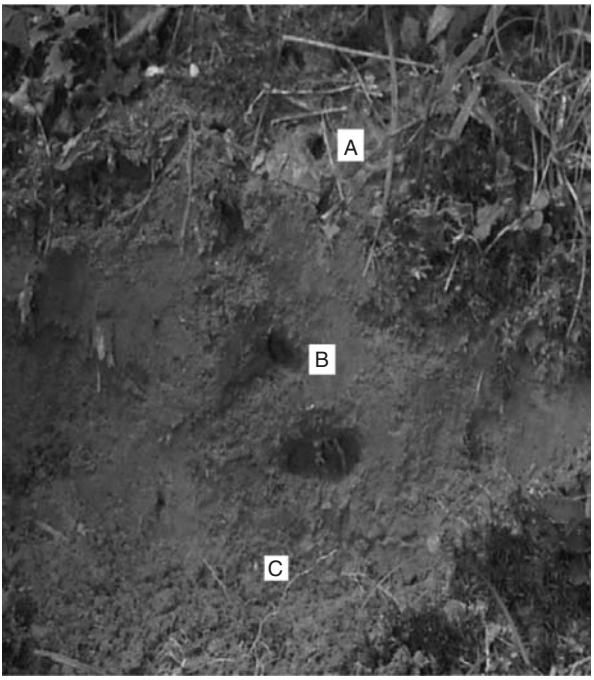


Figure 3.2. An ant colony: *A* is soil brought to surface by ants, *B* is ant holes in the subsurface, *C* is a grub found at of about 10 cm.

The movement of animals through soil and their deposition of organic matter can dramatically affect the soil's structure. As explained in Chapter 2, pushing together soil separates results in the formation of peds, increasing air and water movement through soil and changing the oxidation–reduction conditions and the inorganic and organic species present. Animals, such as worms, which ingest soil solid inorganic particles as they move through soil, cause the degradation and rounding of these particles, and thus have a direct effect on soil inorganic solid components.

Deposition of organic matter will affect areas under and around the deposition. The primary effects are increased microbial activity, increased carbon dioxide, and decreased oxygen content as well as dissolution of molecules into the soil solution. There are also secondary effects caused by the interaction of these molecules with both the inorganic and organic components present. Organic molecules may "dissolve" in the existing soil organic matter. They may also form complexes with inorganic constituents, such as chleate metals present on soil particle surfaces and in the soil solution. Thus, soil samples taken from an area of high animal activity will constitute a matrix that is significantly different from soil taken from an area of low animal activity.

Animals also deposit organic matter on soil even when they do not live in it. Although these deposits are widespread in most cases, they can be concentrated in watering and feeding areas. In all cases soil near and under an organic matter deposition will be affected biologically, chemically, and physically. Before decomposition begins, rain can move both inorganic and organic constituents into and sometimes through the soil profile. During the decomposition process additional organic and inorganic compounds and ions will be produced and leached into the soil.

In addition the physical effects described above, animals can change the soil's biological and chemical reactions. For instance, animal paths become devoid of plants and compacted, thereby decreasing water infiltration and percolation and oxidation–reduction reactions, particularly when there is continual use of the paths [1–4].

3.2. PLANTS

Plants have two parts: the tops and the roots. Both are different and have different effects on soil chemistry and analysis. Because the effects are so different, each part will be discussed separately. The first thing to note about plants is that they all can be divided into algae, fungi, mosses, and liverworts and vascular plants, while the dominant agriculture plants are commonly divided into grasses and legumes. In addition, these types of plants can be annual, biannual, and perennial in their lifecycles. Annual plants are particularly interesting in that both the tops and bottoms die each year and thus add organic matter to soil from both sources.

3.2.1. Tops

Shrubs and trees have woody stems or trunks and moderate to tall growth habits and, along with tall growing woody grasses, such as coconut, are longlived and typically only add leaves to the soil each year. Note that evergreens and needle bearing trees keep their needles all year long; however, needles are continuously lost throughout the year, as are leaves from tropical plants. Organic matter from woody annuals and biennials is added in a similar fashion. Thus, organic matter from roots, stems, and branches is only occasionally added to soil after relatively long periods of time. Addition of organic matter from these types of plants seldom leads to the development of thick O or A horizons.

Although it often seems that leaves, particularly those of deciduous trees, do not decompose, it is observed that the layer of leaves on the ground never becomes thick. In the tropics, where trees grow all year long, the same thing happens; leaves fall continuously during the year and decompose. Thus, the leaves of all plants decompose over a year's period of time, adding organic matter to the soil.

Grasses and other similar plants, which may be annual, biannual, or perennial in their growth habit, do not have woody components, but also add leaves and stems to the soil each year. These leaves and stems decompose over a 1-year period, adding organic matter to the soil surface. Often these leaves seem to decompose faster than do tree leaves; however, in all cases the rate of decomposition will depend largely both the characteristics of the plant material and local environmental conditions.

All components in organic matter affect its decomposition; however, one, the carbon : nitrogen ratio (C/N), is particularly important. Soil organic matter has a carbon : nitrogen ratio in the range of 10 : 1–12 : 1. When organic matter with high C/N ratios, 100 : 1, for example, is added to soil, microorganisms decomposing it take nitrogen from the soil solution and analysis of this soil will result in very low values for inorganic nitrogen. As this organic matter is decomposed, nitrogen will be released. Organic matter with low C/N ratios will release nitrogen to the soil solution. Thus, organic matter will have a dramatic effect on the results of soil analysis. Actively decomposing organic matter will result in changing analytical results over time.

It might be assumed that there will be different organic matter in soil if there are different plants growing on it. This is true when the fresh organic matter and its breakdown products are being investigated. It is particularly evident with Spodosols and Mollisols. Spodosols have a subsurface spodic horizon, which results from decomposing acid detritus, leading to leaching of aluminum and highly decomposed organic matter and often but not necessarily, iron oxides, to form this horizon. In Mollisols the deposition of both grass tops and roots each year leads to the development of a thick dark surface mollic horizon.

Despite these dramatic effects on soil, the organic matter remaining after the breakdown of plant residues, namely, humus, is generally the same the world over. The interaction of humus with chemicals, adsorption, cation exchange, and so on, including those used in analytical procedures, is generally similar. Thus, often the type of organic matter being added to soil, except as noted above, is not as important as the amount of decomposed organic matter already present. However, the components present in humus, specifically, humic and fulvic acids in humus, vary considerable and thus can change some of its characteristics. Soil humus will be discussed in more detail below [5].

3.2.2. Roots

It is reasonable to assume that because roots and tops are part of the same plant, their effects on the soil would be the same. However, this is not the case. Plant roots, because they are in intimate contact with and are constantly extracting nutrients and water from and exuding materials into the soil, profoundly affect its characteristics. This intimate relationship, which includes physical, microbiological, biochemical, bioorganic, and chemical interaction between roots and soil, is illustrated in Figure 3.3.

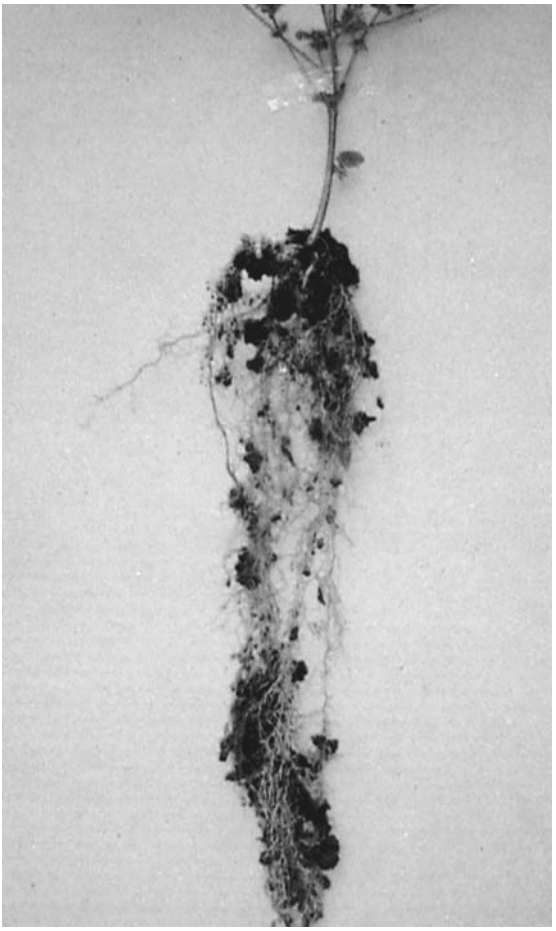


Figure 3.3. Plant roots with adhering soil illustrating the interaction between plant roots and soil.

Any one of these criteria can be used to distinguish an area around the roots, called the *rhizosphere*, from bulk soil. In addition to these general characteristics, the rhizosphere is the area, as shown in Figure 3.4 around plant roots where there is high microbial activity, increased carbon dioxide, decreased oxygen, decreased water and nutrient content, and decreased pH. These conditions develop because of root metabolic activity, exudates, and cells sloughed off by roots. Root exudates are specific for each plant and contain a wide range of organic and inorganic compounds. Both cells and exudates provide "food" for increased microbial activity. The rhizosphere is also an area where contaminants, which are mobile in soil but not taken up or slowly taken up by plants and roots, will accumulate.

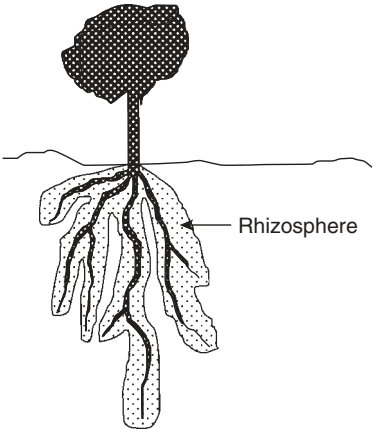


Figure 3.4. Illustration of the concept of the rhizosphere as an area around plant roots.

One well-studied root type is that of the legumes, which are infected by a microorganism called *rhizobia*, resulting in the formation of nodules on the roots. A symbiotic relationship exists between the rhizobia and the legume wherein rhizobia supply fixed nitrogen to plants and plants provide carbohydrates, an energy source, to the rhizobia. Fixed nitrogen is any nitrogen atom bonded to another atom other than nitrogen (N_2), as in NH_3, NO_3^-, $(NH_2)_2CO$; in rhizobia the nitrogen compound produced is ammonia, which is used by plants to produce amino acids.

It is to be expected that soil with a sod cover, that is, with thick grass and roots, will have characteristics different from those of a soil on which few or no plants are growing. These differences will be important in analyzing soil for components of concern.

There are many types of roots, ranging from thick fibrous, deep tap, shallow, tubers, all in one plant community. Some roots explore the soil to significant depth (i.e., as much as 250 cm deep), while others are shallow (i.e., only 25 cm deep). Different rooting depths are found in all plant types; grasses, legumes, shrubs, and trees. Each root type will contribute its own unique exudates and characteristics to its unique volume of soil and the associated soil solution.

Plant roots *respire*, taking in oxygen and giving off carbon dioxide. This is a simple but essential process, and most land plants die if their oxygen source is interrupted for even a short period of time. There are, however, some plants, including crops, which grow with their roots submerged in water, that is, under anaerobic and reducing conditions. Cyprus is an example of such a tree, while rice is an example of a crop. These plants have developed a vascular system that conducts oxygen to the roots, allowing them to function. However, it is important to note that the environment of these roots is very different from that encountered by roots in unsaturated soil. For instance, anaerobic con-

ditions result in increased solubility of iron and other metals along with reduced forms of carbon and sulfur.

Reduced forms are generally more soluble and thus more easily extracted than are oxidized forms. High levels of reduced forms, including some plant nutrients, for instance, iron, may result in levels toxic to plants or other soil organisms. Thus, not only the species but also the extractability and thus the apparent level of soil constituents in the rooting zone will be affected by whether the soil is under oxidizing or reducing conditions at the time of sampling and analysis. Storing soil samples from an aerobic soil under reducing conditions will drastically alter analytical results, as will storing a soil sample from an anaerobic soil under aerobic conditions [6–10].

3.3. MICROORGANISMS

The most diverse group of organisms growing in one location, in soil, are microorganisms. Here we define microorganisms and any organism that is visible only under a microscope. Using this definition, algae, fungi, actinomycetes, bacteria, and even some worms, arthropods, ciliates, and other organisms, will be included in this group. This represents a truly diverse group of organisms capable of carrying out an immense diversity of physical, biological, and chemical changes in their environment.[1] Aerobic, anaerobic heterotrophic, autotrophic, thermophilic, mesophilic, and cryophilic are only some of the different types of microorganisms found in soil. Table 3.1 gives the characteristics of these different types of organisms. An indication of their power and importance is seen when it is noted that some of these organisms can create a whole new cell out of only carbon dioxide, light, and a mix of inorganic compounds, including nitrogen and ions!

Figure 3.5 is a drawing of soil microorganisms; Figure 3.6 is a photomicrograph of common soil microorganisms, which are differentiated by their shape and size. While the actinomycetes and fungi are both filamentous in growth pattern, actimomycetes are smaller and less branched while fungi have larger, more highly branched mycelia. Cocci and bacteria can be found in virtually all the environments, hot or boiling water, high acidity (pH 1), cold (0°C), basic (pH 12), and high salt concentrations. Fungi, which are aerobic, are somewhat more restricted in the environments that they inhabit; however, some species can live in high-osmotic-potential environments, such as sugar syrups, as well as in or on hydrocarbon mixes, such as diesel fuels.

Although all microorganisms in soil are important, most attention is focused on bacteria (often in discussions there is little or no distinction between bacteria and cocci; both are usually lumped together and simply referred to as “bacteria”). This is because they are extremely numerous and

[1] The three references to Web sites given in the Bibliography provide additional information and links to other relevant sites concerning microbial activity in soil.

Table 3.1. Designations of Different Types of Organisms Common in Soil

Organism	Characteristic
Aerobic	Lives in the presence of oxygen and carries out oxidation reactions using oxygen as an electron acceptor
Anaerobic	Lives in the absence of oxygen and uses electron acceptors, other than oxygen (i.e., nitrogen and carbon)
Heterotrophic	Requires preformed organic compounds for energy and cell production
Autotrophic	Obtains energy from oxidation of inorganic ions and obtains carbon from carbon dioxide
Thermophillic	Grows at elevated temperatures; may be any of the types of organisms listed above (>40°C and ≤100°C)[a]
Mesophilic	Grows at moderate temperatures; may be any of the types of organisms listed above (>5°C and <40°C)
Cyrophillic	Grows at low temperatures; may be any of the above types of organisms listed above (<5°C)[a]

[a] Organisms have been reported as growing beyond these temperature ranges.

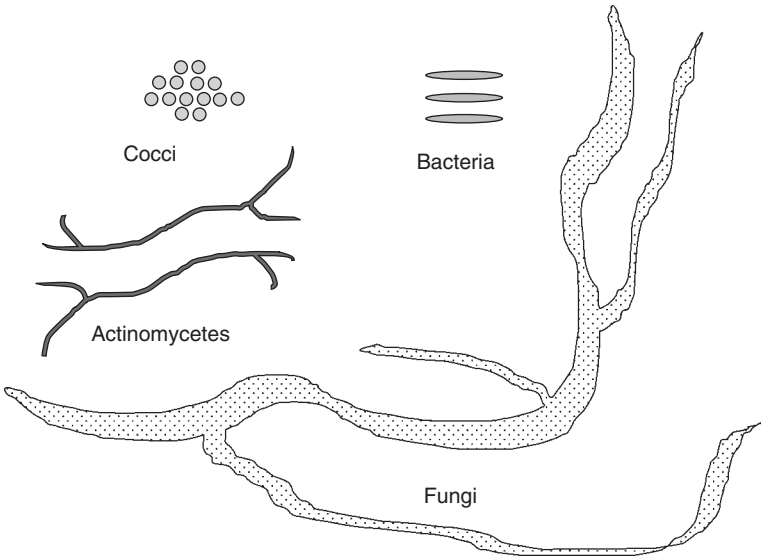


Figure 3.5. Common soil microorganisms (not drawn to absolute or relative scale).

versatile in the reactions they carry out and in well-drained soils they inhabit aerobic (oxidizing), anaerobic (reducing), and microaerophilic zones in soil peds. Microaerophilic zones are those with low concentrations of oxygen. In Figure 3.7 the area marked as *A* will be anaerobic, while the area just inside the mouths of the pore but before the inner water containing areas will be

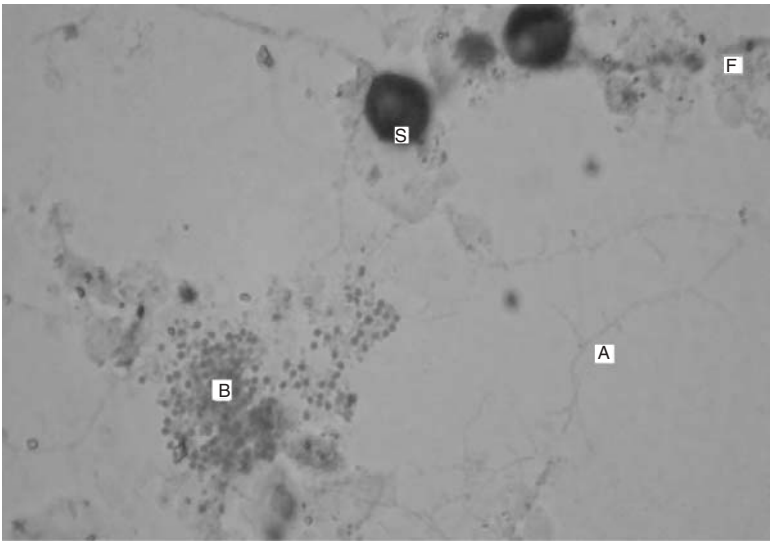


Figure 3.6. A view of soil microorganisms obtained by burying a slide in soil for several days after removal and staining. Bacteria (B), actinomycets (A), spore (S), and fungi (F) can be clearly seen.

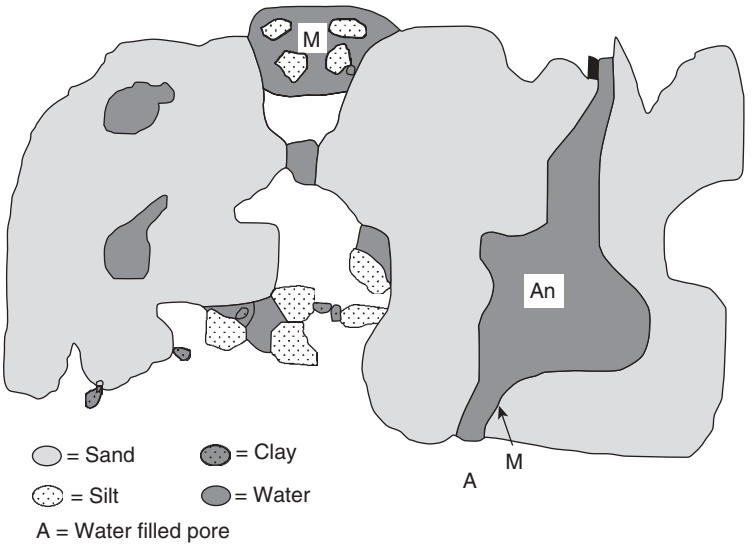


Figure 3.7. A representation of a soil ped showing aerobic areas around peds and at pore mouths (A) and anaerobic areas (An). The pore labeled A will be anaerobic in the middle and will not drain because of the small diameter of the pore mouths. The points labeled M will be microaerophilic.

microaerophilic, while all other areas will be either aerobic or anaerobic. Because of these environmentally different areas, a well-drained soil will have both oxidized and reduced forms of all components present at the same time. Carbon dioxide (CO_2), oxidized and methane (CH_4), reduced carbon, sulfate (SO_4^{2-}), oxidized and hydrogen sulfide (H_2S), reduced sulfur, ferrous (Fe^{2+}) reduced, and ferric (Fe^{3+}) oxidized iron can be found in soil at the same time.

In continually submerged soils there is no oxygen, and so the entire environment is anaerobic and reducing. Under these conditions there will be a predominance of the reduced forms mentioned above, namely, methane, hydrogen sulfide, and ferrous iron.

Soil microorganisms play an extremely important role in cycling of environmental elements such as carbon, nitrogen, and sulfur. The cycling of these elements and others is often represented as their respective cycle (carbon cycle, nitrogen cycle, sulfur cycle, etc.) [11]. Of these the two most important are the carbon and nitrogen cycles. Organisms chiefly responsible for the carbon cycle, animals, plants, and microorganisms change carbon from carbon dioxide to plant and animal tissue and eventually back into carbon dioxide. A critical step in this process is the decomposition of organic matter by microorganisms, and if the organic matter is in contact with soil, these will be soil, microorganisms. During this process other elements important to life are either taken up or released to be used again. One of these other elements, nitrogen, and its cycle have many critical steps carried out by soil microorganisms.

There are many other cycles such as the phosphorus, potassium, halogen, and sulfur cycles; the latter is illustrated in Figure 3.8. All the transformations illustrated are carried out by soil microorganisms. It is interesting that sulfur is converted from its elemental form to either fully oxidized or fully reduced forms by various microorganisms in soil. The starting point in the cycle may be either a reduction or an oxidation depending on the electron status of the starting and ending compounds and the environment where the reactions are occurring. It should also be noted that there are a broad range of organic sulfur compounds, linear, branched, cyclic, and aromatic, which, although not shown, can also occur as part of the sulfur cycle [11].

Microorganisms are important in soil chemistry, soil analysis, and instrumental methods for two extremely important reasons: (1) between taking soil samples and their analysis, these organisms can cause extensive changes in the chemistry and chemical composition of the soil sample, completely changing the species and amount of components found; and (2) almost all extraction procedures will cause the destruction of any cells, animal, plant, and microbial, found in soil with the subsequential release of cellular constituents into the soil solution. Some constituents will be degraded by the extraction process, some enzymes may continue to function, and some cellular components will complex or form chelates with metallic components in the soil. These reactions will lead to a more complex mixture of components than one might expect to be the case. All of these eventualities can affect or change the analytical or instrumental results obtained [11–14].

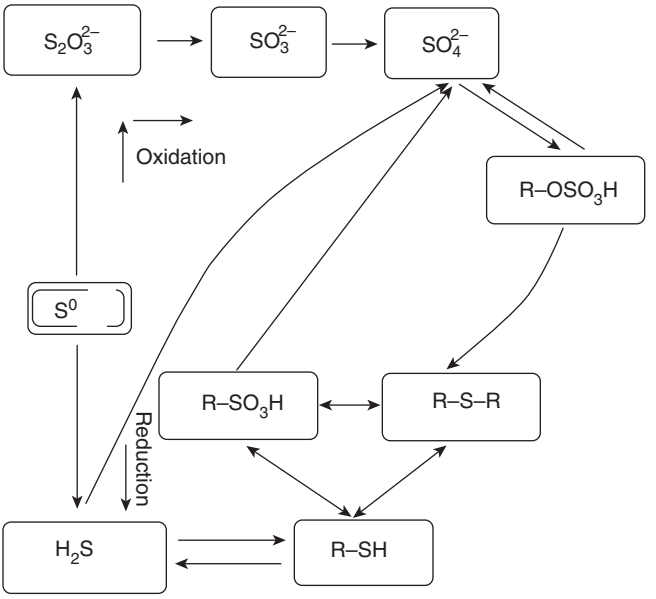


Figure 3.8. The sulfur cycle where S^0 is elemental sulfur, H_2S is hydrogen sulfide, $S_2O_3^{2-}$ is thiosulfate, SO_3^{2-} is sulfite, SO_4^{2-} is sulfate, $ROSO_3H$ represents a sulfate ester, RSO_3H a sulfonic acid, RSR a thioether, and RSH a thiol (adapted from Coyne MS. *Soil Microbiology: An Experimental Approach.* Boston: Delmar Publishers, 1999).

BIOLOGICAL AND ORGANIC CHEMICALS OF SOIL

3.4. BIOCHEMICAL

There are two sources of biochemicals in soil; one is the cellular constituents released when cells are destroyed during the extracting process. It should be kept in mind that any handling of a soil sample will cause the destruction of some of the cells it contains. The simple acts of sieving, air drying, and weighting soil will cause some lyses of cells and release of their contents. Extraction will typically cause complete destruction of all cells in soil, with the release of all their constituent parts. Some parts such as enzymes may continue to function after release from the cell and continue to change the makeup of soil components for some time.

The second source of biochemicals is molecules excreted from cells such as extracellular enzymes and unused organic matter. A typical example is cellulase, which is excreted by fungi such as *Penicillium* in order to break down wood and woody material into sugars that can be used by the organisms. Other common extracellular enzymes found in soil are ureases and amalyases. Often enzymes are associated with clay particles, and in such associations their

activity may be increased, decreased, unchanged, or completely destroyed [15,16].

All biomolecules tend to be large polymer or polymerlike combinations of individual molecular units that can be isolated by either acidic or basic hydrolysis. Lipids and fats are found as integral parts of membranes and cell walls. Polysaccharides are used as structural units and as stored energy sources. Proteins are used to construct muscle and enzymes that also contain metals such as zinc, manganese, and iron. There are many other important biomolecules present at lower concentrations, such as DNA and RNA, which are also released into the soil solution. All can be the source of smaller molecules in the soil solution.

The different groups of biomolecules, lipids, polysaccharides, and proteins, illustrated in Figure 3.9, decompose at different rates depending on their composition. Lipids and fats are slower to decompose in soil because of their insolubility in water. Large polysaccharides are also insoluble in water but are more quickly decomposed than are fats. Proteins and compounds such as DNA and RNA are more quickly decomposed in part because they contain nitrogen (fixed), which is often in short supply in the environment.

Biochemicals will be present in soil during any analysis and can react with components of interest, either organic or inorganic, including sand, silt, and clay particles. Possible reactions include chelation, decomposition, precipitation, solubilization, or dissolving such as dissolving in soil organic matter (humus). Several of these reactions will take place simultaneously and can lead to nondetection of the component of interest or an analytical result that is much lower than the true value [15].

3.5. BIOORGANIC

Bioorganic components in soil include those organic molecules that participate in biochemical reactions; are responsible for the initiation of a reaction, eliciting the formation of a compound or an antibiotic; or are inhibitors. Bioorganic chemistry also uses synthesized models and molecules, to study biological processes such as enzymatic activity. Often these studies are undertaken to develop a mechanism for the reactions of interest. Bioorganic molecules will be present either as components of the synthesis chain or as part of the degradation products. Whenever cells lyse, these compounds will be released into the soil solution. Their moderate size and complexity will allow them to be metabolized rapidly.

Because of their moderate size and complexity, bioorganic molecules can be confused with analytes of interest. They can have the same or similar retentions times, during chromatographic procedures, as the analytes of interest and thus can indicate the presence of more molecules than are actually the case or even the presence of an analyte or contaminant when it is not or has not been added from an external source. It is also possible for gas chromatographic/mass

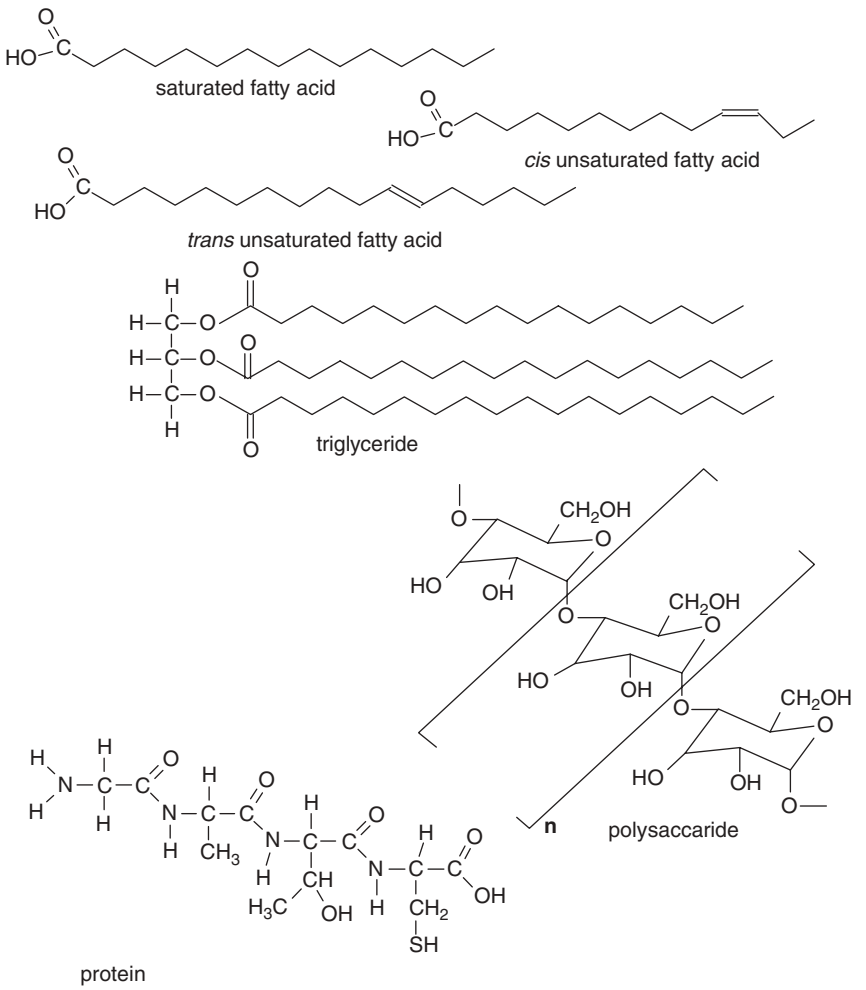


Figure 3.9. Common biological molecules deposited in soil.

spectroscopic (GC/MS) procedures to give confusing results when compounds are not adequately separated by the chromatographic system. These compounds may also mask analytes of interest by reacting or simply associating with them.

3.6. ORGANIC COMPOUNDS

Organic chemistry deals with the relatively simple organic compounds, most of which are easily synthesized in the chemical laboratory and are found in all environments. They are composed of carbon, hydrogen, oxygen, and nitrogen

and may contain smaller amounts of other elements such as the halogens, phosphorus, and silicon. They can be straight-chained, branched, or cyclic, and may contain single, double, and triple bonds in any except the simplest molecules. There are nine principal organic functional groups, as shown in Table 3.2. There are also four important functional groups derived from the acid functional group as shown in Table 3.3.

These functional groups along with the general structure of the molecule will determine its solubility and its ease or resistance to degradation. Typically, as the number of groups capable of forming hydrogen bonds or associating with hydrogen on the water molecule increases, the more soluble an organic molecule will be. Conversely, the longer the straight-chain hydrocarbon portion of a molecule, the less soluble it is. Low-molecular-weight alcohols, aldehydes, ketones, acids, and amines are soluble, while high-molecular-weight members of these families are insoluble. These represent a very large group of compounds with simple and complex structures, including cyclic and multicyclic compounds containing several functional groups and having a wide range of solubilities.

Cyclic organic structures are widespread and common. They can contain any of the functionalities listed in Tables 3.2 and 3.3. Because of bonding angles and steric effects, some will be highly unstable while others will be particularly stable and resistant to decomposition.

Table 3.2. The Organic Functional Groups

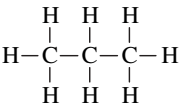
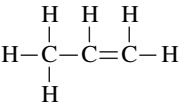
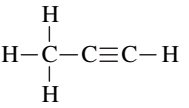
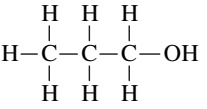
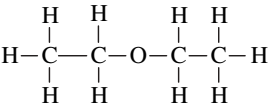
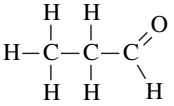
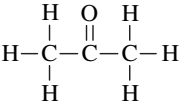
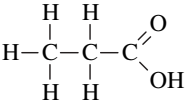
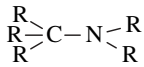
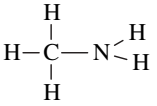
Family Name	Composition	Structure and IUPAC[a] Name
Alkanes	Carbon, hydrogen-bonded with single bonds	 Propane
Alkenes	Carbon, hydrogen, single bonds and containing at lease one double bond	 Propene
Alkynes	Carbon, hydrogen, single bonds and containing at least one triple bond	 Propyne
Alcohols	Carbon, hydrogen, single bonds with at least one —OH (may contain double and triple bonds and other structures)	 1-Propanol

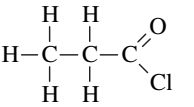
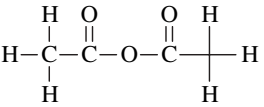
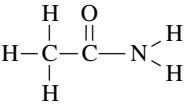
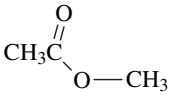
Table 3.2 (*Continued*)

Family Name	Composition	Structure and IUPAC[a] Name
Ethers	Carbon, hydrogen, single bonds with at least one —C—O—C— in the molecule (may include double and triple bonds and other structures)	 Diethl ether
Aldehydes	Carbon, hydrogen, single bonds with at least one —CHO in the molecule (may include double and triple bonds and other structures)	 Propanal
Ketones	Carbon, hydrogen, single bonds with at least one  in the molecule (may include double and triple bonds and other structures)	 Propanone (acetone)
Acids	Carbon, hydrogen, single bonds with at least one  in the molecule (may include double and triple bonds and other structures)	 Propanoic acid
Amines	Carbon, hydrogen, single bonds with at least one  in the molecule (may include double and triple bonds and other structures); R groups may be carbon or hydrogen	 Methyl amine

[a] IUPAC is the International Union of Pure and Applied Chemistry, which determines nomenclature for organic compounds.

Any of these compounds can be found and even synthesized in soil. The simplest example methane (CH_4), is commonly found in the soil atmosphere. It is produced during the decomposition of organic matter under anaerobic conditions, which can occur even in aerobic soils. It is interesting to note that methane can not only be produced in aerobic soils but can also be oxidized by soil bacteria in the same soil.

Table 3.3. Functional Groups Derived from the Acid Functional Group

Family Name	Composition	Structure and IUPAC Name
Acid halides	Carbon, hydrogen, single bonds with at least one —COX[a] in the molecule (may include double and triple bonds and other structures)	 Propanoyl chloride
Anhydrides	Carbon, hydrogen, single bonds with at least one —COOOC— in the molecule (may include double and triple bonds and other structures)	 Ethanoic anhydride
Amides	Carbon, hydrogen, single bonds with at least one —$CONH_2$[b] in the molecule (may include double and triple bonds and other structures)	 Ethanamide
Esters	Carbon, hydrogen, single bonds with at least one —COOC— in the molecule (may include double and triple bonds and other structures)	 Methyl ethanoate

[a] X is always used as a general representation for any of the halogens.
[b] Hydrogens on the nitrogen can be substituted with alkyl groups.

In addition to methane, other simple organic compounds will be found in soil from two different sources. They can be either exuded from roots into the rhizosphere of plants or derived from the decomposition of any organic matter present. During decomposition all organic matter is broken down into smaller and smaller organic molecules until it is finally completely converted into carbon dioxide, water, and humus.[2] The same is true for anaerobic decomposition, except that one of the final decomposition products is methane. Thus, at any given time, intermediate decomposition products from these two sources can be found in the soil solution.

Plant roots excrete a mixture of simple and complex compounds and materials, which differ for different plant species. First, simple acids, both fatty and amino; sugars; and phenolic compounds are commonly detected as exudates from plant roots. Each of the these groups may be composed of a complex mixture of compounds; thus, the most common amino acids can be found along with many common sugars. In addition, a high-molecular-weight compound called *mucigel* is secreted by root tip cells and is thought to lubricate root penetration into soil. Mucigel is somewhat slower to decompose but can also be

[2] Although seldom mentioned, this process is characterized by the release of energy that is utilized by the organism carrying out the decomposition.

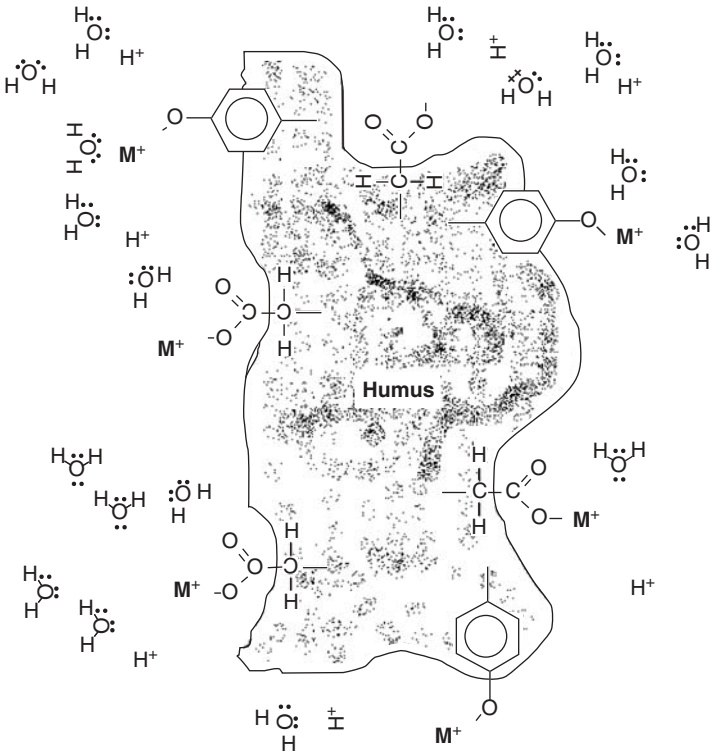


Figure 3.10. Humus with cation exchange sites created by the ionization of phenolic and acidic functional groups. The M^+ represent exchangeable cations.

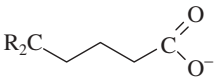
a source of simple organic molecules during its decomposition. These compounds help determine the microorganisms present in the plant's rhizosphere.

No matter what the source of organic matter or the mechanism of its decomposition, an extremely important compound, humus, is formed during decomposition [6–10,17].

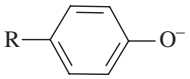
3.6.1. Humus

Humus is the material synthesized or resynthesized during decomposition of organic matter by microorganisms. It is produced under both aerobic and anaerobic conditions and remains after decomposition of the original organic matter is complete. Humus is a complex molecule often described as being a polymer even though no mer[3] unit has ever been found. It is black or dark brown in color and has a high affinity for organic molecules, cations, and water. Organic molecules associate with humus via a process that is similar to the molecule dissolving in it. Cations are held by cation exchange involving both acid and phenolic groups (see Figures 3.10 and 3.11). A base extract of humus

[3] A *mer* is the individual repeating unit from which a poly*mer* is formed. Thus polyethylene is made of ethylene units, the mer, bonded together.



Carboxylate anion



Phenoxy (the anion of phenol 

Figure 3.11. The carboxylate anion (an ionized acid) and a phenoxy (the anion of phenol) groups. The R stands for the rest of the humus molecule to which these groups are attached.

Figure 3.12. A basic extract of an organic soil is shown on the left; a sample of the sodium salt of humic acid is seen on the right.

from an organic soil and a commercial sample of the sodium salt of humic acid are shown in Figure 3.12.

Many researchers have attempted to unravel the mystery of the structure of humus. One approach has been to isolate "fractions" by extracting humus

using various extraction procedures. These procedures result in the isolation of three or more "fractions": humic acid, fluvic acid, and humin. Humic material is isolated from soil by treating the soil with alkali. The insoluble material remaining after this treatment is called *humin*. The alkali solution is acidified to a pH of 1.0 and the precipitate is called *humic acid* while the soluble components are called *fluvic acid*. A lot is known about these components; for instance, they contain three carbon or propyl groups, aromatic moieties with various and usually multiple functional groups, and variable amounts of other components. The isolation of these components, however, has not brought us much closer to a complete understanding of the structure of humus. Applying other extraction procedures will allow the extraction and isolation of other "fractions" of humus; however, humin, humic acid, and fluvic acid are the three main components likely to be discussed in terms of humus.

What is known is that humus is an extremely important component of soil. Even small amounts can cause demonstrable differences in a soil's CEC and its other chemical and physical properties as well. It is active in binding soil particles together to form peds, increases the soil water holding capacity, and increases the absorptive capacity of soil for organic and inorganic constituents, both natural and synthetic. As an example of the importance of humus in terms of soil water, it could be noted that mineral soils absorb and hold 20–40% of their weight in water, while some organic soils can hold 10 times this amount, specifically, 200–400% of their weight in water. This is due in part to the fact that organic soils are much lighter than mineral soils, but nevertheless this increased water holding capacity is dramatic. Increased sorptive capacity is reflected in the increased need for herbicides on soils high in organic matter.

Different soils contain different amounts of humus. Some tropical African soils may contain less than 0.1% organic matter. At the other extreme organic soils, such as Histosols, generally must have 20% or more organic carbon in the upper 80 cm, although this will vary somewhat depending on the conditions of moisture, texture, and depth of the soil. Many agricultural soils contain 1–2% organic matter, although it is not unusual to find soils containing 10% organic matter.

When developing a soil analytical method, it is essential that either the method be applicable to soils of all organic matter contents or that variations of the procedure be applicable to different soil organic matter contents be developed [16,18].

3.7. ANALYSIS

There are a myirad of methods used for analyzing soil for animals, plants, and microorganisms. All are well developed and easily carried out; however, some are more useful when large numbers of samples are to be analyzed, while some are subject to significant error [19–21].

3.7.1. Analysis for Animals and Plants

Determination of the number of animals in a cubic meter of soil is usually done by simple isolation and counting. Plants are usually counted as the number per square meter, hectare, or acre. Determination of root numbers or mass is extremely difficult and is commonly done by assuming that there is a certain mass of roots associated with a plant's top, that is, the ratio of roots to top masses. Determination of microorganisms is usually done using standard microbiological techniques such as dilution plate counts. Because of the extreme diversity of soil microorganism, it is impossible to determine all the organisms present at one time by any one technique. In some cases direct microscopic observation, as was done to produce Figure 3.6, is used to estimate the numbers of microorganisms present [2–4].

3.7.2. Determination of Soil Organic Matter

The analysis of soil for organic matter[4] is straightforward, involving oxidizing it to carbon dioxide and water. Oxidation can be accomplished in a number of different ways, such as by applying air at high temperature, pure oxygen, or chemical oxidation using various common chemical oxidizing agents such as hydrogen peroxide, permanganate, and dichromate followed by titration. Of these, oxidation by hot dichromate followed by titration of unreduced dichromate is most commonly used.

The real problem is the complete oxidation of soil organic matter. As noted previously, this can be associated with mineral surfaces or in pores to which there is difficult access. For these reasons, simple oxidation will not be sufficient, and drastic oxidation procedures will be necessary.

3.7.2.1 Direct Oxidation Using High Temperature and Atmospheric Air

The simplest and least expensive method of determining the organic matter in soil is high-temperature oxidation using air. In this case a weighed soil sample in a crucible is placed in an oven and dried. Subsequently, after a prescribed time at an elevated temperature, it is again removed and weighed, and the difference is taken as the amount of organic matter. This procedure is seemingly simple, straightforward, and easy to perform and does not involve the expense of waste disposal. While oxidation of organic matter with a consequent loss of weight does occur, so do other reactions, which lead to a loss of weight. One such reaction is the loss of waters of hydration from soil minerals. Another is the decomposition of soil minerals. For instance, carbonates, which are very common soil minerals, decompose at high temperature to form

[4] Specific directions for determination of soil organic matter are given in the bibliographic references.

the respective oxide and carbon dioxide, which is lost, resulting in a loss of weight in the sample.

For these and many other reasons, direct oxidation of soil organic matter using high temperature and atmospheric air is not the best procedure for determining soil organic matter content.

3.7.2.2 Oxidation Using Dichromate

The most common method used in determining soil organic matter content is oxidation using dichromate. In this procedure a mixture of potassium or sodium dichromate with sulfuric acid is prepared and mixed with a soil sample. The heat released from mixing the soil with the dichromate and other accessory solutions heats the mixture to around 100°C for a specific period of time. Alternatively, external heating may be used or required. After cooling, an indicator is added and the unused dichromate titrated. The amount of dichromate remaining can be related back to the amount of carbon and organic matter present (see also Chapter 6, Section 6.3). Because of the relative low temperatures and the reagents used, this is a reliable method, which does not lead to loss of other components except carbonates. The loss of carbonate does not involve reduction of dichromate but can cause interferences that must be taken into account [21].

> ***Caution:*** It should be noted that this procedure involves the use of hazardous chemicals by laboratory personnel. It also results in hazardous waste that must be disposed of carefully and properly.

3.7.2.3 Other Oxidative Procedures

Potentially any oxidizing reagent can be used to determine soil organic matter. Of the many other possibilities, the use of catalytic oxidation in pure oxygen and or application of hydrogen peroxide are the two more common methods. Any organic chemical can be oxidized to carbon dioxide and water using catalyst, pure oxygen, and heating. A known amount of material is placed with a catalyst in a stream of oxygen and heated, producing carbon dioxide and water, which are trapped and weighed. From this the amount carbon and hydrogen in the original organic matter can thus be determined. As might be surmised, this method can also be used to determine the amount of nitrogen and sulfur in soil organic matter. This procedure is done on a one-by-one basis and so is slow and time-consuming. For these reasons it is not used where a significant number of samples must be analyzed.

A soil sample can be mixed with hydrogen peroxide and heated to decompose organic matter. This procedure is often used when the objective is to remove organic matter and the amount present is not determined. Problems with this procedure include frothing due to decomposition of hydrogen per-

oxide by components in soil, and questions as to whether all the organic matter is oxidized. Directly measuring the amount of organic matter destroyed is difficult using this method.

There are still other methods of oxidizing soil organic matter. These are not generally or commonly used for a variety of reasons, which can be found by investigating them in the literature [21].

3.8. CONCLUSIONS

Soil biological components are important not only because of their role in mixing and decomposing both the inorganic and organic components therein but also in the addition of organic matter to soil. Organic matter is added by animals, those that live in as well as on the soil, chiefly in the form of manure. Plants add organic matter from their tops and through their roots. Microorganisms are involved chiefly in the decomposition of organic matter deposited by animals and plants. However, they also produce many different inorganic and organic compounds during the decomposition process. One of the most important of these components is humus. Analysis of soil organic matter is accomplished by oxidizing the organic matter using various oxidative chemical procedures. Animals, plants, microorganisms, and organic matter in soil will have a pronounced effect on the soil's characteristics and on procedures used to analyze it.

PROBLEMS

3.1. Name some common types of animals, plants, and microorganisms commonly found in soil.

3.2. Explain how animals can change the physical characteristics of soil. Give some examples.

3.3. In soil microbiology microorganisms are defined by which simple characteristic? Describe the variety of organisms that have this characteristic.

3.4. Explain how animals and plants add organic matter to soil and how this addition is different for these two different groups of organisms.

3.5. What is the rhizosphere, and why is it important?

3.6. What is the difference between a biomolecule and an organic molecule?

3.7. Diagram the breakdown of organic matter in soil and give the products of this decomposition process.

3.8. Explain how humus is formed in soil and why it is important. Give some examples.

3.9. What features of organic molecules control their solubility in the soil solution?

3.10. Describe two ways in which soil organic matter is measured.

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CHAPTER

4

SOIL BASICS IV
THE SOIL AIR AND SOIL SOLUTION

All soil includes air and water. Soil air is similar to atmospheric air in that it contains the same constituents, but differs in that it has different concentrations of these constituents, and that the relative percentages vary over time. In nature soil is never completely dry; it always has a layer of water surrounding the individual soil particles. The dust cloud behind a car driving down a sandy road may contain 1% water on a dry-weight basis, while the dust behind a car on a clayey road may contain 10% or more water. In the field water content is highly variable and can range from 1% to more than 100% in some organic soils. A saturated soil will lose water through percolation, evaporation, and transpiration. As the water content decreases, two things happen. The air content increases, and the water remaining in the soil is more and more strongly held. At some point, which will depend on a soil's texture, water will be held so tightly that it will become unavailable to plants.

Most components coming in contact with water will dissolve, if to only a minute degree, and thus soil water is not pure, but rather a complex solution of inorganic and organic ions, molecules, and gases that are constantly exchanging between the solid, liquid, gaseous, and biological phases. It is in this solution surrounding soil particles that reactions occur, and in which microorganisms live and function. The type and rate of reactions occurring in soil will depend on the soil air and on the soil solution composition, and thus soil air and water are intimately involved in all things happening in and to the soil.

4.1. SOIL AIR

Soil air is made up of the same basic constituents as atmospheric air; however, the ratios of various gases are different and more variable. First and most importantly, virtually all the void volume of the soil can be occupied by either air or water. The amount of air in soil is thus inversely related to the amount of water present. When the air content is around 50% or more of the void volume, as shown in Figure 4.1, the soil is considered to be aerobic and

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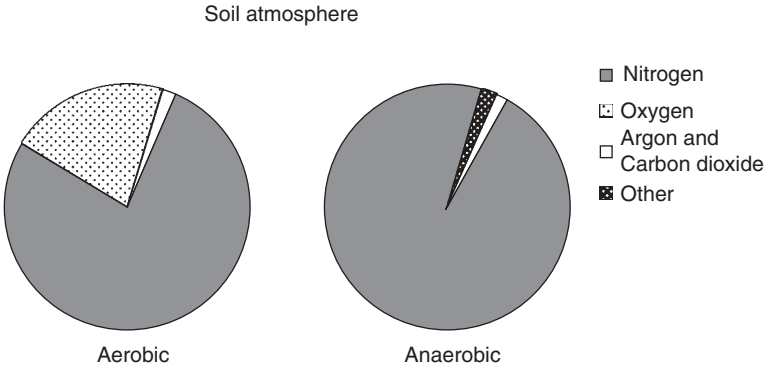

Figure 4.1. Soil atmosphere composition under aerobic and anaerobic conditions.

oxidation reactions predominate. When the void volume is occupied by water, the soil is anaerobic and reducing reactions predominate.

Oxidation reactions in soil, particularly those carried out by microorganisms, and plant roots increase the amount of carbon dioxide in soil air to 10 or more times the concentration in atmospheric air. The consequence of this is that the oxygen content is proportionally decreased. When the soil void volume is almost or completely filled with water, the remaining trapped and dissolved oxygen is quickly utilized by organisms and the oxygen content of any remaining gas is zero. The soil is then anaerobic and reducing conditions prevail (see Figure 4.1).

In addition to oxidation–reduction reactions occurring under various conditions, increased carbon dioxide will also affect soil pH. Carbon dioxide dissolves in water, producing both bicarbonate and carbonate, which will release protons into the soil solution as shown in the following reactions, which illustrate the involvement of CO_2 in control of soil water pH:

$$H_2O + CO_2 \longrightarrow H_2CO_3 \tag{4.1a}$$

$$H_2CO_3 \rightleftharpoons H^+ + HCO_3^- \quad (pK_1\ 6.38) \tag{4.1b}$$

$$HCO_3^- \rightleftharpoons H^+ + CO_3^{2-} \quad (pK_2\ 10.25) \tag{4.1c}$$

$$H^+ + CO_3^{2-} \rightleftharpoons HCO_3^- \tag{4.1d}$$

$$Ca^{2+} + CO_2^{2-} \rightleftharpoons CaCO_3 \downarrow \tag{4.1e}$$

In addition to providing protons, carbonic acid can react with cations to form insoluble precipitates. Both reactions can dramatically alter the composition of the soil solution and soil extracts.

Other gases in the soil atmosphere also change, and thus so will the gaseous components dissolved in soil water, causing its composition to change.

The soil atmosphere also contains water vapor and, in many cases, is at 100% relative humidity. Water vapor evaporating from the soil surface is one mechanism by which water and dissolved components can move upward in a soil profile.

These gases are interesting and important but do not represent all the gases commonly found in analyses of the soil atmosphere. Even in aerobic soils, it is common to find reduced species such as methane. In addition, if ammonium is present in the soil solution, ammonia will be present in the soil atmosphere. Under oxidizing conditions ammonium will be oxidized to nitrite and then to nitrate, which under reducing conditions are converted to gaseous nitrogen oxides, which will also occur in the soil atmosphere. Other gases, such as hydrogen and helium, can be found in the soil under some conditions and at some localities [1–4].

4.2. WATER

Water is a unique molecule, and when it is associated with soil, it is even more unique. The most frequently cited unique characteristics of water are its high melting and boiling points and its ability to dissolve a wide range of molecules and ions. A less often appreciated characteristic of water is it density, which decreases both above and below the freezing point of water with the maximum density actually occurring at a temperature between 3 and 4°C.

These phenomena are related to hydrogen bonding in water, as was discussed in Chapter 2, where it was pointed out that water is the prime example of this phenomenon. In addition, the partially positive hydrogen of water is attracted to electron pairs on any electronegative atom in any molecule, and the partially negative oxygen in water is attracted to any positive atom in any molecule. These attractions can occur between species, molecules, and ions in solution and between water molecules and solid components in the soil. These interactions are illustrated in structures (2.3) in Chapter 2.

One way of thinking about water in soil is to envision it as a layer around and covering a soil particle (see Figure 4.2). As water is removed from outside layers, the remaining molecules are held more strongly. The outermost layers are held with a tension of 0 to –30 kPa,[1] and are removed by the pull of gravity. This is called gravitational water, and normally drains or percolates through the soil and into the groundwater. Soil containing gravitational water contains little or no air and because roots require air to function, this water is generally said to be unavailable to plants.

The next layer of water held between –30 and –1500 kPa, is available to plants and is therefore called *plant available water*. The water present between –1500 and –3100 pKa is held in capillaries so tightly that it is not available to

[1] One kilopascal (kPa) is equal to 1000 pascals, where a pascal is a unit of pressure defined as 1 newton per meter squared (N/m^2).

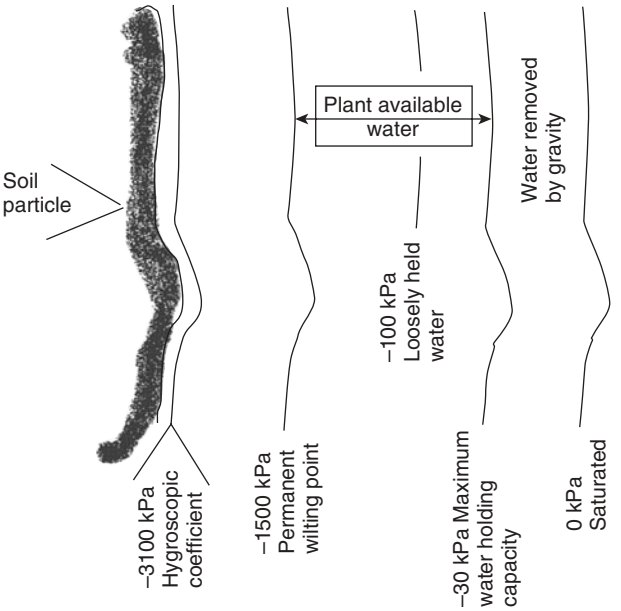

Figure 4.2. Potential of "layers" of water surrounding a soil particle.

plants but can be lost by evaporation. The layer closest to the soil solid is held at more than –3100 pKa and is called *hygroscopic water*. A soil sample, heated in an oven for 24 h at 105°C and then left exposed to the air will adsorb water until a layer of hygroscopic water has been formed, illustrating the strong attraction of water for soil surfaces.

The unit kPa is in common use today and is part of the International System of Units (SI). However, it is also common to encounter the terms *bars* and *atmospheres* (atm) when reading about soil water. One bar is approximately equal to one atmosphere pressure, which is abbreviated atm (–1 bar = –100 kPa).

The movement of water through soil is controlled by the diameter of soil pores and the surface tension of water. Water drains from larger pores and moves down through soil and into the groundwater. In small pores the surface tension of water is strong enough to prevent movement of water into or out of pores. However, because of surface tension, pores will also draw water up from a free water surface until the surface tension is balanced by the pull of gravity. The smaller the pore, the higher the water will be raised. If water, moving down through soil, reaches a compacted zone, for example, having few and extremely small pores, it will move laterally along the dense layer. Thus, water can move down, up, and sideways in soil depending on the soil's pores.

Pores can control water movement in other ways related to size. To understand this control, soil pores can be grouped or classified simply as large, those that allow water to drain or move and small, and those that do not. Water in

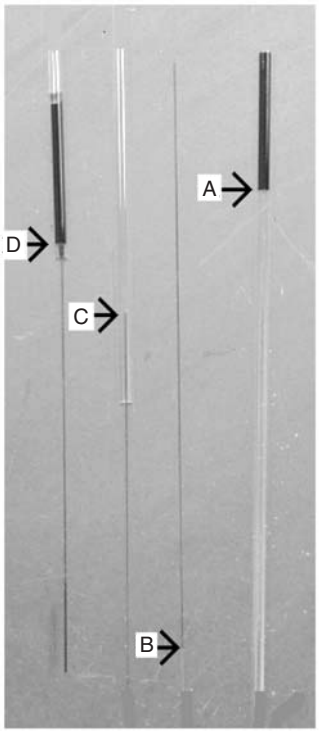

Figure 4.3. A wide-diameter capillary tube (*A*) draws colored a shorter distance than does a narrow-diameter capillary (*B*). Colored water cannot move from the narrow-diameter capillary to the wide-diameter capillary (*C*). Colored water can move from a larger diameter capillary to a smaller capillary (*D*).

large pores will be drawn into smaller pores, but water in small pores cannot move into large pores unless energy is exerted. Thus, water will not move from a sandy horizon—smaller pores—into underlying gravel layer—larger pores—unless the water exerts enough pressure to move from the small pores into the larger pores in the gravel. This relationship between pores and water movement is illustrated in Figure 4.3 using capillaries.

Some pores do not drain because they are simply too small and where the interior is large but the entrances and exits are so small that they prevent drainage as shown in Figure 4.4. Both of these can be called *restricted pores*. Restricted pores can cause problems in any extraction and analysis procedure because they prevent the complete removal of solution from the soil. Exchange with extracting solutions is limited by the slow process of diffusion and can result in the component of interest occurring at low levels even in the last of multiple extractions [5].

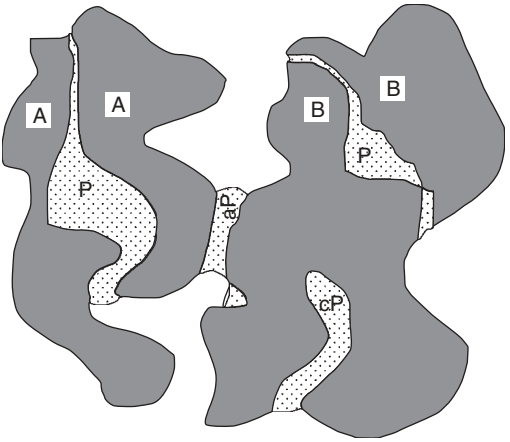

Figure 4.4. Two peds (*AA* and *BB*) each have a pore that does not drain because the mouths are too small. Ped *BB* has a pore closed at one end (i.e., cP). Between the peds is an apparent pore (aP) formed by the close proximity of the peds.

4.3. SOLUBILITY

The solubility of components in the soil solution will be controlled by the innate solubility of the compound in question and the existing soil solution characteristics, particularly salts already present. High salt concentrations will result in salting out and precipitation of some components. Note here that salt concentration is not constant because as the soil dries, the concentration of salt increases and precipitation reactions that may not be reversible when the soil water content is subsequently increased.

In addition to straightforward precipitation reactions, components may dissolve and react with components already present, including atoms on colloidal surfaces. For example, phosphate may dissolve from phosphate rock and react with iron present in the soil solution or on particle surfaces to form an iron phosphate that is insoluble.

4.4. ELEMENTS IN SOLUTION

Elements in the soil solution will be derived from the atmosphere, lithosphere, and biosphere. Thus nitrogen, oxygen, and argon from the atmosphere will commonly be found dissolved in soil water. It is often surprising and sometimes disconcerting to find argon in soil air and water primarily because it is rarely mentioned as a component of air. Although its occurrence may be surprising, it does not represent an unusual situation.

Mercury and the noble metals are found in nature in their elemental forms; however, they are generally unreactive and so their occurrence in the soil solu-

tion is limited. Some elements, such as sulfur, can be reduced to their elemental state by soil microorganisms; however, they both can also be easily oxidized and the oxidized forms reduced and so are rarely found in their elemental form in soil.

4.5. DISSOLVED GASES

Molecular gases, the two most important of which are oxygen and carbon dioxide, dissolve in soil water. Dissolved oxygen results in the soil solution remaining oxidative or aerobic and thus tends to keep components in their highest oxidation state. Microorganisms present will be aerobic and dissolved, and suspended organic matter will undergo oxidative reactions.

Dissolving carbon dioxide produces carbonic acid, which ionizes to bicarbonate and carbonate ions, the reactions for which are shown in reactions (4.1a)–(4.1c). This reaction sequence is extremely important because bicarbonate is a counterion to many cations, is active in buffering the soil solution, and is involved either directly or indirectly in many soil chemical reactions. Bicarbonates are generally more soluble than carbonates, which are generally insoluble. Adding acid to carbonates or bicarbonate results in the release of carbon dioxide and the formation of the salt of the acid cation. The acid is thus neutralized.

At the other extreme, high levels of carbonate or high pH results in the formation of insoluble carbonates, frequently calcium and magnesium carbonates, thus removing them from the soil solution. These reactions thus limit the lower and upper pHs normally found in soil [reactions (4.1d) and (4.1e)]. In addition to carbon dioxide, methane is another common molecular gas in the soil solution. It is largely insoluble in water but will be present because it is constantly being produced by methanogenic bacteria in anaerobic zones in soil. It may seem unlikely that methane would be found in an oxidizing environment; however, as noted above, some pores in soil do not drain and at some distance from their mouths microaerobic and/or anaerobic zones will be found (Figure 4.5). Anaerobic zones are conducive to the formation of methane, hydrogen, hydrogen disulfide, carbon monoxide, or other reduced species in the soil solution.

Any low-molecular-weight organic compound that is normally a gas at standard temperature and pressure (STP) may also be found in the soil solution. These compounds will be produced as a result of the decomposition of organic matter in soil. Many of these will be readily taken up and used by microorganisms and thus their lifespans in the soil solution are short.

4.6. COMPOUNDS IN SOLUTION

The soil solution will contain numerous inorganic and organic compounds derived from the solid components making up the soil. Common compounds

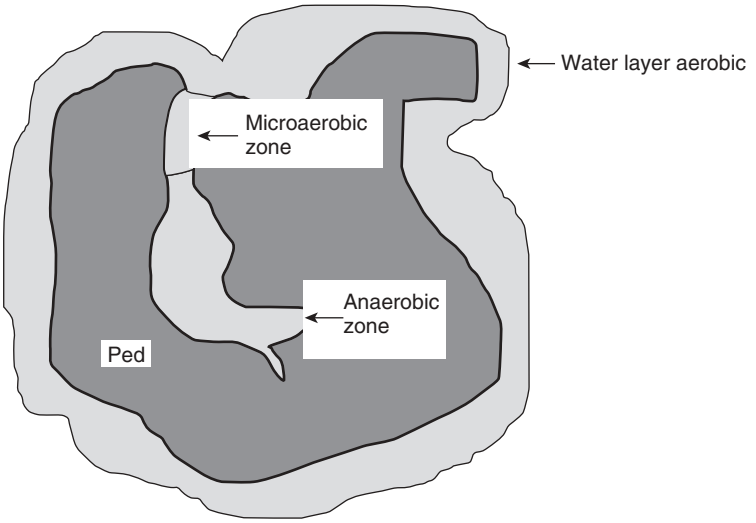

Figure 4.5. Aerobic, microaerobic, and anaerobic zones in a soil ped.

may include, in low concentrations, oxides, particularly those of silicon, aluminum, iron, and titanium. These compounds move down the soil profile sometimes contributing to formations such as the spodic horizon, which can contain aluminum and iron oxides along with highly decomposed carbon.

A large variety of dissolved organic compounds are released by decomposition of organic matter or from the activities of plants and animals in soil. These compounds range from relatively simple molecules such as acetic acid, to relatively complex materials such as enzymes and antibiotics and various cellular components released on cell lyses (see Chapter 3). These and intermediate products serve as sources of carbon and energy for organisms in soil and thus do no persist. However, it is possible for almost any relatively simple water-soluble organic compound to be found in the soil solution at any given time.

Other inorganic and organic compounds are brought into solution by the decomposition of their parent materials. Rocks and minerals will be decomposed by physical, biological, and chemical mechanisms. Enzymes released into the soil solution by microorganisms will decompose insoluble organic materials such as wood. Most of the organic material released with be taken up by the organism releasing the enzyme, but some will find its way into the soil solution.

4.7. INORGANIC IONS IN SOLUTION

Ionic species are generally more soluble in water than are neutral molecules. The polar hydrogens in water are associated with anions and the partially

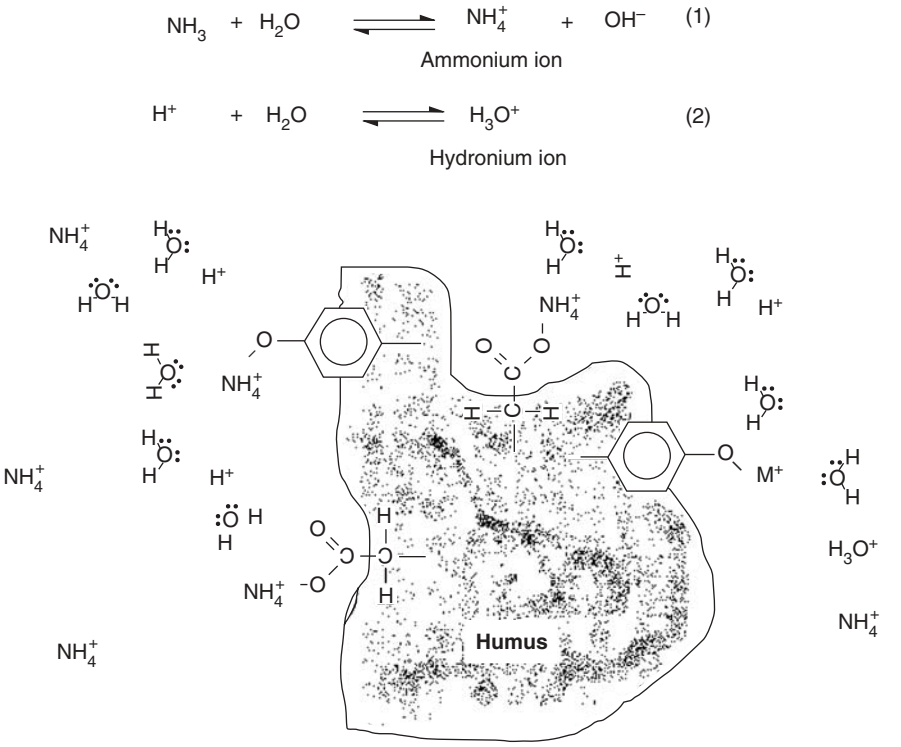

Figure 4.6. Reactions forming ammonium and hydronium ions. Ammonium ions associated with exchange sites on soil humus.

negative oxygens with cations, forming a "shell" of water molecules around them as illustrated in Figure 4.6. Most cations in soil are simple metal ions, although some, such as iron, may be present in multiple oxidation states. An exception to this is molybdenum, which occurs not as a cation but as molybdate, an oxyanion. Two nonmetal cations in soil are ammonium and hydronium (hydrogen ions, protons, associated with water). In contrast there are relatively few simple anions found in soil out side of the halogens, particularly chloride, Cl^-, and bromide, Br^-. Most anions in soil occur as complex oxyanions.

4.7.1. Simple and Multi-Oxidation-State Cations

The most common simple cations in the soil solution are calcium (Ca^{2+}), magnesium (Mg^{2+}), potassium (K^+), and sodium (Na^+). Other alkali and alkaline-earth elements, when present, will be as simple cations also. Iron, aluminum, copper, zinc, cobalt, manganese, and nickel are also common in soil. Iron is present in both the ferrous (Fe^{2+}) and ferric (Fe^{3+}) states, while aluminum will be present as (Al^{3+}). Copper, zinc, cobalt, and nickel can all be present in one

or both of their oxidations states simultaneously. Manganese presents a completely different situation in that it can exist in several oxidation states simultaneously such that it appears to have some intermediate oxidation state.

4.7.2. Multielement Cations

Ammonium and hydrogen (protons) are both present in the soil solution as multielement cations. Ammonia gas reacts with water to produce the ammonium cation, NH_4^+ [Figure 4.6, reaction (1)]. Ammonium acts as a cation in all senses and will be attracted to cation exchange sites on soil particles. Ammonium both in the soil solution and on exchange sites is available to plants.

The hydrogen ion or proton represents a very different situation. When hydrogen (H^+) is released into the soil solution by ionization it loses its electron, the naked proton is naturally attracted to the partially negative oxygen of water and its lone pair of electrons [Figure 4.6, reaction (2)]. The result of this interaction is the species H_3O^+, which is called a *hydronium ion*. This is the true species in the soil solution even though both scientific papers and texts will use the simpler term H^+ when writing equations. The hydronium ion does not act as common cation because of its greater chemical reactivity, especially in cation exchange reactions.

It is important to remember that exchangeable cations (see Figure 4.6), including NH_4^+ and H_3O^+, when attached to exchange sites, cannot be measured directly; they must be brought into solution before analysis can be effected. Thus, extracting solutions must contain a cation capable of replacing all the cations of interest on the exchange sites of the soil. Once in solution, analysis can be carried out.

4.7.3. The Simple and Oxyanions in Soil

Simple anions in soil solution are the halogens, chlorine (Cl^-) and bromine (Br^-). If present, the other halogens will also occur as simple anions. Because the compounds of these anions are generally very soluble, they leach readily out of the soil and so are generally present at low concentrations. Exceptions occur in low rainfall regions where significant, sometimes deleterious (to plants and animals), levels of simple anions can be found.

The two important oxyanions in soil are nitrate and phosphate. Nitrate (NO_3^-) is the predominant oxyanion of nitrogen; however, nitrite (NO_2^-) can also occur in the soil solution. Phosphate can occur as one of three species, $H_2PO_4^-$, HPO_4^{2-}, and PO_4^{3-}, depending on the soil pH. Both nitrate and phosphate are important in plant nutrition and, because of contamination concerns, environmental work. Other important oxyanions are bicarbonate (HCO_3^-) and carbonate (CO_3^{2-}) and the various oxyanions of boron ($H_2BO_3^-$) and molybdate (MoO_4^{2-}).

It is reasonable to expect that since anions and most colloidal particles in temperate region soils have a negative charge, they will repel each other. The

consequence is that anions will pass through soil and not be adsorbed or even retarded. For the simple anions and some of the oxyanions, this is exactly what happens. All the halides, nitrite, nitrate, bicarbonate, and carbonate act in this fashion. However, there are some oxyanions that do not act as expected, and chief among them is phosphate.

Monobasic ($H_2PO_4^-$), dibasic (HPO_4^{2-}), and tribasic (PO_4^{3-}) phosphates react with iron and aluminum at low pH to form insoluble phosphates. In a similar fashion, calcium reacts with phosphate at high pH to form insoluble calcium phosphates. In addition, phosphates will react with clay minerals and organic matter to form insoluble compounds. For these reasons phosphate seldom moves in soil. Exceptions occur when phosphate is associated with some kinds of organic matter, which is moving through the soil and where there is a large excess of phosphate in soil such as in areas where phosphate is mined, such as around the Florida phosphate mines. Other oxyanions will also behave like phosphate, although the exact nature of the reactions leading to their attraction to soil particles and organic matter is not well understood.

Some soils, particularly those in the tropics, have significant anion exchange capacity. For these soils, there is an attraction between soil colloids and the simple halogen and nitrate anions. Bringing these anions into solution for analysis will require an extraction or replacing anion just as does the analysis of exchangeable cations.

4.8. ORGANIC IONS IN SOLUTION

There are three types of organic functional groups—acid, phenolic, and nitrogen—which are commonly ionized in soil (Figure 4.7). Whether the group is

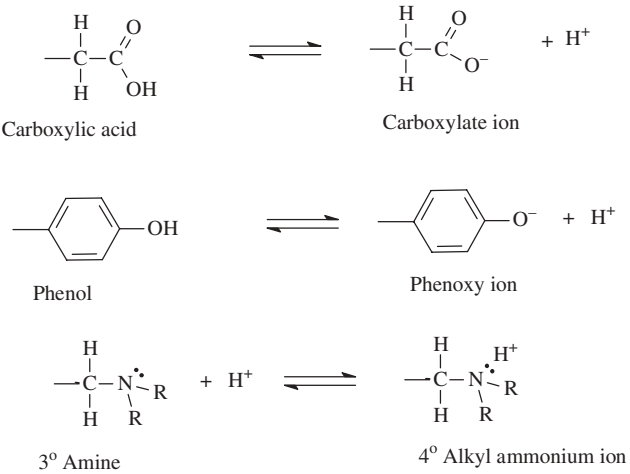

Figure 4.7. Reactions leading to the formation of charged organic species in soil. Note that the unsatisfied bond on the left is attached to some larger organic component in soil.

ionized will be determined by the pK_a of the acid or phenol or in the case of amines, their pK_b. The solubility of organic molecules that contain ionizable groups is greatly increased once they become ionized.

Once ionized, the acid molecule carries a negative charge and can thus attract cations and participate in the cation exchange capacity of soil. The contribution of this source of negative charge will depend on the pH of the soil solution and will change as the pH changes. This is thus a variable or pH dependent cation exchange capacity (CEC).

Because of the variability of this component of soil, the CEC determined at two different pHs should expectedly result in different CEC values; this can also happen if two different laboratories determine the CEC. For this reason it is essential that CEC always be measured in the same way, taking care to define the pH of the solutions used in its determination.

4.9. SOIL pH

In any aqueous solution the pH is a measure of the hydrogen ion or proton activity. However, in many if not most cases, pH is treated as the concentration of protons in solution rather that their activity. The soil solution is no different except that the measurement is much more complex. The complexity arises from two sources:

1. An electrical potential develops at all interfaces. In soil, there are interfaces between solids and solution, solution and suspension, suspension and the electrode surface, and the reference electrode and all these interfaces.
2. The concept of activity is extremely important in soil. Protons or hydronium ions attracted to exchange sites or other components in the system will not be measured as part of the solution composition.

For these reasons, a standard method of measuring soil pH is chosen and all phenomena related to pH or involving pH is related to this "standardized" pH measurement. The most common method is to use a 1:1 ratio of soil to water, typically 10 mL of distilled water and 10 g of soil. In this method, the soil and water are mixed and allowed to stand for 10 min and the pH determined using a pH meter.

This method does not measure exchangeable protons attached to cation exchange sites; therefore, it is also common to use a salt solution (either KCl or $CaCl_2$) instead of distilled water in determining soil pH. The K^+ or Ca^{2+} in the solution exchanges with exchangeable hydronium, thus bringing it into solution where it can be measured. These procedures therefore usually give a pH that is less, more acidic, than that obtained using distilled water. The justification for this approach is that it is thought to more closely relate to the pH experienced by plant roots.

Many other methods of determining soil pH and exchangeable protons are described in the soils literature. Among these are a number of methods designed to determine a factor called the *soil buffer pH*, which is determined by adding a highly buffered solution to the soil and measuring the change in its pH. This change can then be related back to a property called the *total acidity* in soil. It is also used as the basis for making liming recommendations in agriculture. See Chapter 5, Section 5.3.1, for a detailed discussion of pH electrodes for measuring soil pH.

It is often tempting to design a new protocol or change an existing method for pH determination. Keep in mind that when or if this is done, it will change all the analytical methods that use pH as an important component such as the determination of CEC as discussed above. It will also change the interpretation of analytical results and the recommendations for applications of some agricultural amendments (see Chapter 10). Such a change is probably not advisable unless there is some highly significant improvement in the new method.

4.10. THE SOIL SOLUTION AROUND PARTICLES

Dissolved ions and molecules are evenly distributed throughout a pure solution. In soil, this does not take place because soil particles, particularly colloidal particles, both clay and carbon, have sorbed, exchangeable, and peripherally attracted components associated with them.

Cations attracted to colloid surfaces through their waters of hydration are outer-sphere species, whereas those that react directly with the oxygens present in the surface are inner-sphere species. Of the two, the latter species will be more strongly bonded and harder to extract.

Cations form a diffuse layer of ions called the *diffuse double layer* or the *electrical double layer* around soil particles as depicted in Figure 4.8. The existence of the diffuse double layer means that the ions are not evenly distributed throughout the solution; rather, they are more concentrated close to soil particle surfaces and less concentrated farther away. This phenomenon must be kept in mind, particularly when electrochemical analytical methods of analysis are developed [5,6].

4.11. DISTRIBUTION BETWEEN SOIL SOLIDS AND SOIL SOLUTION

All components in the soil solution are to a greater or lesser extent distributed unevenly between the solid and liquid phases. Anions are generally only weakly attracted to soil solids, if at all. Cations are attracted to the soil colloids, while the interaction between soil solids and organic compounds is complex, depending on the structure and functional groups of the organic compound and the nature of the soil solids. However, measuring the attraction or having a measure of the distribution between the two phases is extremely important in understanding the movement of material in soil. It will

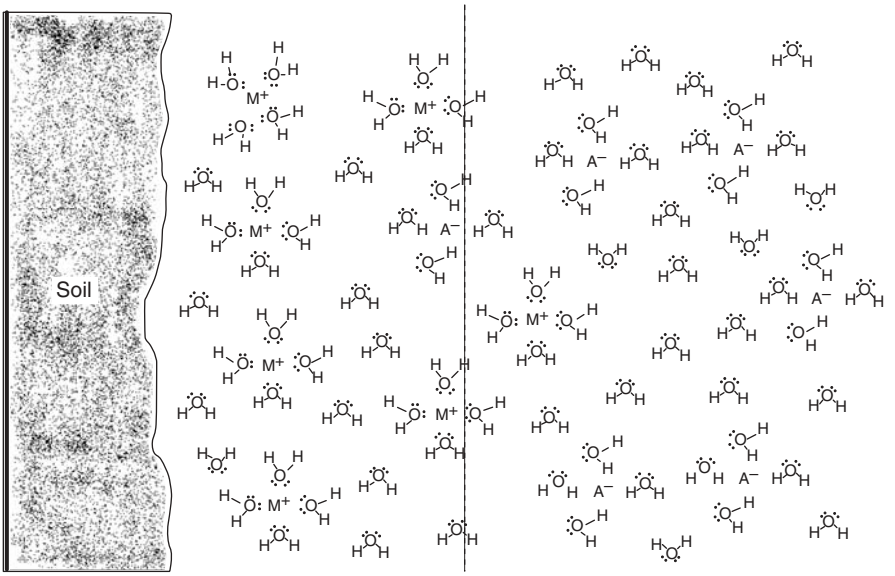

Figure 4.8. A soil particle with a diffuse layer of hydrated ions around it. The dashed line represents the boundary of the layer of tightly held cations. This diagram is not meant to be an exact representation of the diffuse double layer around a soil particle.

also give invaluable information about the time and amount of extractant needed for an extraction procedure.

Two types of distribution coefficients can be measured and used in describing the distribution between solid and liquid phases. The first and simplest is the distribution between total solid and liquid phases. This can be represented by K_d, as given by the following equations (where kg is kilogram and L is liter of soil solution):

$$K_d = \frac{\text{mg component}/\text{kg soil}}{\text{mg component}/\text{L solution}}$$
$$K_{om} = \frac{\text{mg component}/\text{kg organic matter}}{\text{mg component}/\text{L solution}} \qquad (4.2)$$

For K_{om}[2] applied to soil, the numerator would be kg organic matter in soil. Organic matter in soil has a much higher sorptive capacity than does the inorganic component and so it is sometimes more useful to describe the distribution between organic matter and a component of interest, particularly organic components. This can be done using the distribution coefficient K_{om}, which denotes the distribution between organic matter and water. The equation for this distribution is also given in equations (4.2).

[2] A K_{oc} where oc = organic carbon could be calculated if organic carbon is substituted in the equation.

These constants are determined by mixing a solution of known concentration, measured in mg/L, with a known amount of soil, measured in kilograms. After a period of time the solution and solid are separated and the amount of a given component in solution is measured. From this data a K_d can be calculated. This procedure is often performed for various periods of time and at various concentrations of a target compound in solution. In the determination of K_{om}, the amount of organic matter in the solid phase, namely, soil, is determined (see Chapter 3), and this amount is used as the kilogram value of organic matter.

From these two equations, the larger the amount of component sorbed to the solid phase, the larger the K_d or K_{om} and the less likely it is to move in the soil. Because K_d and K_{om} are determined using the compound in a pure solvent, usually water, and a soil suspension, these constants do not necessarly give information about how difficult the extraction of a component is likely to be when a specific extractant is used, such as mixed solvents or extractants using specific extracting or complexing agents [7,8].

4.12. OXIDATIVE AND REDUCTIVE REACTIONS IN THE SOIL SOLUTION

Many chemical reactions occurring in soils are acid–base reactions; however, oxidation–reduction reactions are more frequently the cause of confusion and may complicate the interpretation of analytical results. The reason is that reduced species, for instance, ferrous iron and the bivalent manganese, are seldom assumed to occur in the liquid and solid phases in a well-aerated soil, although they usually do.

During and after a rainfall the soil will quickly become anaerobic because microorganisms and plant roots use up dissolved and trapped oxygen. Under these conditions, reduction of various constituents takes place. When the soil drains or dries, air replaces the water and there is movement of oxygen back into the pores. However, not all pores drain immediately, and some pores never drain at all. Both these situations lead to anaerobic zones. Even when soils become aerobic, the reaction leading to oxidation of the reduced species may not be fast enough to remove all reduced species before the next anaerobic event.

Drying soil at an elevated temperature will result in the loss of water from these normally filled pores, thereby allowing reactions that would not otherwise take place. Also, chemical reactions take place faster at higher temperatures, resulting in reactions taking place much more rapidly than they normally would in soil. This not only changes the amount of compounds in the soil sample but will also change the ratios of the components present and may even lead to the formation of compounds not naturally present in soil at all. For these reasons soil is not dried at elevated temperatures before analysis [9].

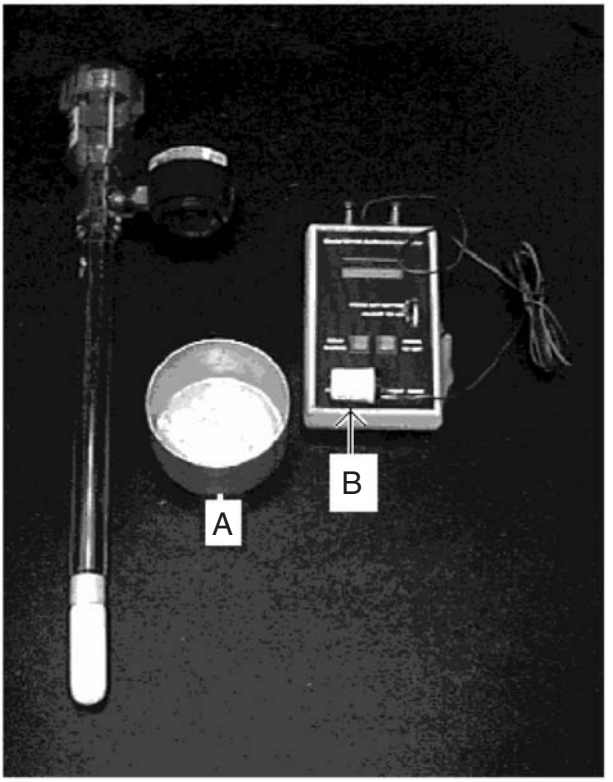

Figure 4.9. Shown from left to right are a tensiometer, soil drying can (*A*) and porous block (*B*) and meter for determination of soil water.

4.13. MEASURING SOIL WATER

Soil water can be measured using either laboratory or field methods. Common laboratory measurements include the percentage of water on a dry-weight basis and water content as a function of pressure. For the analyst the determination of percentage water on a dry-weight basis is the most important. Field methods include tensiometers, porous blocks (shown in Figure 4.9), psychrometers, time-domain relfectrometry (TDR), neutron probe, and others designed primarily to determine plant available water.

Soil water content may be reported in a number of different ways. Most commonly it is reported as the amount of water in grams per gram of oven dry soil, that is, the mass water content (see equation 4.3a below). It may also be reported as the volume of water per volume of soil, that is, the volumetric water content. For analytical purposes it is most often simply presented as a percent of water on a mass : mass ratio, that is, grams of water per gram of oven-dry soil. In field measurements, the water content of

soil is reported as the kilopascals (kPa) of pressure holding the water in the soil. The importance of reporting soil water in this fashion can be seen in Figure 4.2.

4.13.1. Laboratory Methods

The most important laboratory measure of soil water is the percentage water on a dry-weight basis. In most cases, soil will be extracted or analyzed while still moist to minimize changes that occur during drying. To obtain comparable data from multiple analyses, the soil sample weight is corrected using the percent water on a dry-weight basis. Because the water content of soil is highly variable, the dry weight is used, as it is more constant. Soil is typically dried at 105–110°C for 24 h in a drying cup as shown in Figure 4.9, and the amount of water lost is divided by the dry weight of the sample and multiplied by 100. The basic, simplified formula is given in equation (4.3a); formula representing the actual calculation usually made (for soil dried in a drying can; see also Figure 4.9) is given in equation (4.3b):

$$\%\text{water} = \frac{\text{soil wet weight} - \text{soil dry weight}}{\text{soil dry weight}} \times 100 \tag{4.3a}$$

$$= \frac{[(\text{soil wet weight} - \text{can weight}) - (\text{soil dry weight} - \text{can weight})]}{(\text{can weight} - \text{soil dry weight})} \times 100 \tag{4.3b}$$

Thus the can weight must be subtracted to carry out the calculation.

The percentage of water on a dry-weight basis is used to calculate the dry weight of soil taken for an analysis. First, a 50-g sample of soil is taken and dried. A second 25-g sample is taken and analyzed. The dried sample is found to weigh 48 g and thus lost 2.0 g of water; thus:

$$\%\text{water} = \frac{2}{48} \times 100 = 4.2\% \tag{4.4a}$$

The water in the moist sample, taken for analysis, is found using this percentage:

$$\frac{4.2}{100} = 0.042 \tag{4.4b}$$

$$\text{Dry-weight sample analyzed} = \frac{\text{weight of sample taken}}{1 + 0.042} = \frac{25}{1.042} = 24\text{ g} \tag{4.4c}$$

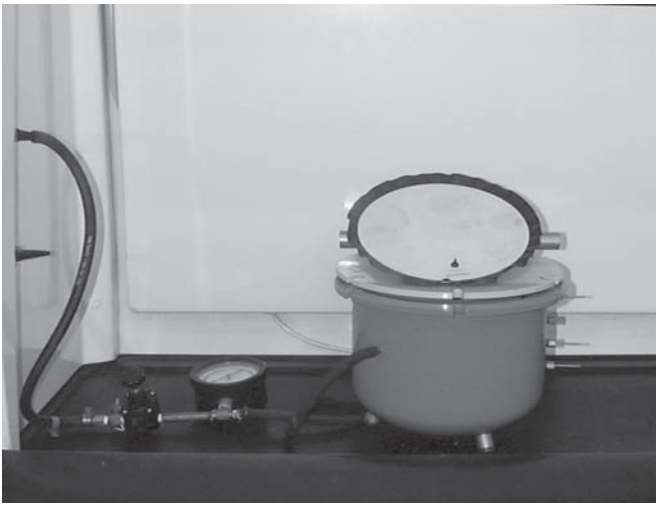

Figure 4.10. Pressure apparatus for measuring the amount of water held in soil at different pressure; pressure gauge is to the left, and a pressure plate is on top of the vessel.

Table 4.1. Common Instruments for Measuring Soil Water Content in the Field

Instrument	Useful kPa Range	Characteristics
Tensiometers	0 to –86	Limited range and depth
Thermocouple psychrometers	50 to –10,000	Wide range but limited accuracy
Porous blocks	–100 to –1500	Accuracy and range are limited
Neutron probe	0 to –1500	Cannot be used in highly organic soils
Time-domain reflectrometry (TDR)	0 to –10,000	Accurate and can be installed at various depths

The other common laboratory method uses pressure plates and membranes to measure the amount of water held by soil at various pressures (Figure 4.10). One advantage of this method is that it gives a pressure value that can be used in many calculations relevant to movement of water in soil. The plates in the apparatus shown are used in the pressure range of –10 to –30 kPa. Other similar pressure plate apparatus can be used to determine water at pressures to –1500 kPa [10].

4.13.2. Field Methods

Field methods of measuring soil water are designed primarily to measure water, in the range of –10 to –1500 kPa of pressure. However, different instruments have different ranges as shown in Table 4.1. Tensiometers, porous

blocks, and thermocouple psychrometers are usually installed in the field and measurements are taken on a regular basis. Neutron probe and time-domain reflectrometry (TDR) equipment are usually carried to the field each time a measurement is made. In addition, the neutron probe requires an access hole into which it is lowered to determine the of water content. These methods provide data that are seldom generally used in soil analysis and will not be discussed further [11–16]. Additional information about these methods can be found in the text by Brady and Weil [17].

4.14. CONCLUSION

The compounds contained in soil air are basically the same as those in atmospheric air, but are more variable. Also, the volume of soil occupied by air varies greatly. Water is a unique molecule in both its physical and chemical characteristics. It has higher than expected boiling and melting points and can dissolve a great variety of compounds. In the soil it is even more unique in that it occurs in the liquid, gaseous, and solid (frozen) states. The water content of soil is highly variable, ranging from air dry, with as little as 1% water to saturated where all void spaces are filled with water. The soil solution contains many inorganic and organic compounds ions and gases, the concentration of which changes dramatically when soil water content increases or decreases.

In the laboratory soil water content is measured by drying in the oven and with pressure plate apparatus. A number of different field measuring methods are used mostly to determine the amount of water available for plant use. Drying soil can change the form and species of components present, and for this reason, most soils are dried carefully at or only slightly above room temperature before analysis.

PROBLEMS

4.1. Describe the differences between atmospheric air and soil air.

4.2. Soil water can be thought of as existing as layers around soil particles. Explain how these layers are differentiated.

4.3. Explain how pores can affect the composition of both the soil atmosphere and the soil solution.

4.4. Using equations, illustrate how soil carbon dioxide affects both the pH of soil and the ions present in the soil solution.

4.5. Give both general and specific examples of how organic compounds can lead to the formation of cation exchange sites in soil.

4.6. What role would K_d play in the extraction of a component from soil?

4.7. Using any available resources, describe how pressure place methods of determining soil moisture levels work.

4.8. Explain why tensionmeters and porous block methods are most useful in the field.

4.9. A soil sample is taken from a field. Half the sample, 50 g, is dried at 105°C for 24 h, after which time it is found to weigh 45 g. What is the percent moisture of this soil sample on a dry-weight basis?

4.10. The undried soil sample in Problem 4.9 is extracted, analyzed for phosphate, and found to contain 5 µg of phosphate. What is the concentration of phosphate on a dry-weight basis?

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CHAPTER

5

ELECTRICAL MEASUREMENTS

In making measurements using electricity, all its parameters—voltage, amperage, resistance, capacitance, frequency, and dielectric characteristics—can be used singly or in combination to obtain information about various conditions in the medium through which electrons are moving. All soils contain many ions of many different sizes and complexities: simpler hydrated K^+, more complex NO_3^-, organic ions, and charged solids. All contribute to the electrical characteristics of a soil and its solution.

One easily demonstrated electrical characteristic of moist soil is seen in the production of electricity when two different metals, namely, copper and zinc, are inserted into it (assuming that the reaction is spontaneous). This is not unexpected because any salt containing solution, adsorbed in media such as paper or cloth, and placed between these same two electrodes, will cause a spontaneous reaction, which will produce electricity. The source of this flow of electrons is a chemical, oxidation–reduction reaction, represented as the following two half-reactions, for copper and zinc:

$$\begin{aligned} &\text{Oxidation:} \quad Zn^0 \rightarrow Zn^{2+} + 2e^- \\ &\text{Reduction:} \quad Cu^{2+} 2e^- \rightarrow Cu^0 \end{aligned} \tag{5.1}$$

Production of electricity in this way was the original method used to produce electricity (i.e., an electric current) for scientific experiments. Thus, if copper and zinc are inserted into wet soil, electricity will be produced by the same process, although a mixture of salts will be involved. Figure 5.1 shows copper and zinc strips inserted into a moist soil and connected to a voltmeter, which displays the resulting voltage, demonstrating the existence of salts in the soil. This simple setup is not, however, used to measure soil characteristics such as salt content or pH.

This electricity generation characteristic, along with the salts, their movement through soil, and the diffuse double layer must be kept in mind when making any soil measurement using electricity or electrodes.

Although all characteristics of electricity have been used to investigate soil and its properties, only a limited number are used routinely. The most common

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

Figure 5.1. Zinc and copper strips inserted into wet soil and producing a voltage as shown on the voltmeter.

are those used for the determination of pH, salt content, and soil water content. Of these three, pH is the most common measurement, and frequently the first measurement made prior to all other determinations. Although pH can be determined by many methods, for soil the most common is to use a pH meter and electrode. Conductivity or resistance is used to measure soil salt content, while several different electrical characteristics of soil are used in determining its water content. In addition to their inherent importance, the pH, salt, and water content are important in determining how other analysis must be carried out or in determining the effects or interferences that these parameters have on other analytical methods, particularly spectroscopy and chromatography.

5.1. THE BASIC ELECTROCHEMICAL CELL

The terms *cathode* and *anode* often cause confusion and to a certain extent the characteristics of an electrode depend on whether they are being viewed from the outside in or the inside out. One way to keep the terms straight is that *anode* begins with a vowel, as does *electron*; that *cathode* begins with a *consonant*, as does cation; or that both cathode and cation begin with a *c*.

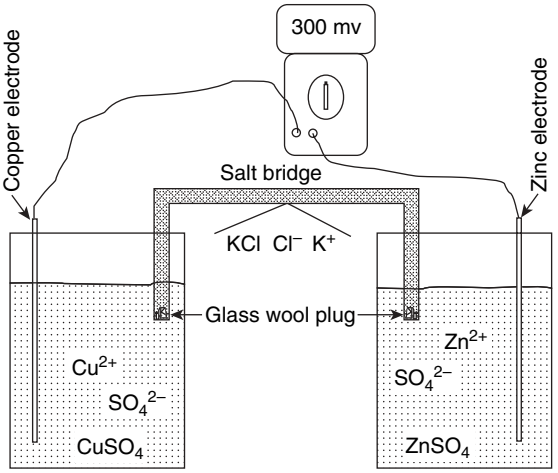

Figure 5.2. A typical electrochemical cell.

A basic electrochemical cell (depicted in Figure 5.2) consists of a copper wire in one container with a solution of copper sulfate and a zinc rod in a different container with a zinc sulfate solution. There is a salt "bridge" containing a stationary saturated KCl solution between the two containers. Ions flow freely in the salt bridge in order to maintain electrical neutrality. To complete the cell, a wire is connected to each rod and then to a measuring device such as a voltmeter.

5.2. ELECTRICITY GENERATION IN SOIL

The generation of small electrical currents in soil is possible and may affect any electrical measurement made therein. More recent theoretical and experimental work has shown that charged particles, including ions, passing through microchannels under low pressure (i.e., 30 cm of water), can generate small amperages. In addition, amperages from multiple channels are additive and might be a useful method of generating electricity or of making a new type of battery [1,2].

Soil solids have channels having charged surfaces, such as those in clays, and what is termed an *electrical double layer* on the surfaces and salts in the soil solution moving over and through them. The generation of electricity in soil by this mechanism is thus possible. Although generation of electrical currents under standard extraction conditions may not be of concern, they may be in other soil analytical procedures. In situ measurements, taken with electrodes buried in soil, may record or be susceptible to interference from such currents. This will be particularly true for electrodes buried in fields, during

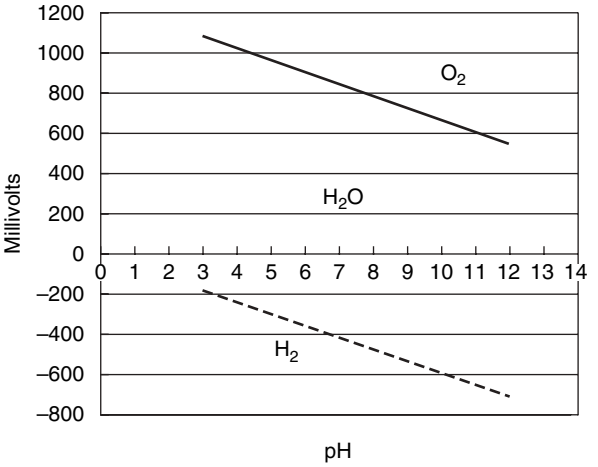

Figure 5.3. Electrode potentials for the oxidation of oxygen and the reduction of hydrogen at gaseous pressure of 1 atm and pH levels common in soil.

rainfall or irrigation events and the subsequent percolation of water through the soil profile. It will be more pronounced in soils, such as *Aridisols*, which naturally contain more salts and soil with more clays, that is, with Bt horizons, particularly for high-activity clays, with high cation exchange capacity and high surface area.

5.3. POTENTIOMETRY (ELECTRODES IN SOIL MEASUREMENTS)

Numerous different types of electrodes are used in electrochemical analysis of soil. Simple elemental electrodes such as platinum, mercury, and carbon are the most frequently used, while other unreactive metals such as gold and silver and the more reactive copper and zinc and others have been used. During analysis both stirred and unstirred solutions and suspensions are used. In some cases electrodes are rotated or, in the case of mercury, dropped into the solution being analyzed.

Soil scientists represent the electrode potential as Eh and the standard potential as Eh^0,[1] which is measured in millivolts or volts. All (non-oven-dried) soil contains water, which limits the possible upper and lower potentials. The upper potential is controlled by the oxidation of water. Under highly oxidizing conditions oxygen would be oxidized to oxygen gas, namely, O_2. The lower potential is limited by the reduction of hydrogen or protons, specifically, H^+, which would lead to the formation of hydrogen gas. Figure 5.3 shows the elec-

[1] The standard electrode potential is that potential generated by an electrode when compared to a standard hydrogen electrode under standard conditions, which typically include a temperature of 298 K (degrees Kelvin).

trode potentials for the oxidation of oxygen and the reduction of hydrogen at common soil pH levels.

Although the reduction of hydrogen and the oxidation of oxygen determine the lower and upper limits of electrochemical redox measurements in soil, it is still possible to have both hydrogen and oxygen produced by biological processes. Photosynthetic production of oxygen by algae and phototrophic bacteria occurs in all soils. Likewise, the biological production of hydrogen gas occurs under anaerobic conditions. Thus it is possible to find oxygen and/or hydrogen gas being produced in soil.

In soil analysis, pH, selective ion, oxidation–reduction (redox), electrical conductivity cells, and oxygen electrodes are commonly used. For each of these measurements a different specific electrode along with a separate or integral reference electrode will be needed. In some cases, with extended use or long exposure to soil or soil–water suspensions, electrodes may become polarized. When this happens, erroneous results will be obtained and depolarization will need to be carried out using the electrode manufacturers' directions [3].

5.3.1. pH

In soil one can conceive of the presence of three "types" of protons (see Chapter 6, p. 116). However, those in solution, associated with water molecules forming hydronium ions, are the only measurable protons. Other protons will be associated with cation exchange sites, are exchangeable, and contribute to soil buffering. They cannot be measured directly but can be exchanged with cations from salts or buffer solutions, and once in solution they can be measured. The third "type" of proton is bonded to either inorganic or organic soil components and normally will be regarded as being covalently bonded. However, they may be part of a functional group from which they may be easily removed and thus become part of the protons in solution. Both organic acid and phenolic groups are examples of compounds, which have protons, that fall into this category.

The absolute pH of soil cannot be known absolutely; however, standardized methods for measuring soil pH have been developed.

A combination pH electrode, as illustrated in Figure 5.4 and shown in Figure 5.5 (*A*), is most commonly used in determining soil pH. However, two separate electrodes, one pH sensing (i.e., the H^+ glass bulb in Figure 5.5) and the other a reference electrode, may be used and may be best in cases where fouling of the reference electrode is a particular problem. Care must always be taken to avoid scratching or breaking the pH sensing bulb when making a pH measurement, because while some pH electrodes have robust pH sensing electrodes, others are quite delicate. Figure 5.6 shows a pH electrode in a soil suspension, which is connected through a card, which is a pH meter, to a laptop computer that provides data output.

Electrodes may be attached to a pH meter, which can be analog, digital, or, as mentioned above, to a computer. Several different connectors are used

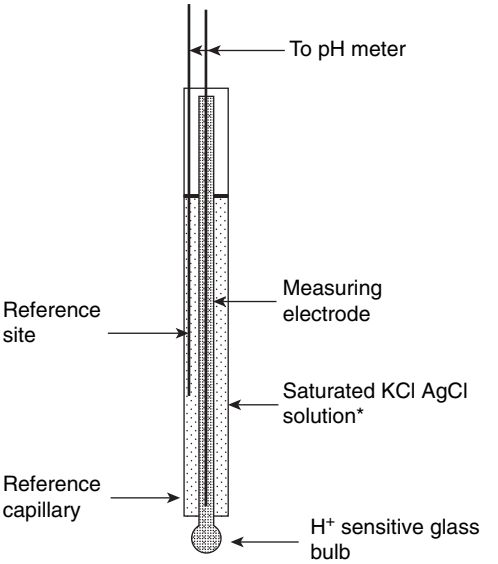

Figure 5.4. Typical setup for a combination pH electrode. Note that the saturated KCl/AgCl solution can be different depending on the electrode manufacturer.

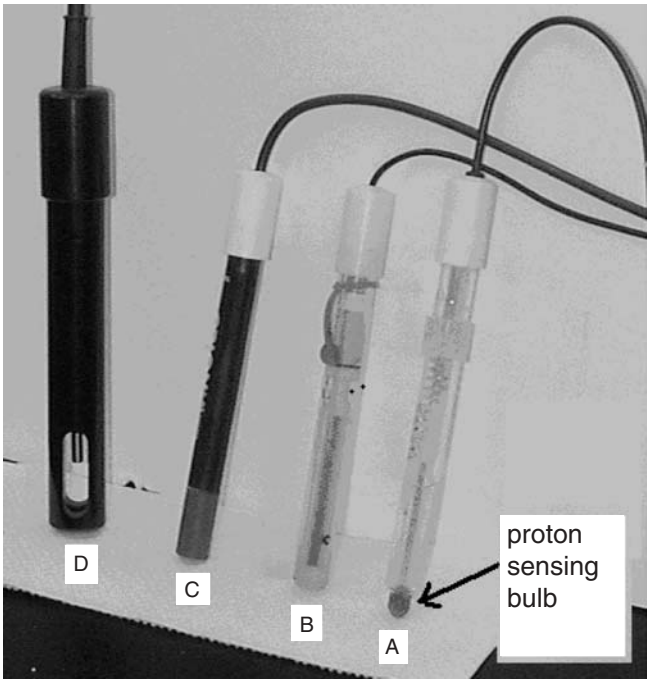

Figure 5.5. A combination pH electrode is shown (*A*). The dark-colored tip is the pH sensing glass electrode. The reference electrode is not visible. A reference electrode (*B*) for ISE measurement along with a nitrate-selective electrode (*C*) are also shown. Electrode *D* is used for measuring salt content in aqueous solutions.

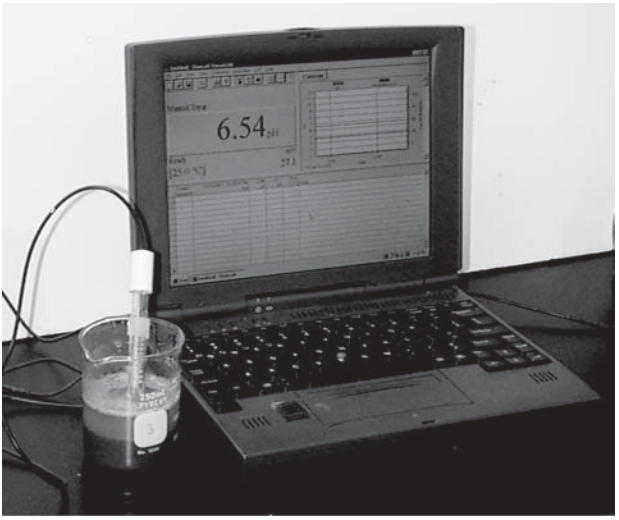

Figure 5.6. A pH electrode in a soil suspension. The electrode is connected to a computer that shows the readout.

on both electrodes and pH meters; thus, when purchasing pH and reference electrodes, one should ensure that they have the correct connectors for the meter being used. Even robust electrodes must be treated with care and kept wet with the appropriate solution as directed by the manufacturer. Regardless of the make, pH meters are usually robust and should need little maintenance, although care should be taken to keep them dry.

Although maintenance of pH meters is low, standardization is essential and must be done on a regular bases. The starting point in standardization is to adjust the meter and electrodes using a standard buffer solution of pH 7.00 and adjusting the meter to this pH. The second step is to set a second point, which is in the range of pH levels expected to occur in the measurements. Thus, if the pH values are all expected to be acidic, the second point will commonly be set using a buffer of 4.02; if they are expected to be basic, then a pH 10.00 buffer will be used. If both acidic and basic pH values are likely, the meter can be standardized using all three buffers.

Standard buffers can be purchased as already prepared solutions or as powders dissolved in distilled or deionized water. In this latter case the powder is typically dissolved in 100 mL of distilled or deionized water for use. Buffers may be color-coded, such as green for pH 7.00, orange for pH 4.02, and blue for pH 10.00.

In practice the electrode is first rinsed with distilled water and placed in the pH 7.00 buffer and the meter adjusted to read 7. In some cases the meter will automatically make the adjustment needed. The electrode(s) is (are) rinsed and placed in the second buffer and the meter adjusted. This two-point standardization is usually enough for most soil pH measurements; however, if it is not, the third buffer can be used in the same way.

Standardization is an essential component of soil pH measurement, and care must be taken to ensure good buffers. If the buffers show any indication of contamination, such as material floating in them, soil, or microbial growth, they must be discarded and new buffers prepared. All pH buffers will support microbial growth, which will interfere with electrode function and change the buffers pH.

Measurements made using pH meters and electrodes are temperature-sensitive; that is, the pH reading obtained depends on the temperature of the solution. Some pH meters have a temperature probe such that a temperature correction is automatically made during the measurement. However, if this is not the case, the pH meter must be set to the proper temperature if accurate measurements are to be obtained.

Fouling of the reference electrode or the reference side of a combination electrode is a common problem in soil pH measurements. Fouling can be caused by salts, organic matter, and clay. Each electrode manufacturer will provide specific cleaning procedures that will help keep electrodes functioning properly; however, in many cases no amount of cleaning will be effective and the electrodes will need to be replaced.

Depending on the extraction method to be used or developed, determination of pH using a standard method should be used. In different countries and geographic areas different standard methods will be in common use; for example, in Ohio (USA), a 1:1 soil:water suspension is used while in Zimbabwe a 1:1 soil:0.01 M $CaCl_2$ is commonly used. The pHs determined by the standard method, used in a particular area, are used in many other procedures and methods and are used to make recommendations and predictions about the environment. Changing the method will mean that the validity of all these relationships will need to be reestablished or new relationships determined. For these reasons it is generally not advantageous to spend time and money developing a new methodology for determining soil pH unless there is a highly significant economic benefit in doing so [4–6].

5.3.2. Ion-Selective Electrodes

Ion measurements, using ion-selective electrodes (ISEs),[2] are very similar to pH measurements and typically are carried out using a pH meter capable of accepting ion-selective electrodes. Each ion requires a specific electrode, some of which will be combination electrodes similar to combination pH electrodes, while others will be only the sensing electrode, called a *half-cell*. For these half-cell electrodes, special reference electrodes, which have a high flow of reference electrode solution, are used. There are two important differences between pH and ion-selective measurements: (1) the latter may require that the solution be separated from the soil before measurement is made and (2)

[2] Ion-selective electrodes were originally called *ion-specific electrodes*, and this term may still be encountered.

the reference electrode and meter must be specially designed for use with this type of electrode. Both ion-selective and the reference electrodes used with them are shown in Figure 5.5, letters *C* and *B*, respectively. Table 5.1 gives common ion-selective electrodes useful in soil analysis. This is not an exhaustive, list and new selective electrodes are being developed on a daily basis.

Ion-selective electrodes are standardized using standard solutions of the ion dissolved in water or a solution designed to keep all samples at about the same ionic strength. Standardizing solutions can be purchased or prepared in the laboratory and typically, as seen in Table 5.1, cover several orders of magnitude often between 1 and 10^{-6} or 10^{-7} molar. Measurements are made at the various concentrations and a standard or calibration curve prepared (see Chapter 8, Sections 8.8.2, 8.9, and 8.10). Usually the meter can be programed to read the concentration of the ion directly once a suitable curve is obtained. Raw data can also be entered into a spreadsheet, which can be programmed to calculate the amounts of ion present in any units desired.

As with pH measurements, a specific amount of water or ionic strength adjusting solution is added to soil mixed and allowed to stand. The ion-selective and reference electrodes are then inserted in either the suspension or solution, filtered from the soil before measuring. When a stable reading is obtained, it is recorded.

Ion-selective electrodes are subject to interference from ions other than the one they are designed to measure. The Na^+ ion-selective electrode is susceptible to interference from other single positive species (i.e., K^+, NH_4^+), and the same situation will hold for ion-selective electrodes designed to measure negatively charged species (see Table 5.1). Generally the electrode will be less sensitive to these interfering ions than to the ion it is designed to measure so that low levels of interfering ions may not make a significant difference in the measurements being made. In other cases interfering ions can be precipitated or complexed to remove them from solution before measurement is made.

Interfering ions are problematic when analyzing environmental samples, particularly soil and soil extracts. Samples of these materials may contain unknown combinations and concentrations of ions. For these reasons ion-selective electrodes are most useful in two situations. The first is where the soil composition with regard to ions is well known and routine repetitive analysis is to be made. The second is where a preliminary screening is to be done and followed up by detailed laboratory analysis. In this latter case potential interfering ions will be determined and the validity of the original screening accessed [7,8].

5.3.3. Redox

Redox is an abbreviation for reduction and oxidation. It is based on the fact that one component cannot be oxidized without another being reduced. The most common oxidizing, electron accepting, agent is oxygen. In soil other electron accepting agents such as ferric iron and nitrate can also serve as

Table 5.1. Ion-Selective Electrodes and Their Characteristics

Electrode/Ion[a]	Type	Molar Range[a]	Interferences[a]
Ammonia	Gas sensing/combination	$1.0–5 \times 10^{-7}$	Volatile amines, Hg^{+}
Bromide	Solid state	$1.0–5 \times 10^{-5}$	S^{2-} must not be present; CN^{-}, I^{-}, NH_3, Cl^{-}, HO^{-}
Cadmium	Solid state/combination	$10^{-1}–10^{-7}$	Pb^{2+}, Hg^{2+}, Cu^{2+}
Calcium	Half-cell	$1.0–5 \times 10^{-7}$	Na^{+}, Pb^{2+}, Fe^{2+}, Cu^{2+}, H^{+}
Carbon dioxide	Gas sensing	$3 \times 10^{-2}–10^{-5}$	Volatile organic acids
Lead	Solid state/half-cell	$0.1–10^{-6}$	Cu^{2+}, Ag^{+}, Fe^{3+}, Cd^{2+}, Ag^{+}
Nitrate	Half-cell	$1.0–7 \times 10^{-6}$	ClO_4^{-}, I^{-}, ClO_3^{-}, CN^{-}, Br^{-}, NO_2^{-}, HS^{-}, CO_3^{2-}, HCO_3^{-}, Cl^{-}
Nitrogen oxide	Gas sensing/combination	$5 \times 10^{-3}–3.6 \times 10^{-6}$	CO_2, volatile weak acids
Oxygen	Gas sensing/combination	0–20 ppm	—
Perchlorate	Half-cell	$1.0–7 \times 10^{-6}$	I^{-}, NO_3^{-}, ClO_3^{-}, NO_2^{-}, CN^{-}, NO_2^{-}, HCO_3^{-}, CO_3^{2-}, Cl^{-}
Potassium	Half-cell	$1.0–10^{-6}$	Cs^{+}, Na^{+}, NH_4^{+}, H^{+}
Redox/ORP	Combination	—	—
Sodium	Half-cell	Saturated to 10^{-6}	Ag^{+}, K^{+}, H^{+}, Li^{+}, Cs^{+}, Tl^{+}

[a] This is not an exhaustive list of ISE electrodes or number of interferences. Also, manufacturers' specifications must be consulted as different materials and construction may result in different interferences and sensitivities.

oxidizing, electron accepting, agents. Likewise, there are a number of reducing, electron donating agents; chief among these is hydrogen. Generally speaking, in soil reduced species are more soluble and more easily removed from the soil. For this reason the oxidation–reduction condition of a soil sample can be important in any extraction or analytical procedure.

In accessing the redox potential it is also essential to know the pH of the medium being analyzed. There is a direct relationship between pH and the potential measured:

$$E = -59.16\,\text{mV} \times \text{pH}$$

For each unit of increase in pH (decrease in H^+), there is a decrease in millivoltage. In soil, this simple equation does not hold because it does not take into account the complex electrical characteristics of soil and thus cannot be used in soil analysis. However, it does illustrate the fact that any measurement or consideration of the oxidation–reduction situation in soil must also take into account the soil's pH.

In soil stable redox reactions occur between the limits of the oxidation and reduction of water as shown in Figure 5.3. The *y* axis (ordinate) shows millivolts (mV); the *x* axis (ordinate), pH. Chemists and physical chemists will use these two terms in describing what happens in a redox reaction. Environmental and soil chemists will refer to the measured voltage [expressed in millivolts (mV)] as Eh; thus these types of graphs are called Eh–pH diagrams. Eh is defined by the Nernst equation [equation (5.3a)], which can be simplified to equation (5.3b) when the activities of (Red) and (Ox) are equal. Many redox couples have the relationship shown in equation (5.3c), and thus their reactions as electron acceptors or donors is straightforward (however, in soil significant variations from this simple relationship exist where $m \neq n$):

$$\text{Eh} = \text{Eh}^0 - \frac{RT}{nF} \ln \frac{(\text{Red})}{(\text{Ox})^n (\text{H}^+)^m} \tag{5.3a}$$

$$\text{Eh} = \text{Eh}^0 - \frac{m}{n} 0.059\text{pH} \tag{5.3b}$$

$$\frac{m}{n} = \frac{\text{H}^+}{\text{e}^-} = 1 \tag{5.3c}$$

where Eh^0 = standard electrode potential
R = gas constant
T = absolute temperature
n = number of electrons
F = Faraday constant
(Red),(Ox),(H^+) = concentrations of reduced, oxidized species, and hydrogen ion, respectively

In addition to the simple Eh–pH graph shown in Figure 5.3, three-dimensional Eh–pH graphs can be produced. Known quantities of a pollutant can be added to a number of different soil suspensions and its degradation at different combinations of Eh and pH measured. In this way the optimum conditions for the decomposition of the pollutant in question can be determined. An excellent example of this is the decomposition of pentachlorophenol and hexahydro-1,3,5-trinitro-1,3,5-triazine in soil and water under various Eh–pH conditions as illustrated in the papers by Petrie et al. and Singh et al. [9,10].

In soil analyses knowledge of Eh–pH data can be used in three ways. It will provide information as to the form or species of pollutant present (see also Chapter 10). It can also be used to determine which extraction procedure is best suited for extraction of a component from a soil sample. Potential changes in species, movement in the environment, and conditions suitable for bioremediation or natural attenuation can also be derived from this type of measurement.

In this discussion so far all the systems are well defined, at equilibrium and at a constant 25°C. None of these conditions occur in soil in the environment. Soil is not a pure system and all the components affecting redox reactions are seldom known, defined, or understood, and a host of different redox couples, many unknown, are likely to be present. Unless it is possible to take into account all couples present, it is not possible to describe the exact redox conditions in a soil without measuring it.

Even though very small soil samples may be well defined, large samples and field size areas are not. Soil is never at equilibrium even though it may appear to be so over short periods of time. Reactions occur, and microorganisms continue to function in soil samples after they are taken. In tropical conditions soil temperatures will vary significantly even where there is little change in air temperature. Heating by sunlight and cooling by radiation will always occur. Variable shading by trees and other vegetation will add to soil temperature variability, and both rain and its subsequent evaporation will have a cooling effect on soil.

In spite of the limitations, Eh–pH data will provide information about the condition of a soil in terms of it being in an oxidizing or reducing condition. Thus it will indicate the prominent redox conditions of species present. It will also indicate what changes in Eh or pH may be desirable to effect the desired extraction or analysis for the compound or species of greatest concern [9–11].

5.3.4. Gas Electrode

The gas electrode is similar to the ISE electrodes and usually works on the same basic principles. Here the electrode looks much like a standard ISE electrode and is, except that it has a gas-permeable, water-impermeable membrane in the tip. Gas present in the environment passes through the membrane and reacts with reagents in the interior, producing a chemical change that is directly related to the development of a potential. This potential is thus

directly proportional to the partial pressure of the gas to which the electrode is exposed.

Electrodes are available for most common gases, including oxygen, carbon dioxide, and ammonia. For oxygen and carbon dioxide their natural concentration in air can be used in standardization while other gases require standard gas concentrations. Because of the importance of oxygen in biological processes, a number of different types of oxygen sensing electrodes have been developed. Some may be compatible with pH meters; others will not. It is thus important to make sure that the correct electrode is obtained for the instrument to be used.

There are three concerns in using gas sensing electrodes in soil: (1) some electrodes have membranes that have a limited shelf life and must be changed regularly; (2) the membrane is relatively delicate, and so electrodes must be placed in soil carefully and cannot be subject to movement; and (3) many membranes must be kept moist to function properly and thus cannot be used in dry soils or situations where the soil may dry out during measurement [7].

5.4. VOLTAMMETRY

Voltammetry is the oxidation or reduction of a species at an electrode. In this case the electrode is the source or the sink for the electrons being exchanged during the reaction. By measuring current (amperage) and potential (voltage) in a system, either stirred or unstirred, one can obtain information about not only the species present but also the amount present. In a typical experiment, only the oxidized form of a component might be present and the negative potential might gradually increase until a spike is observed. This is the potential at which the oxidized species is being reduced. Because each oxidized species has a different potential where it is reduced, this spike can be used to identify the species. Numerous analytical techniques are based on this electrochemical phenomenon. Some common examples are stripping voltammetry, cyclic voltammetry, and polarography.

The standard potentials of practically all oxidation and reduction reactions, especially those common in the environment and soil, are known or can easily be determined. Because of the specificity and relative ease of conducting voltammetric measurements, they might seem well suited to soil analysis. There is only one major flaw in the determination of soil constituents by voltammetric analysis, and that is that in any soil or soil extract there is a vast array of different oxidation–reduction reactions possible, and separating them is difficult. Also, it is not possible to begin an investigation with the assumption or knowledge that all the species of interest will be either oxidized or reduced.

In a well aerated soil it is expected that all species will be in their highest oxidation states; however, this does not happen for reasons elucidated in previous chapters. In a well-aerated soil, both ferrous and ferric iron can exist

along with elemental iron.[3] Zinc, copper, and especially manganese can apparently exist in a mixture of oxidation states simultaneously in soil. Add to this a multitude of organic species that are also capable of oxidation–reduction reactions, and the result is truly a complex voltammetric system [12,13].

5.5. ELECTRICAL CONDUCTIVITY

The electrical conductivity (EC) of soil or water depends on the amount of salts present. As shown in Figure 5.1, salts are always present in soil. In humid regions they are at low concentration and do not affect plant growth, while in semiarid and arid regions and near salt lakes or oceans they may be at high concentration and have detrimental effects on plant growth. In all these cases the electrical conductivity is simply measured by determining the amount of electricity passing through a cell of known dimensions and configuration when it contains a salt solution. In the case of soil, the electrical conductivity of a soil paste or solution extracted from soil is related to the soil's salt content.

5.5.1. Whole-Soil Paste

Direct determination of the conductivity of a soil is carried out by making a paste and placing it in a special, standard, cell containing two electrodes. The cell is made of a circular nonconductor with flat-strip electrodes on opposite sides of the cell. A soil paste in water is prepared and added to the cell; the top is leveled and the electrodes attached to a meter that measures its resistance, which is then related back to the soil's salt content.

5.5.2. Water and Soil Extracts

Electrical conductivity is usually determined on solutions of salt in water. A soil sample is mixed with water until a paste, which is allowed to stand overnight, is obtained. After standing the paste is filtered, and the EC of the solution obtained is determined. A conductivity cell for water, shown in Figure 5.5 (*D*), a sample is simply placed in the cell, or the cell is inserted into the water and a measurement made. Standardization is carried out by preparing standard solutions of salt, usually sodium chloride (NaCl) in distilled water and the electrical conductivity determined.

The basic unit used to represent in electrical conductivity is siemens (S). For direct current it is the reciprocal of the resistance, and for alternating current it is the reciprocal of the impedance (both in ohms). For soils dS/m are the units used where 0.1 S/m = 1 dS/m.[4]

[3] The occurrence of elemental iron in soil is not common but is possible where there is contamination, for example, from an accident.

[4] From *Encyclopaedia Britanica Deluxe Edition* 2004 CD-ROM.

Soils with pH <8.5 and an electrical conductivity of <4 dS/m are considered normal. Soils in the same pH range but with EC >4 are said to be *saline*. Soils high in sodium but with EC <4 and are called *sodic soils*, while those with EC >4 and high sodium are saline–sodic soils. Sodium is represented as either the ratio of exchangeable sodium to total exchangeable cations, termed the *exchangeable sodium percentage* or the *sodium absorption ratio*, which is the ratio of sodium ion to the square root of the calcium and magnesium ions [2,14–17].

5.6. TIME-DOMAIN REFLECTOMETRY (TDR)

Time-domain reflectometry involves the use of two or more substantial metal rods inserted into soil. The rods are parallel and are attached to a signal generator that sends an electrical input down the rods. The time it takes the signal to travel down the rods is dependent on the soil's apparent dielectric constant, which in turn is proportional to the amount of water in the soil. On reaching the end of the rods, the signal is dissipated and the amount of dissipation is related to the amount of salt in the soil. This instrument can thus measure both the water content and the salt content of the soil into which it is inserted.

Depending on the type of data needed and the experiment to be done, rods can be either moved from place to place or left in place for measurements made over a period of time [18].

5.7. POROUS BLOCK

The porous block is used to determine the water content in soil by changes in the resistance between two electrodes encased in a porous material buried in soil. In a common porous block design, two electrodes are encased in gypsum connected to wires such that the blocks can be buried to various depths in a soil profile. The gypsum block is buried in soil, and its moisture content comes to equilibrium with the soil water content. As the water content of the block increases, the resistance between the two electrodes decreases and vice versa. The change in resistance can then be related back to the soil water content. This basic idea is used with "blocks" constructed of materials other than gypsum. The gypsum block is designed for fieldwork and finds it greatest use there (see Figure 4.9) [18].

5.8. OTHER METHODS

In addition to the porous block, there are a number of other types of measurement made in soil involving electrodes that are not in direct contact with the soil. An example is the thermocouple psychrometer, which involves a Thomson thermocouple in a ceramic cell buried in soil. The thermocouple

cools when a current is passed through it, causing water to condense on the thermocouple. When the electricity is turned off, the condensate evaporates at a rate inversely proportional to the relative humidity in the soil. A voltage generated by the cooling junction is measured and related to the soil moisture content. This moisture content is related to both the matrix and osmotic potentials of the soil being investigated.

There are many additional methods and variations on the methods discussed above (see Section 4.13). Most are designed for determination of soil water content in the field and are rarely used in laboratory analysis of soil components such as available plant nutrients or contaminants [18–20].

5.9. CONCLUSIONS

Soil has electrical characteristics associated with its components, salts, ions in solution, and the diffuse double layer. All, singly or in combination, can affect electrical measurements in soil. Electrodes inserted into soil are used to measure various soil characteristics, most often soil pH, salt, and water content. Fouling of electrodes by salts, organic matter, or inorganic components, including clay, is an important potential source of error in any soil measurement involving electrodes. Because of the potential errors, electrodes must be standardized frequently during procedures that involve multiple measurements over an extended period of time. Analytical procedures for the determination of soil characteristics using electrodes have been developed and are used in conjunction with other soil procedures and measurements. Development of a new method or procedure will require detailed investigation of the relationship of this new method to previously developed methods and to associated or dependent procedures or measurements.

PROBLEMS

5.1. Diagram a basic electrochemical cell. Diagram a similar cell using soil instead of water as the supporting medium.

5.2. What do the terms Eh and Eh^0 stand for? What types of electrodes are used for the determination of Eh in soil?

5.3. Describe the basic design of a pH electrode. What "kinds" of protons in soil can a pH electrode can measure?

5.4. Diagram an ISE electrode. What characteristics of ISE electrodes make them difficult to use for direct soil measurements?

5.5. In terms of Eh–pH, what limits the range of Eh–pH values in soil?

5.6. Explain, giving examples, why voltammetry is seldom useful in direct soil measurement of components present.

5.7. Describe the two common methods of determining salt content in soil using electrical conductivity.

5.8. Soil water content can be measured in the field using a number different types and arrangements of electrodes. Describe two of these methods in some detail.

5.9. Describe some of the limiting or completing factors involved in using ISE electrodes in making measurements on soil or soil extracts.

5.10. What types of metal electrodes are best suited to making electrical measurements in soils?

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CHAPTER

6

TITRIMETRIC MEASUREMENTS

Titration is a general word used in many different disciplines. Any time a solution of known concentration is used to find the amount of an unknown component in another solution, it can be called a titration. Although this type of analysis is very old, it still finds widespread use in chemical analysis. Titrations are used in soil analysis to determine soil acidity, soil organic matter, and various constituents isolated from soil, particularly ammonia.

Common chemical titrations are acid–base, oxidation–reduction, precipitation, and complexometric and have been in use for a very long time. In all cases the basic concepts are contained in a classical acid–base titration. A known amount of acid is placed in a flask and an indicator added. The indicator is a strongly colored compound that is one color at acid pHs and a different color at basic pH. A base of precisely known concentration, the titrant, is then added to the acid until the pH becomes neutral as indicated by the indicator changing color. From the amount of titrant added, the amount of acid originally present is determined. This is a typical titration; however, a pH meter can also be used to detect the endpoint using a pH electrode, by following the electrode potential. Meters, usually pH meters, capable of measuring millivolts (Eh) or ion-selective electrodes (ISE), can also be used in a similar same way.

The endpoint in a titration is a little bit different from the end of a reaction. What is desired is to know when all the acid is titrated. This happens when the titration curve, shown in Figure 6.1, changes from acid to base. This change occurs when it passes pH 7 (for a typical strong acid–base titration), neutral, pH and when a color indicator is used, when there is a change in color. If the milliliter of acid added is plotted against pH, then a titration curve such as that shown in Figure 6.1 is obtained. The endpoint is the portion of the curve with the greatest slope.

In most cases a curve is not drawn and the endpoint is taken as the milliliters (mL) used just before the color change takes place. There should be a half-drop of titrant difference between the change from one color to the next. In many cases the color change is very light but distinctive. If the titration curve is plotted, then the endpoint can be determined by inspection or by

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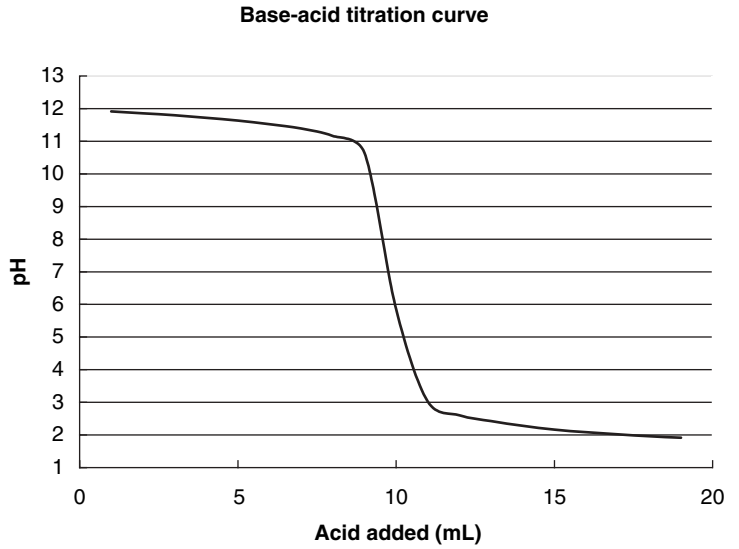

Figure 6.1. Curve obtained titrating a standard base with a standard acid.

taking the first or second derivative of the data. Both the first and second derivatives give an inflection point where the greatest slope of the titration curve occurs, thus showing the endpoint.

Although this explanation of titration has been simple, the same basic idea is applied to all forms of titation. In each case there is a slight change in pH, millivoltage, or ISE reading with added titrant followed by a sharp change through the endpoint followed again by a slight change.

Soil and soil suspensions are colored and difficult to see through; thus it is difficult to directly titrate them. There are typically only two cases where direct titrations of soil are carried out. Titration of soil has been used to determine the amount of amendment needed to bring the soil to a desired pH. The second direct titration is in the determination of soil organic matter where organic matter is oxidized with chromate and the unreacted chromate is titrated (actually called a *backtitration*) to determine, by subtraction, the amount of dichromate reduced and thus the amount of organic matter present.

In other titrations the component to be titrated is separated from soil and subsequently titrated. The simplest of these is the determination of soil ammonia. However, all forms of nitrogen in soil are important, and so methods of converting these to ammonia, distilling it, and determining its concentration by titration constitute an extremely important set of procedures.

Other environmental analytical procedures using titration can be found in the United States Environmental Protection Agency's (USEPA) Web site compilation of methods, particularly the 9000 series methods [see Bibliography].

6.1. SOIL TITRATION

Typically acid soils are titrated with a sodium or calcium hydroxide [NaOH or $Ca(OH)_2$] solution and basic soils with hydrochloric acid (HCl) and are most commonly followed using a pH meter. Carbonates in basic soils release CO_2 during any treatment with HCl, thus making the titration more difficult. For this reason carbonates are often determined by other methods. It is important to keep in mind that basic solutions react with carbon dioxide in air and form insoluble carbonates. This means that either the basic titrant is standardized each day before use or the solution is protected from exposure to carbon dioxide in air. Specific descriptions of titrant preparation, primary standards, and the use of indicators and pH meters in titrations can be found in the texts by Harris [see Bibliography], Harris and Harris [1], and Skoog et al. [2].

The complex nature of soil makes hydrochloric acid the preferred acid titrant because the other common acids can be involved in reactions, which are preferably avoided. Both sulfuric and phosphoric acids are di- and triprotic, respectively, and each proton has a different pK_a. In addition, in high concentration both are dehydrating agents and in low concentration are hydrating agents and thus can be involved in other than the desired titration reactions, potentially leading to erroneous results. Nitric acid, on the other hand, is monoprotic but is also an oxidizing reagent and can react with organic matter to produce nitro compounds and thus produce erroneous results. The use of sulfuric, phosphoric, and nitric acids may not be a problem with well-defined systems but can be a problem when applied to undefined systems, especially soils.

Oxidation–reduction reactions and titrations are often easy to carry out, and in many respects oxidation–reduction titrations are the same as an acid–base titration. For instance, a standardized oxidizing solution, often permanganate, is added to a solution of an easily oxidized species of interest. Permanganate is dark purple in color and colorless when reduced, making the endpoint easy to determine. Other oxidizing reagents can be used, and strongly colored molecules can be used as indicators in the same way as in acid–base titrations. Also, a platinum electrode coupled to a reference electrode can be used to determine the endpoint using most pH meters.

Most oxidation reactions are between specific metal cations or metal oxyanions and cations. The problem that arises when applying oxidation–reduction reactions to soils is that all soils contain a complex mixture of oxidizable and reducible cations and anions and organic matter, which means that it is impossible to determine which is being titrated. An exception to this is the oxidation of organic matter where an oxidation–reduction titration is routinely carried out. Organic matter determination will be discussed in Section 6.3.

Precipitation titration are typified by the titration of chloride with silver or vice-versa. In this case interferences with the precipitation reaction may occur because of components in the soil, and the soil itself may interfere with detection of the endpoint. Thus precipitation reactions are rarely applied

Table 6.1. Titrametric Methods Used by USEPA

Method	Species Titrated	Titrant
9014	Cyanide	Silver nitrate
9034	Sulfides	Iodine
9253	Chloride	Silver nitrate

directly to soil; however, they can be applied to soil extracts. Common environmental titration methods described in the USEPA methods are summarized in Table 6.1 [1,2].

6.1.1. Backtitration

In a backtitration an excess amount of standardized reagent is taken and reacted with an unknown amount of a component of interest. When the reaction is completed, the remaining unused reagent is titrated and the amount of component of interest is determined by difference. In the kjeldahl procedures described below, freed ammonia is distilled into an acid of known concentration. When all the ammonia has been distilled, the remaining unreacted acid is titrated, and one can calculate the amount of ammonia distilled (and thus in the original material being investigated) from the difference between the amount of original acid present at the start and the amount remaining at the end.

Backtitrations are common in soil analysis, as they are used in both nitrogen and organic matter determinations. Backtitrations are highly valuable analytical techniques and are applicable to other environmental analysis as well.

6.2. pH TITRATION OF SOIL

Titration of soil pH is an old method that is not widely used today. Basically, a soil suspension is prepared and titrated with a standardized base, often sodium hydroxide, although various basic calcium compounds such as calcium oxide (CaO) and calcium hydroxide [$Ca(OH)_2$] can and have also be used. Because of the dark color of many soils, they are often titrated using a pH meter as the indicator of the endpoint. A setup for the titration of soil is shown in Figure 6.2. Titration is slow in that it takes some time, after the addition of titrant for the semblance of equilibrium to be reached. Once this happens, a reading or simply another addition of titrant can be made.

A titration curve for an acid soil suspension to which 1 mL of a calcium hydroxide titrant is added and the change in pH followed for 2.3 min is shown in Figure 6.3. As can be seen, the pH initially increases and then falls back toward the original pH. The curve not only has a sawtooth pattern but is also curved in the reverse direction from a standard acid–base titration curve such as that shown in Figure 6.1.

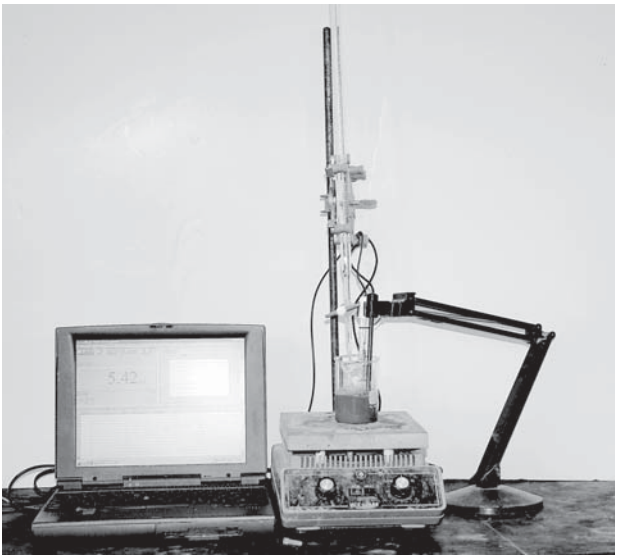

Figure 6.2. Setup for titrating and recording pH change using a pH meter card in a laptop computer.

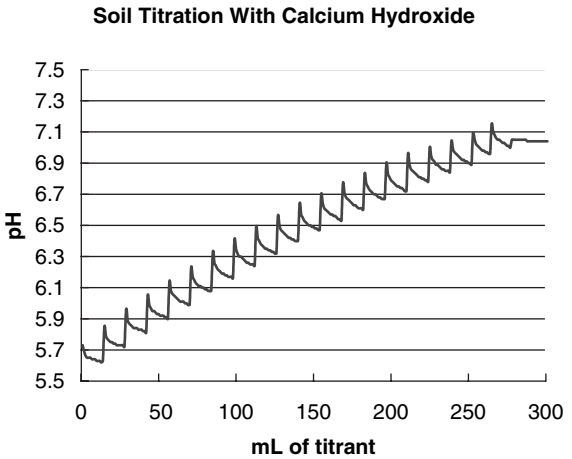

Figure 6.3. Stepwise titration of acidic soil with calcium hydroxide.

The initial pH rise as shown in Figure 6.3 and fallback is interpreted to be a result of two reactions. The initial rapid pH increase is a result of neutralization of free acid in the soil solution. Often this is represented as H^+; however, in aqueous solutions it is better represented as H_3O^+, the hydronium ion. The slower subsequent decrease in pH is a result of reequilibration

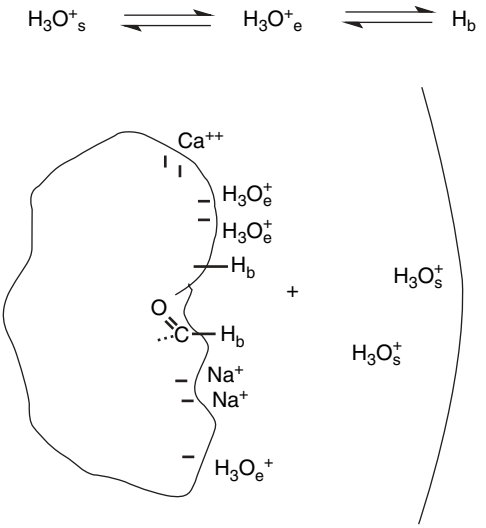

Figure 6.4. An equation showing the equilibrium between bonded protons (H_b), exchangeable protons ($H_3O^+_e$), and soluble protons ($H_3O^+_s$) is given above and illustrated below.

between H_3O^+ in solution and on exchange sites (Figure 6.4). In addition there may be a release of weakly held protons from either or both inorganic or organic constituents in soil. It might be envisioned that there is an equilibrium between all three sources of protons and that the decrease is a return to reestablishing this equilibrium.

In Figure 6.4 these three sources of protons are illustrated and designated as $H_3O^+_s$ hydronium ions in solution, $H_3O^+_e$ hydronium ions on exchange sites and H_b protons bonded to some soil constituent by either a covalent or polar covalent sigma bond. As discussed in previous chapters, measuring soil pH using a salt solution results in a lower pH being found. Here the cation provided by the salt replaces protons or hydronium ions on exchange sites, and thus they are in solution and can be measured. When a base such as NaOH is added to soil, the Na^+ cation will exchange with protons or hydronium ions on exchange sites in a similar manner. In addition, every proton exposed to the soil solution will have a pK_a value and thus be released or bonded depending on the pH of the solution. What is seen is that as the solution is made, more basic protons from all these various sources are potentially released into solution.

If a slow continuous addition of base is made to the same soil used in Figure 6.3, a similar titration curve without the sawtooth pattern is seen. Figure 6.5 shows the titration curve obtained by the continuous slow addition of 0.1 M NaOH. Again the curve is not a smooth line, and irregularities found in this titration are seen in other titrations of this same soil. Note that no distinct

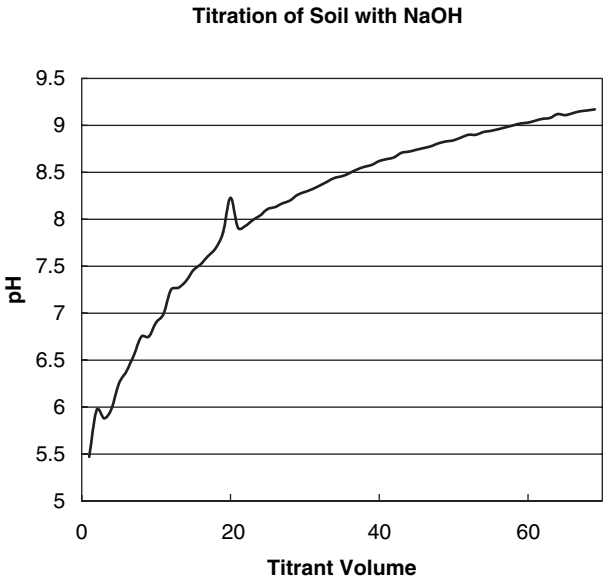

Figure 6.5. Titration of 50 g of soil suspended in 50 mL of distilled water with 0.1 M NaOH using a pH meter. Titrant was added slowly and continuously with stirring.

titration endpoint is seen here as there is in Figure 6.1. However, it is possible to determine the amount of base needed to bring this soil to pH 6.5, which is a typical pH desired for crop production.

This also explains why the pH of any extracting solution is important. Depending on the pH of the extracting solution, the component(s) of interest may be in the form of an ion or a polar or neutral molecule. This, in turn, determines whether it will be solvated by the solvent chosen as the extractant. If a certain pH is needed for an extraction process, then titration of a soil can be carried out in order to determine how much base or acid would be needed for the process. This would be useful in cases where removal of a contaminant from a spill site or a field is required.

***Caution*:** Lowering or raising a soil's pH to effect remediation, especially on a large scale, is not feasible for four reasons: (1) changing the pH of soil to any great extent requires large amounts of acid or base because soil is highly buffered, (2) soil is destroyed at both very high and very low pH levels, (3) a large amount of material that cannot be readily returned to the environment is produced, and (4) the material is no longer soil!

Because of the complex nature of soil and the soil solution, it is rarely possible to directly determine specific soil constituents by titrating soil or soil solutions using a pH meter, selective ion electrode, or a platinum electrode (with appropriate reference electrode (see Chapter 5 for more details on this subject) [3].

6.3. ORGANIC MATTER

Soil organic matter can be divided into many fractions; however, the first distinction between fractions is the active fraction, that is, the fraction undergoing active decomposition and the stable fraction, namely, the fraction that is relatively stable, mostly humus. The most common method of determining soil organic matter does not differentiate between these two types. All organic matter is oxidized using a strong oxidizing agent, most often potassium or sodium dichromate in sulfuric acid. To effect complete oxidation, heating, which can be done using a hotplate or by mixing the acidic and dichromate solutions, is required. When the reaction is completed, unreacted dichromate is titrated, in an oxidation–reduction titration, and the difference is used as the amount of organic matter present.

This titration uses an indicator, the color of which is difficult to see because of the soil present. The indicator, however, is much easier to see with natural rather than fluorescent lighting.

Other methods for determination of soil organic matter are available [4]; however, they are not as commonly used as is dichromate oxidation, commonly called the *Watley–Black* method. Usually these methods are both more time-consuming and less accurate than is the dichromate oxidation titration method. Keep in mind that the dichromate oxidation of organic matter is the standard by which all other methods of determining soil organic matter must be compared [4,5].

> ***Caution*:** Chromates, including potassium and sodium dichromate, are hazardous materials, as are sulfuric and phosphoric acid used in the oxidation of soil organic matter. Great care must be exercised in using these chemicals.

6.4. AMMONIA

Ammonia is a gas that reacts with water to form ammonium as follows:

$$H_3N + H_2O \rightleftharpoons NH_4^+ + {}^-OH \tag{6.1}$$

Figure 6.6. Setup for performing kjeldahl analysis of soil. On the left are two different types of kjeldahl flask; to the right of the flask is a heating block used to heat the text-tube-shaped flask during digestion and on the right, a steam distillation unit.

The equilibrium lies to the right unless the solution is rendered basic, at which point the equilibrium shifts to the left and ammonia gas is released. This is the basis for a common method for the determination of ammonium in soil.[1] Soil is suspended in water and placed in a kjeldahl flask. The suspension is rendered basic by the addition of a strong (5–50%) sodium hydroxide solution, and the flask is immediately attached to a steam distillation setup. Steam distillation of the suspension carries the released ammonia to an Erlenmeyer flask, catching the distillate in a standardized acid solution that is subsequently backtitrated, by acid–base titration. The amount of ammonia in soil can be calculated from the endpoint of the titration. This procedure is similar to a standard kjeldahl determination and can be carried out using the same equipment, although no digestion is needed.

A steam distillation apparatus for determining the ammonia released by the digestion of organic matter is shown on the right-hand side of Figure 6.6. This same apparatus can be used to determine ammonia in soil as described above. A flow diagram for the determination of ammonia in soil using a kjeldahl apparatus is given in Figure 6.7 [6].

[1] A cation containing solution is added before ammonium determination to release exchangeable ammonium.

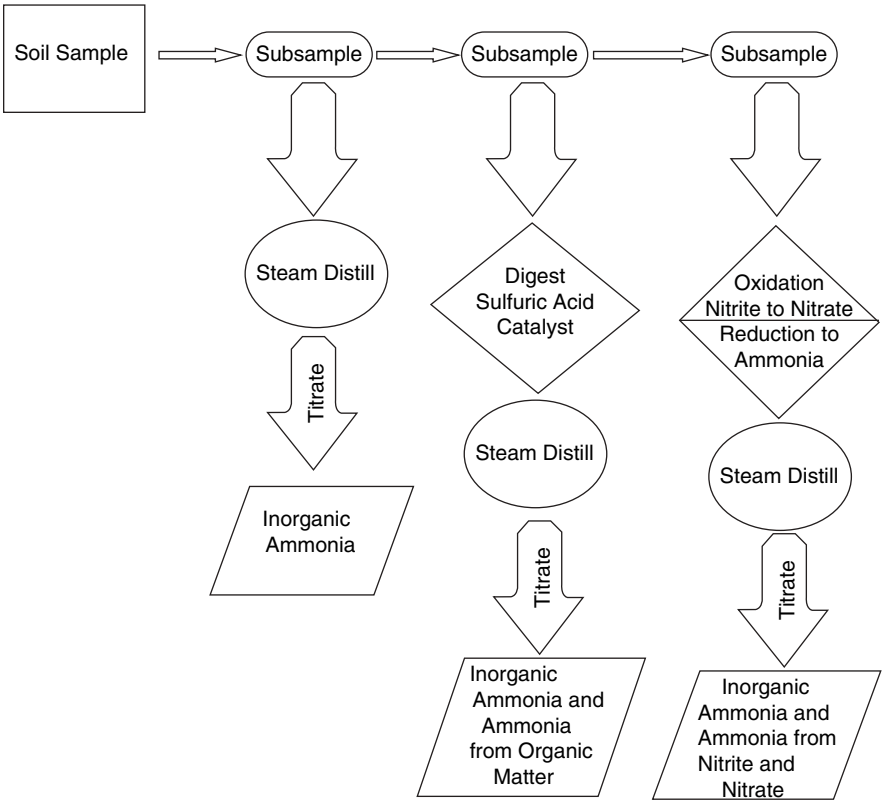

Figure 6.7. Flow diagram for determination of ammonia, organic nitrogen, nitrite, and nitrate using kjeldahl apparati [6].

6.5. KJELDAHL—ORGANIC NITROGEN

***Caution*:** Extreme caution must be exercised in carrying out a kjeldahl procedure. Digestion involves the use of concentrated sulfuric acid and in the past selenium and mercury have both been used a catalyst. Older equipment may be contaminated with these elements and should be treated with caution. During the digestion process copious amounts of *choking, corrosive, and toxic* sulfur dioxide fumes are released. These fumes must be removed by evacuating them using an aspirator or some other appropriate method. Standard kjeldahl equipment will come with components and directions for removing SO_2 safely. Ammonia distillation involves use of highly concentrated caustic sodium hydroxide solutions, which must be handled with care. Sodium or potassium hydroxide spilled on skin is best removed with a dilute solution (1–2%) of acetic acid in water followed by washing with soap and water.

Nitrogen in soil organic matter is mostly in the form of proteins and amino acids. Although the specific analysis for these important and interesting compounds can and is done, it is more often the case that the total inorganic and organic nitrogen in soil is determined. This is because inorganic nitrogen compounds are used by plants and are of environmental concern. Decomposition of organic nitrogen containing compounds results in the release of ammonia into the soil solution, where it immediately reacts to form ammonium. Once in this form, it is readily oxidized by soil bacteria to nitrite and finally into nitrate. Because of the ready conversion of organic nitrogen into inorganic forms and the ready interconversion of inorganic nitrogen in soil, its total concentration, both inorganic and organic, is important. Kjeldahl is the basic and most commonly used method for determination of total nitrogen in soil.

In a total soil nitrogen analysis a soil sample is first digested in a kjeldahl flask to convert all organic nitrogen into inorganic ammonium. Two kjeldahl flasks are shown on the left-hand side in Figure 6.7; the flask with the bulb at the bottom is an older-type kjeldahl flask, while the large test tube is a newer-design digestion tube. Digestion is accomplished using concentrated sulfuric acid and a catalyst. A salt such as potassium sulfate is added to increase the boiling point of sulfuric acid such that decomposition of organic matter occurs more readily. This mixture plus soil is heated until all organic matter has been destroyed. In Figure 6.6 a heating block for heating the kjeldahl flask is shown next to the distillation unit. After digestion the solution is cooled and a concentrated basic solution, usually 50% NaOH, is added and the released ammonia steam-distilled into a receiving flask containing a standard acid that reacts with the ammonia. On completion of the steam distillation, the unreacted acid is titrated and the amount of ammonia distilled is calculated by difference.

The kjeldahl procedure has been used for many years to determine the nitrogen in human tissues and in both animal and human foodstuffs. For these materials, the procedure works well and is straightforward. For soil, such is not the case. All soils naturally contain some ammonium, and when the steam distillation is carried, out this distills along with the ammonium produced by the decomposition of organic matter. This then gives a measurement of the total ammonium in soil after digestion. It cannot distinguish between ammonium derived from organic matter and from the soil itself.

If a soil kjeldahl organic nitrogen determination as described above has been carried out, then this can be used along with simple ammonia steam distillation, to measure the amount of nitrogen from each source, that is, inorganic ammonium and organic matter. However, this still does not provide a measurement of the total nitrogen in soil because it does not account for that present as either nitrite or nitrate. See Figure 6.7 for a flow diagram for determining all nitrogen in soil using a kjeldahl apparatus.

6.6. NITRITE AND NITRATE

Both nitrite and nitrate are highly mobile in soil and easy to extract. However, it is also possible to reduce each individually to ammonia and subsequently steam-distill the ammonia, capturing and titrating it as described above for ammonia. If this procedure is to be followed, naturally occurring ammonia in soil must first be determined as described above. After this step, a reducing agent is placed in the flask and nitrite and nitrate reduced to ammonia. The soil is then rendered basic again and the ammonia steam-distilled and titrated. If both nitrate and nitrite are reduced at the same time, the combined amount of both is obtained. At this point selective reduction of either nitrate or nitrite with subsequent distillation and titration will allow for calculation of the amounts of all three forms of nitrogen in the soil sample.

The determination of nitrite and nitrite in soil can be accomplished by reactions carried out before digestion or steam distillation to oxidize nitrite to nitrate and to reduce nitrate to ammonia. Alternately, nitrite can be reacted with an organic compound, typically salicylic acid and subsequently digested in a typical kjeldahl procedure. This procedure starts with an organic molecule that reacts with nitrite to form a nitro compound that is subsequently reduced to an amine. The amine is then subject to digestion just as with organic matter in soil. In this way the total nitrogen content in soil can be determined. Also, when this procedure is combined with those described above, the amounts of the various forms of nitrogen in soil—organic, ammonia, nitrite, and nitrate—can be determined. This is thus an extremely powerful method for elucidating the nitrogen status of soils. Figure 6.7 gives a flow diagram for determining nitrite and nitrate in soil using a kjeldahl apparatus.

The procedures described above determine soil nitrogen that is often taken to be the total nitrogen in soil. Because very few other organic or inorganic nitrogen compounds are commonly found in soil, there is little call for additional analytical procedures. However, there are some exceptions to this. It may be necessary to determine the gaseous oxides of nitrogen, formed during denitrification. This is typically accomplished using gas chromatography (see Chapter 9). Also, using simple steam distillation ammonia trapped in clay structures will not be determined. Such determination requires complete destruction of soil minerals and the subsequently released ammonia determined.

Although kjeldahl procedures are capable of providing information about all the common nitrogen components in soil, it is a time-, labor-, equipment-, and reagent-intensive procedure. Typically kjeldahl equipment, namely, a digestion–steam distillation apparatus, can accommodate 6, 12, or more digestion tubes or flasks and distillation, and titration can be done quickly; however, it will take hours to do any significant number of samples. For ^{15}N work, additional steps, time, and money are required to convert ammonia to nitrogen gas and determination of ^{15}N by mass spectrometry (see Chapter 8) [6].

6.7. CARBONATE DETERMINATION

Carbonates decompose under acidic conditions with the release of carbon dioxide:

$$CaCO_3 + 2HCl \rightarrow CaCl_2 + CO_2\uparrow + H_2O \quad (6.2a)$$

$$CO_2 + 2NaOH \rightarrow Na_2CO_3 + H_2O \quad (6.2b)$$

To determine the carbonate, a soil sample can be placed in an Erlenmeyer flask and a 0.1 molar solution of hydrochloric acid (HCl) is added until no more carbon dioxide is released. The amount of HCl consumed is used to calculate the amount of carbonate present. The reaction shown is for calcium carbonate; however, all carbonates in the soil will also be decomposed, and thus this is a method for determining the total carbonate content, not just the calcium carbonate. Additionally, inaccuracies can be caused by other components in soil that can react with the HCl.

Another approach would be to measure the amount of carbon dioxide produced either by measuring the volume of gas released or by reacting the carbon dioxide, in a separate flask, with base and determining the amount of base remaining after all the carbonate has precipitated and been removed from the solution.

Alternatively, the weight lost when the carbonate is reacted with HCl can be determined. Heating carbonates results in their decomposition. Thus soils containing carbonate can be heated to the appropriate temperature and the weight loss measured. In this approach the loss of organic matter and water of hydration of various components in soil must be corrected for in order to determine the weight of carbon dioxide lost [7].

6.8. HALOGEN ION DETERMINATION

Inorganic halogen containing compounds are usually very soluble salts. They commonly occur as simple, single, negatively charged anions in soil. There are two common exceptions to this generalization. First, fluorine is commonly found bonded to phosphate in insoluble minerals called *apatites*, which are calcium phosphate fluorides. The second are halogens, which are sigma-bonded to carbon.

The halogen anions are easily leached from soil with water and can be determined using silver nitrate as a titrant. The letter X is commonly used to represent halogens and thus may be interpreted to be any of them (i.e., F, Cl, Br, or I) but is not generally used for At. The following reaction of silver with halogen X^- ignores other possible counterions, namely, nitrate and the cation associated with the halide:

$$X^- + Ag^+ \rightarrow AgCl\downarrow \tag{6.3}$$

Reaction (6.3) is a precipitation titration that can be done with or without an indicator. Silver as a cation is very reactive and is an oxidizing agent. For instance, it can oxidize aldehydes, including aldoses (sugars with an aldehyde functionality). For this reason it is possible to obtain inaccurate or misleading results with titrating a soil extract with silver nitrate.

The most common halogen in soil is chloride while both bromide and iodide occur but are uncommon [8]. The occurrence of either of these anions in soil would be cause for concern. Analysis for these other halides could be carried out using either capillary electrophoresis or high-performance liquid chromatography (HPLC) (see Chapters 8 and 9).

6.9. pH–STAT TITRATIONS

In another type of titration, termed *pH–stat*,[2] the system is maintained at one fixed pH during a reaction. This type of titration has been applied to bioreactors where neither the starting material nor the product of the reaction is titrated but rather acidic or basic byproducts or coproduced acid or base are measured. In biological reactions this may be CO_2, HCO_3^-, or CO_3^{2-}. The system, maintained at a fixed temperature during the reaction of interest, is titrated with an automatic titrator set to maintain the specific pH. Bioreactors may be sealed to ensure that no liquid or gas is lost or gained, or they may be open to allow the exchange of gases.

The titrant used in pH–stat procedures is usually a dilute (perhaps 0.01 molar) acid or base. Two different sets of data can be obtained from a pH–stat titration. The amount of a substrate consumed or product formed can be determined by the total amount of titrant used. Because the titrant is added over a period of time, the rate of reaction can also be determined.

Even a small soil sample can be considered as a bioreactor, and so the pH–stat method is applicable to soil. However, although this method is applicable to the study of processes occurring in soil, it is not particularly useful in routine analysis of soil components [9].

6.10. CONCLUSIONS

The titrimetric determination of soil constituents is most commonly applied to a limited number of soil analyses, namely, organic carbon, nitrogen compounds, carbonates, and chlorides. Determination of acid content by titration is seldom done because the titration curves are not amenable to typical titra-

[2] *Stat* stands for static and derives from the fact that the pH is held static during the progress of the reaction.

tion analysis. Because of the color of soil and the fact that it is a suspension when stirred, it is often necessary to remove the constituent of interest before titration. In other cases it is possible to do a direct titration using an appropriate indicator. However, even in these cases detection of the endpoint is difficult.

Because of the complex nature of titration curves obtained using pH, ion-selective electrodes, or mV (Eh) measurements on whole soils, these methods except for organic matter determination, are seldom used.

PROBLEMS

6.1. Explain why the endpoint of a titration is at neither end of a titration curve.

6.2. Explain why titration is not a generally useful method for discovering the acidity of soil.

6.3. Suggest areas in soil where there might be organic matter that is not determined by dichromate oxidation (refer to earlier chapters).

6.4. Make a flow diagram that shows how to determine all different forms of nitrogen found in soil.

6.5. Look up the titrimetric method for the determination of cyanide and describe it.

6.6. Give the equation for the reaction of carbonate with acid. Describe two ways in which titration might be used to determine carbonate.

6.7. In environmental analysis ^{15}N can be used to determine where nitrogen moves in the environment. Tell how ^{15}N containing inorganic compounds might be isolated from soil and how it could be specifically determined. (*Note*: You might wish to consult Chapters 7–9 in answering this question.).

6.8. Describe pH–stat titration in detail.

6.9. Describe the two types of information that can be obtained about a reaction by using the pH–stat method of titration.

6.10. During most titrations the solution or suspensions are mixed sometimes continuously. Considering this, why might it be a good idea to use an indicator during a complexiметric titration?

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CHAPTER

7

EXTRACTION

Soil analyses involving instrumentation generally have extraction procedures associated with them. Thus, in addition to the references cited below and listed at the end of this chapter, additional understanding of extraction procedures, how they are designed and used, can be found along with the descriptions of spectroscopic and chromatographic methods and the corresponding references cited and listed in Chapters 8 and 9.

As has been pointed out in previous chapters, soil is a complex mixture of inorganic and organic solids, aqueous and gaseous solution, and suspension of inorganic and organic ions, molecules, and gases, which can be sorbed, dissolved, or free. In most cases analysis for a particular component first involves isolation of that component from all the other myriad soil components, specifically, the soil matrix. Isolation may involve physical separation such as precipitation or distillation or, more often, as a first step, after sieving is extraction of soil using an appropriate extracting solvent or solution. Once isolated, the component can be directly or indirectly measured. Direct methods usually involve spectroscopy, while indirect methods involve production of a colored product by reaction with an applicable reagent and carrying out a colorimetric measurement.

Both direct and indirect measurement involves the preparation of a standard or calibration curve. With an accurate calibration curve, the concentration of the component can be determined taking into account interferences, dilution, and extraction efficiencies.

7.1. ISOLATION

Physical methods of separation and isolation of components from soil are commonly used in soil analysis, for instance they are particularly useful in nitrogen compound determination, as described in Chapter 6, and by head space analysis, as described below.

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

Organic compounds, including pollutants, can be desorbed from soil by heating using a procedure called *headspace analysis*. Heat-desorbed compounds can then be determined by gas chromatography. A soil sample that does not fill the container is sealed inside the container using a cap containing a septum. The container is brought to a predetermined temperature and allowed to equilibrate. A syringe is inserted into the sample container, and a sample of the headspace gas is removed and injected into a gas chromatograph (GC) or a gas chromatograph–mass spectrometer (GC/MS) for separation and identification of the components released from the soil.

There are other ways of removing components of interest from the soil matrix without using a solvent per se. Sparging, solid phase and solid-phase microextraction (SPME) are three of the most common of the nonsolvent methods.

In sparging, a soil sample is placed in a container and a flow of gas, usually pure helium or nitrogen, is passed through it to remove components of interest. The soil may also be heated to remove components with lower vapor pressures. Gas exiting the container flows over a sorbant that removes or traps the components of interest. After a preset period of time at a prescribed temperature and flow rate, the flow is stopped and sorbed components are extracted from the sorbant and analyzed by GC or GC/MS.

Alternatively, the sorbant used in sparging can be placed directly in soil to sorb components of interest. After a prescribed period of time, the sorbant is removed from soil, extracted, and analyzed by GC or GC/MS. In direct sorption there is always the question of contact between the sorbant and soil components. Therefore these procedures must be investigated to ascertain and validate their precision and accuracy.

Direct heating of soil in a gas chromatograph inlet and gas chromatographic analysis of released compounds from soil has also been used to analyze for absorbed compounds [1–9].

7.2. EXTRACTION

As a first step in any soil extraction, a decision between an aqueous and nonaqueous extracting solvent or solution is made. Typically inorganic compounds are extracted using either one of two types of aqueous solutions or in the simplest cases water. One type of aqueous solution containing a cation is designed to extract cations attached to exchange sites in the soil. Another is an aqueous solution containing ligands designed to form a strong attachment to the compound of interest. For simple, highly soluble compounds, a simple water extraction may be sufficient [10].

In extracting cations and, in some cases, anions, the extracting solution must contain ions appropriate for accomplishing two tasks; they must (1) be capable of exchanging with the ion of interest and (2) not interfere with subsequent analytical procedures or analysis. Most commonly in soil the clay and organic matter carry a net negative charge and thus attract cations, resulting in soils

having cation exchange capacity (CEC). There are, however, some soils that have significant anion exchange capacity, and this possibility must never be overlooked.

In cation exchange reactions typically both the charge and concentration of the cation in the extracting solution are important in determining its ability to exchange with cations already on the exchange sites. Generally, the larger the charge on the cation, the more effective it is in replacing other cations, especially those with less charge. Cations with less charge but at high concentration will replace cations with higher charge. This latter approach is generally the one taken in carrying out analysis involving cation exchange; thus an extracting solution containing a high concentration of lower charged cation is used in the extraction [11,12].

In the case of compounds not attracted or attached to cation or anion exchange sites in soil, an aqueous solution containing a ligand or a mixture of ligands and auxiliary compounds that form an attraction for the compound of interest and result in the formation of a highly soluble compound, complex, or species is chosen. A solution of this ligand in distilled water is prepared and used to extract the soil [13–15].

Another approach that has been used is to add a surfactant to aqueous extractants, particularly when insoluble components are to be extracted or are present. Typically soaps and both ionic and nonionic surfactants have been used. Because of the complex nature of some surfactants and their effect on viscosity, care must be taken to make sure that they do not adversely affect subsequent analytical procedures [16–18].

In a similar fashion organic compounds are typically extracted using organic solvents or mixtures of organic solvents. In this case, because of the constant occurrence of water in soil, the solubility of water or the mutual solubility of the compound of interest and water in the extractant will be an important consideration.

An attempt to circumvent these problems is to use mixed solvents, which are soluble in each other as well as in water. Thus a mixture of acetone, which is miscible with water, and hexane, which is a hydrophobic hydrocarbon insoluble in water, could be used as a soil extractant. The idea is to have a solvent that will dissolve in water and yet also dissolve hydrophobic contaminants in soil (see Section 7.2.1).

Nonaqueous or hydrophobic extractants can also be used, although typically their usefulness is limited. Examples of these types of extractants are halogenated solvents such as the freons and chloroethanes. A problem with all these types of extractants is that water in nondraining pores is not accessible to them and thus any contaminant surrounded by water will not or will only partially be extracted.

In the case of all of the above mentioned extractants, there are three important questions:

1. Will the extractant extract the component or contaminant of interest?
2. Is the extractant compatible with the analytical procedures to be used?

3. What is the distribution of the component between the soil matrix and the extractant?

The best answer to questions 1 and 2 both is affirmative, but if it is negative, then the incompatibility must be removed. One method of accomplishing this is to remove the initial extractant and redissolve or suspend the component of interest in an analytically compatible solvent. Another approach would be to sorb the component on a suitable sorbent and subsequently extract it from the sorbent into a suitable, compatible solvent. The simplest and best approach is simply to extract the component of interest into a compatible solvent directly. Regarding question 3, extraction time and/or the need for multiple extractions must be sufficient to extract all the component of interest. This information is usually obtained by carrying the extraction out over varying times or varying the number of times and determining which gives the highest extraction efficiency.

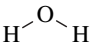
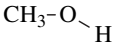
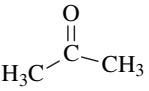
7.2.1. Solvents

> ***Caution*:** Common organic extracting solvents are both volatile and flammable. Ignition can be caused by hot surfaces without the need of sparks or flames. They will also form peroxides, particularly diethyl ether, which are explosive when concentrated, especially when heating is involved. Also, some solvents may be toxic or carcinogenic or both. Always consult the U.S. Environmental Protection Agency (USEPA) or Material Safety Data Sheets (MSDSs) before using any solvent.

In all environmental work, particularly soil analysis, the purity of solvents used is of extreme importance. This stems from three sources. Soil is extremely complex in its composition, which can lead to solvent contaminants being confused with or interfering with the separation and detection of the analyte or analytes of interest; see Chapters 8 and 9 for more details on these types of interference. The second source results when extractants are concentrated prior to analysis. In this case any impurities present will also be concentrated, but they may not all be concentrated proportionately. The third source is the sensitivity of modern instrumentation, which results in detection of very low solvent contaminant concentrations.

In soil analysis for agricultural purposes, micronutrients, such as iron, boron, and copper, are important and as the name implies, are present in low concentration. As these are micronutrients, sensitive methods of analysis are needed to measure them. If present at low concentrations, they are not sufficient for optimum plant growth. If present at high concentrations, they are toxic. In environmental analysis determination of low concentrations is there-

Table 7.1. Common Extraction Solvents and Their Structure and Characteristics

Name	Structure	Characteristics
Water		Dissolves many compounds and is present in many other extractants; can cause interference in analytical and instrumental methods
Methanol		Miscible with water; can dissolve some hydrophobic or sparingly soluble materials; can cause interference in analytical and instrumental methods
Acetone		Miscible with water; dissolves many hydrophobic compounds; low boiling point
Diethyl ether	$C_2H_5—O—C_2H_5$	Very low solubility in water; low boiling point; dissolves most hydrophobic compounds
Hexane	$CH_3(CH_2)_4CH_3$	Hydrophobic—insoluble in water; dissolves hydrophobic compounds; has higher boiling point than diethyl ether or acetone
Methylene chloride	CH_2Cl_2	Hydrophobic—insoluble in water; dissolves hydrophobic compounds; has low boiling point

fore essential. Some toxic elements, such as arsenic and selenium, are commonly present in soil and it is essential to differentiate naturally occurring levels from contamination. Organic contaminants, both industrial chemicals such as poly(chlorinated biphenyl)s (PCBs) and agricultural chemicals such as insecticides and herbicides, must be determined at low concentrations. In all these cases this is best done when there are no interferences from contaminants in the solvents, including water.

There are a limited number of solvents used in common soil extracting procedures, ranging from water to the nonaqueous and hydrophobic. Each has its own uses and limitations. The most common solvents and their chemical structure and characteristics are listed in Table 7.1. Solvents are available in a variety of purities designed for various uses, and thus it is important to carefully read the description of the solvent and its grade before use. High purity does not guarantee compatibility with the extraction, analysis, or instrumentation to be used.

Many organic solvents are sold containing small amounts of additional compounds, generally used to increase their stability and shelf life. Diethyl ether is available in several different "stabilized" forms. In some cases these stabilizing agents, such as alcohols, may interfere with subsequent analysis. In addition to stabilizing agents, ethanol is often denatured, which means that it is not drinkable. Depending on the manufacturer, the denaturing agents may be other alcohols or other compounds. Thus, not only must the purity but also any additives present must be known.

All nonaqueous solvents may contain traces of water, even when they are said to be or are sold as anhydrous. If truly anhydrous solvents are needed, additional drying (see Section 7.3) may be needed.

7.2.1.1 Water

Water is a common soil extracting solvent. Pure water for use in extraction or as the solvent for extracting solutions would seem to be a simple matter, and in the past it has been much simpler than today. This is due in part to increasing demands on the purity of the water used and partly on changes in technology used in water purification. Distilled, doubly distilled, and sometimes triply distilled water might have been called for in the past or in older literature and procedures. Today there are many other types of purified water, and some extractions may call for water purified by a specific procedure.

In some situations it may be desirable to combine one or more purification procedures in order to obtain water of sufficient purity for the extraction and analysis to be undertaken. It is common to combine deionization with reverse osmosis to obtain what is commonly referred to as *deionized water* or *DI water*. In some cases deionization or filtration may be desirable before distillation is carried out. Or all three purification methods may be combined to produce highly purified water.

Highly purified water must be kept under conditions that maintain its purity. Clean containers are usually constructed of glass or plastic; plastic is the preferred container material today because highly purified water will dissolve small amounts of glass. Also once in the container, the water must be protected from contamination with foreign material and gases. Carbon dioxide, from the atmosphere, will dissolve in water and form bicarbonate and carbonic acid. Highly purified water can be protected from carbon dioxide by storing it under a nitrogen atmosphere or by filling the container completely and having displacing air pass through an upside-down drying tube filled with a layer of ascarite between two layers of indicating drieate or for short time periods sodium hydroxide as illustrated in Figure 7.1. Although this will protect the water from carbon dioxide, it will not prevent the water from absorbing other gaseous atmospheric contaminants. If the absorption of other potential contaminates is of particular concern, drying tubes containing other absorbents arranged in tandem can be added. All drying tubes must be changed as soon as deterioration is seen. Water must always be checked to verify its purity.

A way to avoid contamination problems occurring during storage is to prepare purified water fresh just before use. However, even in this case care must be taken to ascertain the purity of the water being produced [19].

7.2.1.2 Organic Solvents

Common laboratory solvents are most frequently used as they come from the supplier and will be supplied with some information as to their purity. However, the purity statements provided by the manufacturer or supplier may not provide

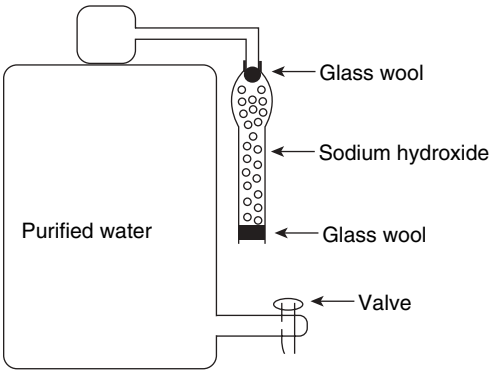


Figure 7.1. Purified water container protected from carbon dioxide by a drying tube containing sodium hydroxide (*Caution*: adsorption of water by N_2OH produces caustic solution).

all the information that the researcher needs. In all cases solvents must be protected from exposure to light, which can cause the formation of degradation products that may cause interferences during analytical procedures. Common organic solvents and their characteristics are listed in Table 7.1.

Solvents come in many grades that are designated in different ways. There are 7 grades of hexane and 10 grades of hexanes offered by one supplier. These range from 98.5% hexane through American Chemical Society (ACS) grade, to various chromatographic and other grades. In some cases it will be essential to use the specific grade designated for a particular analysis; in other cases further purification may be necessary. The most important example is the case of water where a solvent designated as being anhydrous may still contain enough water to interfere with reactions, instrumentation, or analysis that are highly sensitive to water. Additional drying may be called for, and care must be taken to make sure that drying agents introduce no contamination.

Solvents must be protected not only from light but also, as with water, from absorbing vapors and from the atmosphere. Solvents must be kept tightly sealed and where the highest purity must be maintained, stored under nitrogen. Another approach that can be used is to purchase solvents in individual, small-volume, containers where the whole bottle is used in one analysis, in one day or over a short period of time [20].

7.3. DRYING AGENTS

In all cases where drying agents are to be used, both their composition and purity must be ascertained to ensure that they will not introduce any interfering contaminates to the material, including soil, being dried (see Sections 7.4, 7.5.2, and Table 7.2). As with water and solvents, exposure of drying agents to the atmosphere allows for the introduction of foreign components, in this

Table 7.2. Characteristics of Drying Agents

Drying Agent	Characteristics: Capacity/Speed/Intensity[a]	Restrictions
Calcium chloride	High/medium/high	Reacts with many functional groups
Calcium sulfate	Low/fast/high	Applicable in most situations
Magnesium sulfate	High/fast/moderate	Applicable in most situations
Potassium carbonate	Medium/medium/moderate	Reacts with acids and phenols
Sodium sulfate	High/low/low	Hydrate decomposed on heating
Molecular sieve 5 Å	High/high/high	Suitable for most solvents

[a] Capacity = amount of water absorbed, speed = rate of water removal, intensity = level of water remaining after drying (e.g., "high" means that smallest amount of free water remains after drying).

case including water. Thus these agents must always be kept tightly sealed and checked for purity from time to time. The purchase and use of small, individually packaged amounts of drying agents is another way to limit the possibility of introduction of contaminants.

7.4. EXTRACTION PROCEDURES

The actual extraction procedure may be as simple as adding the extractant to the soil sample in an Erlenmeyer flask and mixing. It might be more complex, involving Soxhlet, ultrasonic, microwave-assisted, accelerated solvent or supercritical fluid extraction. In extracting soil using hydrophobic extractants the procedure may call for drying the soil before extraction. Air drying may be sufficient; however, mixing the soil with a drying agent; often anhydrous calcium sulfate may be required. A specific amount of drying agent will be called for, or the procedure may call for adding an amount of drying agent equal to the weight of soil or the addition of drying agent until the soil is dried to a prescribed consistency. Common drying agents are listed in Table 7.2.

7.4.1. Simple Mixing

Mixing is commonly accomplished using a reciprocal shaker equipped with clips for holding the flask to the shaker platform. The shaker can then be set to the prescribed orbits per minute and time for the extraction. This type of mixing works well when a large number of samples need to be extracted at one time. Alternatively, the sample could be stirred using a mechanical or magnetic stir bar stirrer.

7.4.2. Soxhlet Extraction

Soxhlet extraction (see Figure 7.2) is used extensively and is the standard method for extracting oils from a wide variety of materials, including plant materials and contaminated soils. The round-bottom flask is filled half-full with the extractant; the sample to be extracted is placed in a thimble positioned in the siphon extractor and a condenser attached to the top of the extractor. A mantle, placed under the flask, heats it so as to maintain a constant flow of condensed solvent into the thimble. When the extractor fills with extractant, it is automatically siphoned back into the flask. Extracted components that are normally higher-boiling than the extractant remain and are concentrated in the flask. After the prescribed extraction time, the extractant is analyzed for the component(s) extracted from the sample.

Soxhlet extraction can be used with organic solvents or water and often results in a solution that is ready to analyze. Figure 7.3 shows the results of extracting 10 g of soil using a Soxhlet device and a magnetic stirrer. The two flasks on the left were extracted for 8 h using a Soxhlet extraction procedure. The leftmost flask is a hexane extract that is absolutely colorless, while the center flask is a water extract. The right-hand flask is the solution obtained when 150 mL of distilled water was mixed, using a magnetic stirrer, for 8 h followed by filtration. The aqueous Soxhlet extraction has a deep yellow color,

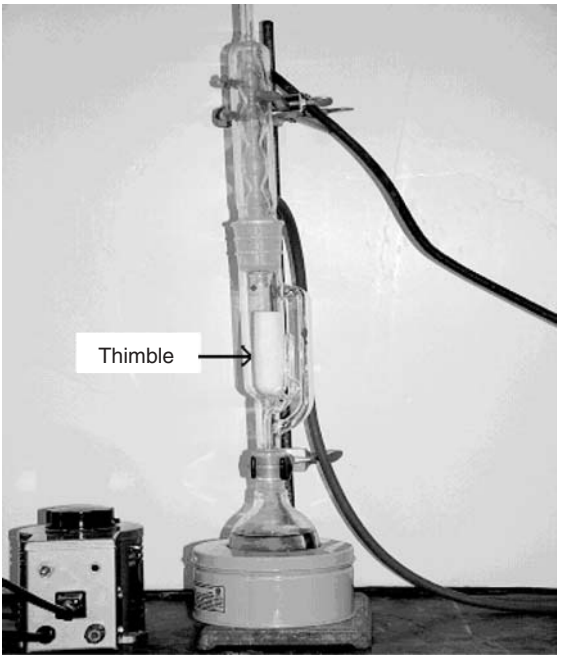


Figure 7.2. Soxhlet setup for extraction of solid samples.

Figure 7.3. Solutions obtained by Soxhlet and magnetic stirring of soil with either water or hexane.

while the magnetic stirred extract is cloudy even after being filtered 2 times. All three solutions have ultraviolet absorptions as a result of components extracted from the soil [21].

7.4.3. Ultrasonic Extraction

Ultrasonic extraction or sonication involves disruption and mixing a sample and extractant using *ultrasound*, which is sound above the hearing range of approximately 10,000 Hz. A high-frequency electrical current is applied to a piezoelectric crystal that causes a metal tip attached to it to oscillate at high frequency. This produces a series of compressions and relaxations in the extraction mixture, causing disruption and mixing in the extraction vessel. A typical ultrasonic "horn" is shown in Figure 7.4. A complete ultrasonic extraction setup involves both the "horn" and a high-frequency source [7,22].

7.4.4. Microwave-Assisted Extraction

> ***Caution*:** Microwave-assisted extraction must always and only be carried out in microwave ovens and digestion containers, with appropriate sensors, especially designed for this purpose.

Microwave-assisted extraction involves sealing a sample and extractant in a special microwavable digestion container, placing it in a special microwave

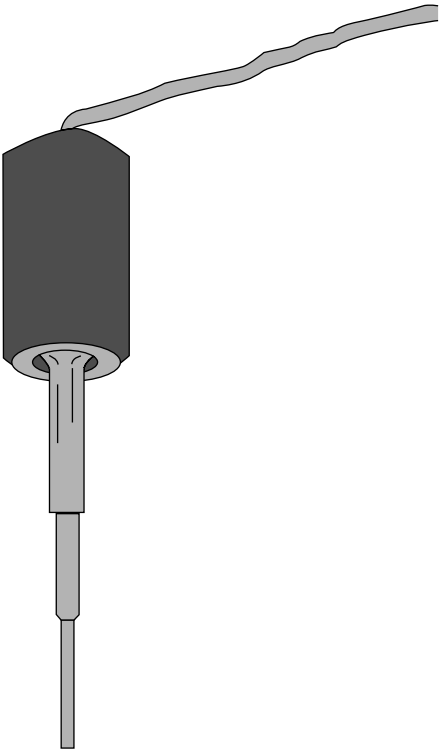


Figure 7.4. Ultrasonic "horn" for disruption and extraction of soil samples.

oven, and microwaving for the designated time period. Microwaves heat the sample with consequent increase in container pressure, both of which decrease the time needed for and increase the efficiency of extraction. Both temperature and pressure are monitored and can be controlled during microwave-assisted extraction. Because high temperature and pressure are produced, it is essential that both the containers and microwave be designed specially for this process. A microwave assisted microwave and extraction containers are shown in Figure 7.5 [8,21,23,24].

7.4.5. Supercritical Extraction

Supercritical fluid extraction (SFE) involves extracting a material using carbon dioxide maintained as a supercritical fluid. To do this, the carbon dioxide must be kept above its critical temperature and pressure such that it is maintained in its liquid state (liquid CO_2) during the extraction process, for example, 350 atm and 80°C. A soil sample is placed in an extraction vessel that can be maintained at the necessary pressure, and liquid CO_2 is passed through the soil and into a collection container. In some cases additives may be introduced

Figure 7.5. A microwave-assisted instrument showing extraction vessels and attachment for sensors (courtesy of Dr. Courso and Dr. Conklin, University of Cincinnati).

to the liquid CO_2 before extraction to change the extraction conditions. A diagram of a liquid CO_2 extraction setup is shown in Figure 7.6.

An advantage of supercritical extraction is that when the extract is brought to room temperature and pressure, the carbon dioxide is released as a gas, leaving the extract free of extraction solvent (assuming that no additives have been used) and thus in a concentrated state ready for further cleanup or direct analysis [21,25,26].

7.4.6. Accelerated Solvent Extraction

Physically, accelerated solvent extraction (ASE) is similar to supercritical extraction, except that in this case the extractant is normally a liquid at room temperature and pressure. However, it is heated to above its boiling point but kept in its liquid state by keeping it under pressure. The advantage of this process is that it allows for a greater range of extraction parameters in that solvents of varying functional groups, polarities, and so on can be used to effect the extraction. The extraction process can be more selective in the type of material extracted, and the speed of extraction can be increased [9,27,28].

7.4.7. Solid Phase and Solid-Phase Microextraction

In solid phase and solid-phase microextraction a solid adsorbant is placed directly in soil for a period of time. The solid adsorbant adsorbs compounds

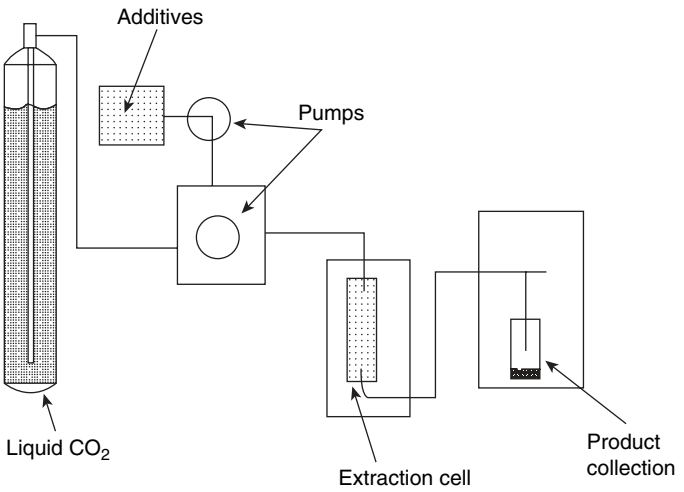


Figure 7.6. Setup for supercritical extraction of soil.

of interest from the soil that are later desorbed or extracted from the adsorbant and analyzed by chromatographic and spectrophotometric methods. There are two issues with this type of analysis: (1) whether there is significant contact between soil and the adsorbant such that a representative sample of the compound of interest is obtained and (2) whether it can be ensured that the adsorbant is not completely saturated such that some of the compound of interest is not adsorbed, resulting in an inaccurate analytical result. If these two conditions can be adequately addressed, then this procedure can be used successfully [1–5,7–9].

7.5. EXTRACT CLEANUP

Although extraction methods are designed to extract a specific component of interest, it is rare that the extract will be clean enough for direct analysis. During extraction many compounds and water will be dissolved in the extracting solution. Some of these components will be innocuous, but some will interfere with further analysis or decrease either the accuracy or precision of the analysis, and some may prevent successful completion of the analytical process. Interfering or potentially interfering components thus must be removed before further analysis is undertaken.

7.5.1. Suspended Particle Removal

Soil contains many very small and colloidal organic and inorganic particles. These particles are hard to separate or remove from extractants and can cause

serious interferences during analytical procedures. The standard method of separation is filtration, which can be accomplished using standard laboratory paper filter papers. Often laboratory personnel will crease filter paper with such force that small tears or holes are torn in the paper, allowing soil to pass though during filtration. In other situations the porosity of the paper may be sufficient to allow clay particles and organic matter to pass through the paper. In this case refiltering through the same paper will sometimes be sufficient to remove all particles. Sometimes two layers of the same filter paper will be sufficient. Some combination of these approaches used together should remove all particles.

Sometimes, however, none of the above mentioned procedures will be sufficient, and other filtering methods may be required. Small filtering disks that fit on a plastic syringe and have porosities of 0.22 or 0.45 μm will effectively remove particles from soil extract. These disks clog rapidly with soil, and so it is essential that the suspension be filtered through filter paper before using filtering disk, particularly if significant quantities of filtrate are needed. Either the suspension to be filtered can be pulled into the syringe, the filter disk added, and the suspension expressed out of the syringe; or the disk can be put on the syringe, the plunger removed, suspension poured into the syringe, the plunger inserted, and the suspension expressed, thereby filtering it.

An alternate procedure is to use a centrifuge to remove suspended material from soil extracts. Centrifugation is very effective in removing small particles; however, caution must be exercised when removing centrifuged suspensions from the centrifuge and centrifuge tube because any motion may resuspend particles, thus undoing the centrifugation process. The supernatant, when carefully removed, can be used directly for analysis. In many cases this is faster than filtration, especially when a large quantity of solution free of suspended material is required.

7.5.2. Sorption Cleanup Methods

There are a myriad of sorption cleanup procedures available. Some are simple, requiring little sample preparation and manipulation; others are more complex. Common cleanup materials along with their advantages and disadvantages are listed in Table 7.3. These sorption materials can be used in glass columns similar to chromatographic columns, or they may be purchased as prepared columns, termed *solid-phase extraction columns*, the use of which is simply called *solid-phase extraction* (SPE) (see discussion above).

The basic method involves passing the extract through a column containing an appropriate solid sorbing material. One of two cleanup processes can occur. The component of interest can be sorbed and thus separated from other components. It is later released by extraction and analyzed. The other process involves the component passing through the column while unwanted components are retained on the column. Both of these can be effective cleanup

Table 7.3. Some Common Extraction Cleanup Methods

Method Based on Sorbent Used	Advantages	Disadvantages
Alumina	Diffferent pH and activity ranges available for different cleanup needs	May decompose or irreversibly sorb compounds
Florisil	Cleanup of chlorinated hydrocarbons, nitrogen, and aromatic compounds	Has basic properties and may not be compatible with acids
Silica gel	Separation on the basis of differing polarity	Components with the same polarity will not be separated
Gel permeation	Components separated on the basis of size	Different components of the same size will not be separated

procedures; however, the former has the advantage of both separating the component of interest from other materials and concentrating it.

In either case materials commonly used as sorbants are stationary phases used in chromatography. Because of their common chromatographic use, these sorbants are well described in the literature and their characteristics and sorbtive capacities known. Thus all three aluminas—acid, neutral, and basic—have been used, along with silica gel, Florisil, and gel permeation for different compounds and environmental samples.

Solid-phase cleanup separates and concentrates components of interest. However, care must be taken to ensure that the capacity of the column is not exceeded. If it is, some of the component of interest will not be retained on the column and thus lost, resulting in analytical results lower that the true amounts present.

7.5.3. Water Removal

Water is always present in soil; even air dry soil contains a significant amount of water. The temptation is to remove all water before analysis, but to accomplish this, soil must be dried at a temperature above 100°C. This procedure works but causes irreversible changes in soil characteristics such that its extraction and subsequent analytical results are inaccurate, at least when compared with the results obtained on "fresh" unheated soil samples. The issue then becomes how to circumvent these problems.

The most common and standard method is to allow soil to dry naturally at room temperature or sometimes a little above room temperature (35°C maximum) before extraction or analysis. This will result in a soil sample con-

taining one to several percent water. The soil is weighed and extracted as if it were dry (see Chapter 4, Section 4.13.1). It must be kept in mind that under these conditions the extractant will pick up and in some cases become saturated with water that will need to be removed before proceeding with the analysis.

Another approach is to add an inorganic drying agent such as anhydrous sodium sulfate to the moist or wet soil. This latter approach is limited by the contact between the drying agent and the soil. Although adding a drying agent to soil will remove some water, it cannot be expected to remove all water. Thus, even in this case the extractant will contain some water after extraction and may need to be dried before analysis can be undertaken.

Because even air dry soil will have a variable amount of water, analytical results are reported on the bases of the dry weight of soil taken. This procedure is described in Chapter 4 and expressed in equations (4.3) and (4.4).

After extraction organic solvents can be dried using a number of common drying agents. Usually these are inorganic salts that react with water in a solvent and become hydrated. Not all drying agents are suitable for all solvents or solvents containing certain functional groups; thus care must be taken in selecting the drying agent. Common drying agents and their characteristics are listed in Table 7.2 [20].

Drying conditions are specific for specific extractions procedures, and thus the extraction method used will determine the drying required for the method used.

7.6. EXTRACT ANALYSIS

Extracts may be analyzed using by colorimetry, spectroscopy (Chapter 8), chromatography, or a combination of chromatography and spectroscopy (Chapter 9). Spectroscopic, chromatographic, and chromatographic/mass spectroscopic analysis will most often be carried out directly on the clean, dry extract.

7.7. CONCLUSIONS

A variety of methods, both physical and chemical, are used to separate components of interest from the soil matrix. This is usually accomplished using one of a number of different procedures, solvents, and solutions. Once isolated, the extracted component will be cleaned by one of several purification procedures that may also result in concentration. In other cases the extractant will be concentrated after the cleanup process is completed. In many instances one of the most important contaminants to remove will be water. Once cleaned, the extract may be analyzed by a wide variety of spectrophotometric and chromatographic methods.

PROBLEMS

7.1. What type of extractant would you chose to extract the following compounds from soil? Explain the rationale for your choice.
(a) LiCl
(b) CH_3CH_2OH
(c) $CH_3(CH_2)_{10}CH_3$

7.2. List the different "grades" of water available and describe how they are different and what residual materials they might contain.

7.3. A 100-g soil sample from the field was dried overnight at 105°C and later was found to have a weight of 95 g. Calculate the percent moisture on a dry-weight bases [see Chapter 4, equations (4.3) and (4.4)].

7.4. A researcher develops an extraction–cleanup procedure using a sandy soil. The procedure works well and is reported in the literature. Another researcher uses the same procedure on a clayey soil and an organic soil and obtains very poor, inconsistent results. What factors might account for these results? (You might want to use information from previous chapters to help answer this question.)

7.5. Describe in detail two extraction procedures applied to soil. (You might want to consult some of the references in the bibliography to answer this question.)

7.6. Describe two cleanup methods and list the advantages and disadvantages of each.

7.7. Explain why drying soil is both an important and a difficult procedure. Explain why all nonaqueous soil extracts may need to be dried. (You will need to consult previous chapters to answer this question.)

7.8. Can solid-phase extraction be used to concentrate an extract?

7.9. Using alcohol in ether as an example, explain why the stabilizers added to solvents may cause a problem in an extraction procedure.

7.10. What are the similarities between hexanes and petroleum ether? How do their characteristics affect chromatographic analysis? (You might want to consult Chapter 9 in answering this question.)

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CHAPTER

8

SPECTROSCOPY

The electromagnetic spectrum is a continuum of wavelengths λ (it can also be expressed as frequency; the reciprocal of wavelength and in electron volts). Within this spectrum visible light represents a very small part: the region of wavelengths around 10^{-5} cm. All regions of the electromagnetic spectrum have been used to analyze environmental constituents, including those in soil. In some cases the radiation is passed through the material being investigated and the absorbed frequents related to the components it contains. In other cases radiation is reflected or refracted from the sample and information is gathered as a result of changes in the radiation occurring during reflection or refraction.

How electromagnetic radiation is used to investigate the characteristics of a sample depends on how it interacts with matter. X rays are diffracted by atoms and thus are used to elucidate the arrangement of atoms in a material, commonly crystals. Absorption of ultraviolet and visible light results in the movement of electrons from one orbital to another. Electrons in double or triple bonds and nonbonding electron pairs are most subject to this type of interaction. Infrared radiation interacts with matter by changing its vibrational and rotational modes. When atoms are placed in a strong magnetic field, the spins of their electrons and protons interact with radiofrequency (RF), wavelengths. These methods, namely nuclear magnetic resonance (NMR) spectroscopy and electron paramagnetic resonance (ESR) spectroscopy, are used to determine the environment of nuclei and electrons.

The movement of electrons between orbitals in atoms can also be used to gain information about the elements. Energy from an outside source, such as heat from flames, a furnace, an electrical arc, plasma; ultraviolet or visible light; and X-rays or gamma rays, can move electrons from one orbital to another. In this process light is absorbed, and the absorbed wavelength can be used to determine the type and amount of element present. The other possibility is that the excited electron will fall back to its original position, emitting a wavelength of light specific for that particular transition. Measuring the wavelength and the amount of light provides information as to the kind and amount of that element present. The method of exciting the electrons varies with different types of instrumentation, but the basic process is the same. This technique,

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

although technically applicable to all elements, is most sensitive and therefore most commonly used in the detection and quantification of metals.

All analytical methods that use some part of the electromagnetic spectrum have evolved into many highly specialized areas of use and ways of extracting information. The interaction of X rays with matter represents an excellent example of this diversity. In addition to straightforward X-ray absorption, diffraction, and fluorescence, there are a wide host of other techniques that are either directly X-ray-related or come about as a secondary result of X-ray interaction with matter such as X-ray photoemission spectroscopy (XPS), surface-extended X-ray absorption fine-structure spectroscopy (SEXAFS), Auger electron spectroscopy (AES), and time-resolved X-ray diffraction techniques, to name only a few [1,2].

No attempt will be made to thoroughly investigate all these X-ray or other specialized spectroscopic techniques. Only the main, common, or routine methods and instrumentation used in soil analysis will be discussed.

8.1. SPECTRAL OVERLAP

Spectra can be obtained as either absorption, where the material of interest absorbs defined wavelengths of radiation, or emission spectra, where the material of interest emits definite wavelengths of radiation. All compounds and elements absorb and emit numerous wavelengths or bands of electromagnetic radiation. If two compounds or elements are placed in a beam of electromagnetic radiation, all the adsorptions of both compounds will be observed. Or if the same mixture is excited, all bands emitted from both compounds will be present in the emission spectrum. This leads to the possibility of three types of interference: two different compounds absorbing in the same place in the spectrum, two different compounds emitting in the same place in the spectrum, and one compound emitting while another is absorbing in the same place in the spectrum. This latter case, however, is usually only observed in atomic and X-ray spectroscopy. One or more of these types of interference can be present in an analytical procedure, especially when applied to soil and soil extracts.

In situations where all the components of a mixture are known and all their characteristics fully understood, correction for any or all of the types of interference described above can be made. However, in soil and soil extracts it is generally impossible to identify all the components and fully understand all their characteristics. Thus, it is especially important to rule out, compensate for, or eliminate all possible types of interference when carrying out a soil analysis. This is generally accomplished by isolation of the component of interest from most, if not all, of the soil matrix components with which it is associated (see Chapter 7) [3].

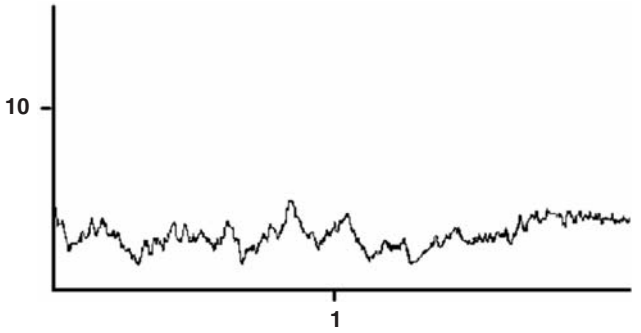

Figure 8.1. Baseline noise from a gas chromatograph.

8.2. NOISE

Every measurement has noise—random changes in the results measured; that is, if an instrument is left to make measurement without any sample, the baseline will not be a straight line but will be a random recording of instrument output. Figure 8.1 shows the noise in the baseline of a gas chromatograph at maximum sensitivity. When an absorption or peak is vastly greater than the noise, there is little question of its authenticity. When it is not much greater than the noise, the issue becomes whether it is real or is noise.

There are two ways to approach this issue, and both should be investigated in any questionable measurement. First, it is often assumed that any absorption or other measurement that is 3 or 4 times larger than the noise is real. This is a good start; however, there are other more scientific approaches to this problem. If repeated measurements on different subsamples or aliquots produce the same absorption or measurement, then it is probably a real result and not noise. On the other hand, if on repeated measurement adsorptions in a spectrum occur in exactly the same location and have exactly the same characteristics such as shape and area under the peak, then they are probably not due to the sample because some variation in measurement always occurs. This type of problem is usually a result of instrument malfunction, which must be investigated.

8.3. X-RAY DIFFRACTION

X-ray diffraction is a very powerful tool used extensively to identify the crystalline clay minerals in soil. It is also used to study the characteristics of the clay minerals in terms of shrink–swell characteristics and occluded components. It is not, however, applicable to amorphous clays found in some tropical soils and common in Andisols. It is carried out by irradiating, at various

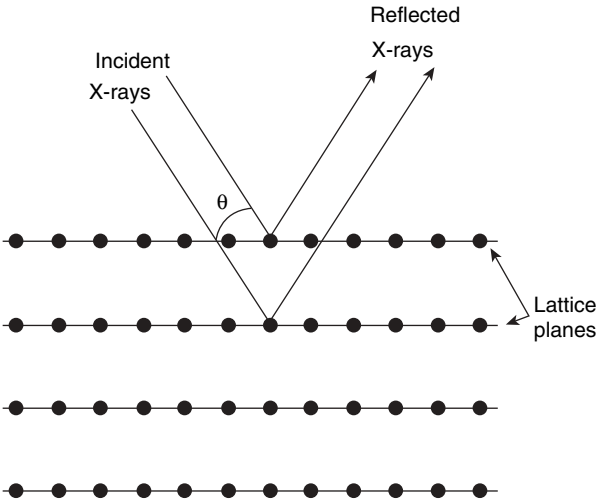

Figure 8.2. The diffraction of X rays from crystal planes.

incident angles, a soil clay sample. When the incident angle θ, shown in Figure 8.2, results in constructive interference of the reflected X rays the distance between the layers can be calculated using Bragg's law (see the text by Atkins and de Paula [4] for an explanation of Bragg's law). In addition to the distance between lattice planes, the swelling characteristics of various clays are different when they are exposed to different solvents. This provides additional information about the type of clay present and its characteristics.

In practice, clay from a soil sample is prepared on a microscope slide and dried and its X-ray diffraction measured; subsequently the same clay is placed in atmospheres saturated with, for example, glycerol, with subsequent X-ray diffraction and again the distance between layers determined. Changes in the diffraction pattern, or the lack thereof, will help identify the type of clay present [5].

8.4. X-RAY FLUORESCENCE

Exposure of elements to a broad spectrum of X rays results in the ejection of electrons from their inner shells. Electrons from outer shells falling into these vacancies emit radiation, an X-ray photon, of specific wavelength (see Figure 8.3). Analysis of this radiation, X-ray fluorescence (XRF), allows for the identification of the element from which the photon is emitted. Instruments for carrying out this analysis can be either laboratory-size or a handheld unit that can be taken to the field. The excitation radiation for the X-ray fluorescence instrument must pass through a window. The window material,

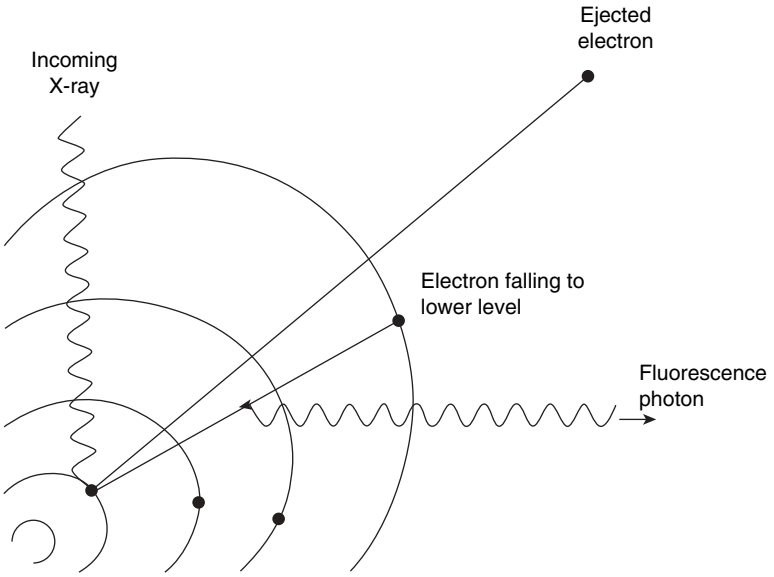

Figure 8.3. Diagram showing the source of X-ray fluorescence photons.

often a metal, will, in part, determine the range of elements that can be detected.

When used in direct soil analysis, X-ray fluorescence suffers from the fact that it is largely a surface phenomenon. For this reason, only surface elements will be determined. However, because it is a surface phenomenon, it has been extensively used to study sorption on the surfaces of soil components. An extensive list of investigations using XRF to investigate various sorption mechanisms is given in the text by Sparks [6].

Other characteristics of XRF that can limit its usefulness are the surface area observed and surface contamination. In XRF the surface area measured is small, meaning that a large number of determinations must be made in order to obtain a representative sample of the elements present. In addition, transport and storage of uncovered soil samples can lead to surface contamination that will subsequently appear as part of the soil constituents.

The sensitivity of X-ray fluorescence determinations is better for elements with higher mass number and less for lighter elements. Often in soil analysis lighter elements are of greater interest, and this makes application of this method more difficult. Typically determination of elemental composition in the part per million (ppm) range is, however, achievable.

Fluorescence determinations are best made on samples of homogeneous particle size, and this is not the normal state of soil. Grinding and carefully sieving soil before analysis can minimize problems associated with particle size heterogeneity. Another approach has been to fuse soil with borate or dilute it

with cellulose or another suitable diluent. Very thin layers of soil may also be prepared and used for quantitative analysis.

Other limitations involve both the mass absorption coefficient of soil components and secondary and tertiary excitation. The mass absorption coefficient can be calculated and used to correct fluorescence determinations if the exact composition of the material being analyzed is known. This is not possible in soil. Secondary or tertiary excitation occurs when X rays emitted by an element other than the one of interest cause emission of fluorescence of the element of interest. These potential sources of error are possible in any soil analysis using X-ray fluorescence.

Some of the these limitations, but not all, can be overcome by making an extract of the soil and determination of the elemental composition of the extract. This approach can eliminate or minimize problems associated with the limitation of X-ray fluorescence analysis to a particle surface [7,8].

8.5. ATOMIC SPECTROSCOPY

Excitation of electrons in an atom promotes them to a higher energy level, and when they fall back to their original level, they release energy of the same wavelength as the energy absorbed. When this energy is in the visible range of the electromagnetic spectrum, it gives rise to what is termed a *line spectrum*, which consists of discrete wavelengths, or lines of light unique to each element that are absorbed or given off. Early chemists used these unique lines to identify new elements as they were discovered. Samples of a new (or thought to be new) element were put in an electrical arc and the light emitted dispersed using prisms and recorded using photographic film. This is the original spectrometer.

Although such instruments as described above are available, they are not typically used in soil analysis. Today samples are most often aspirated into a flame or torch and the diagnostic wavelength detected and quantified by photomultipliers. Modern spectrometers are different because of the use of many different methods of heating samples and the range of wavelengths available. Today, because of increased sensitivity of instrumentation and detectors, more of the spectrum is available for this type of analysis, typically both the visible and the ultraviolet regions. Thus wavelengths ranging from 200 to 900 nm commonly can be used for the analysis of elements present.

The basic idea of exciting electrons and isolating unique, diagnostic, wavelengths and measuring the amount of light emitted is useful for routine measurement of soil. This is called the *emission mode* (EM). Potassium, one of the three most important plant nutrients, and sodium, which poses problems in some arid soils, along with calcium are easily and routinely measured using the EM mode, which is also commonly used to determine the concentration of these elements in blood. An instrument capable of determining elements in both the EM and atomic absorption (AA) modes is shown in Figure 8.4. Using

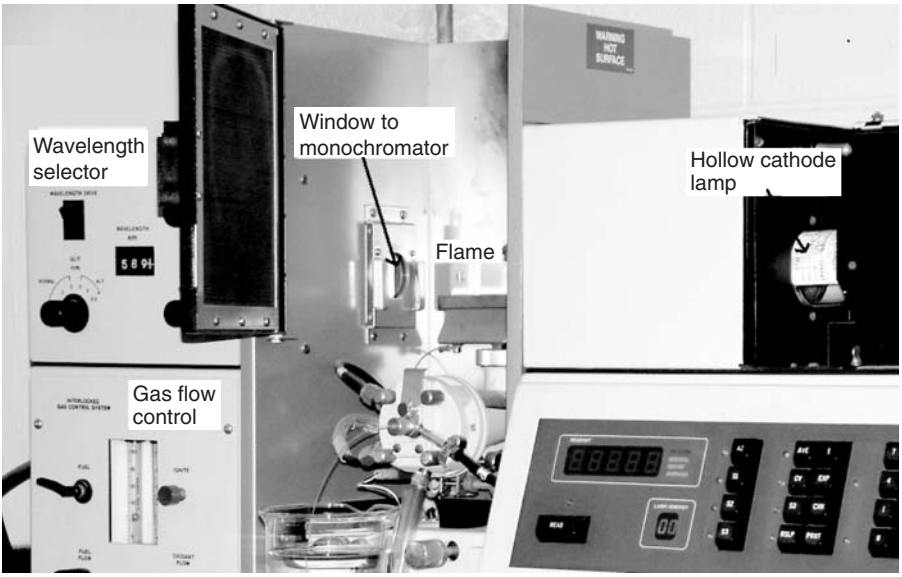

Figure 8.4. An atomic absorption spectrophotometer capable of functioning in either the EM or AA mode.

this instrument, the composition of many samples can be determined very quickly.

The analysis of a well-characterized sample by atomic spectroscopic analysis is straightforward. The instrument is adjusted to isolate and direct the analytical wavelength of light to the photomultiplier. It can be adjusted for maximum sensitivity by introducing a known sample of the element of interest into the flame and adjusting the instrument, including the flame, to maximize the reading obtained. The sample is then introduced into the flame, and the amount of light in the analytical wavelength emitted is measured and recorded. A standard curve (see Section 8.8) is prepared and used to determine the amount of element in the samples.

Even when soils are from the same area, significant variations in concentration and content are found. This leads to the possibility of diagnostic wavelength overlap. Even though each element has a unique spectrum when it is taken alone, two elements may produce wavelengths of light that are the same or very close together. This can lead to the overdetermination of the quantity of an element because the total amount of light measured is partially from the desired element and partially from another element.

Matrix interferences can be observed in a number of different forms. Components in the extract may interfere with excitation of electrons. Absorbence of light by unexpected, metals or organic compounds, either generally or specifically, would be another example. Another would be complexation with

extract components such that the metal is protected from the heat source. Other interferences such as changes in the viscosity of the extract that can affect the accuracy of analysis are also possible, although they are less common.

Whenever soil samples are being analyzed by any atomic spectroscopic method, it is essential to make sure that no interfering, overlapping wavelength elements are present in the soils. If they are then steps must be taken to correct for these interferences.

8.5.1. Excitation for Atomic Emission

For the alkali-earth metals, as noted above, a simple flame of almost any type can be used to excite the metals. However, in order to determine a wide range of metals, it is common to use either an acetylene-air or acetylene-nitrous oxide flame as the source of energy to excite the atoms. The burner is long with a slot in the top and produces a long narrow flame that is situated end-on-end to the optics receiving the emitted light.

Light given off by the excited electrons falling back to ground state is passed through slits, isolated using a grating, adjusted to the analytical wavelength, and the amount of light is measured using a photomultiplier or other light detecting device. The wavelength of the light specifies the element present while its intensity is directly related to the amount present.

Introduction of sample into the flame is accomplished by Bernoulli's principle. A capillary tube is attached to the burner head such that gases entering the burner will create suction. The sample is thus aspirated into the burner, mixed with the gases, and passed into the flame. Heating in the flame excites electrons that emit light of distinctive wavelengths as they fall back to their original positions in the elements' orbitals. Because a number of electrons can be excited and there are a number of orbitals into which they can fall, each element emits a number of different wavelengths of light particular to that element. In the case of atomic emission, one of these, usually the most prominent or strongest, is chosen as the primary analytical wavelength.

Generally the higher the temperature of the sample, the more sensitive will be the analysis. Thus, in addition to the two types of flames discussed above, a third excitation source, which is not a flame but a plasma from an inductively coupled plasma (ICP) torch, is used (see Figure 8.5). Argon support gas is seeded with free electrons that interact with a high-frequency magnetic field of an induction coil gaining energy and ionizing argon. The reversing magnetic field causes collisions that produce more ions and intense thermal energy resulting in high-temperature plasma into which the sample is introduced.

While the first two flames are used both for emission and absorption spectroscopy, ICP is used for emission spectroscopy. The three are arranged in order of increasing temperature. Both acetylene—air and acetylene—nitrous oxide can be used in the same instrument and the flames can be adjusted to be oxidizing or reducing to allow for increased sensitivity for the element

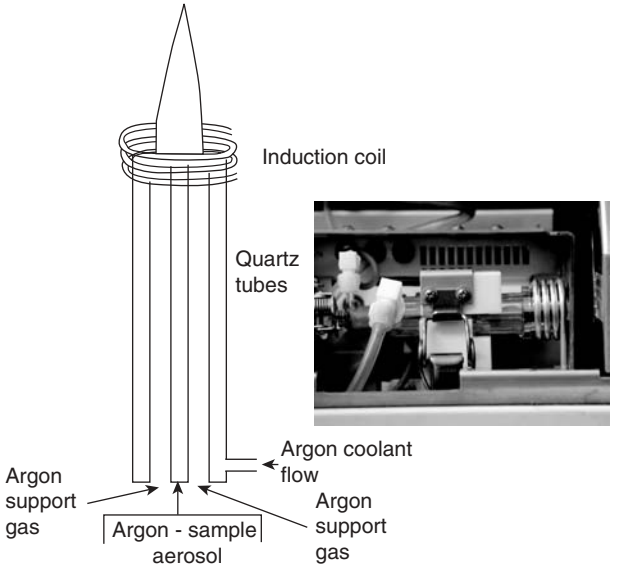

Figure 8.5. Left is a diagram and on the rights is a photograph of an ICP torch.

being analyzed. ICP is carried out using a separate instrument and generally has a significantly higher sensitivity than do flame instruments. There are a number of hyphenated variations on the basic EM and ICP instrumentation such as ICP-OES [(ICP optical emission spectroscopy); also sometimes referred to as ICP-AES (atomic absorption spectroscopy)] and ICP-MS (ICP–mass spectroscopy).

AA and ICP instruments can be equipped with multiple detectors so that analyses for more than one element at a time can be accomplished [3,9–13].

8.5.2. Atomic Absorption

In atomic absorption light emitted by the element of interest, from a hollow cathode lamp (HCL), is passed through the flame of an atomic absorption spectrometer (the same instrument is used for EM except now configured for the AA mode). In this case the same burner and flame as described above and shown in Figure 8.4 is used. The source of the light is a hollow cathode lamp, also shown in Figure 8.4, where the cathode is made of the element of interest and thus, when excited, emits the analytical wavelengths of light needed for the analysis of that element. In a majority of cases a different lamp will be needed for each element for which an analysis is required. The amount of light absorbed by the element in the flame is directly proportional to the amount of that element present. Atomic absorption is significantly more sensitive than flame emission for most metals [14,15].

8.6. ULTRAVIOLET AND VISIBLE SPECTROSCOPY

Information about a soil extract can be obtained from the ultraviolet and visible regions of the spectrum. These are wavelengths ranging within 190–400 and 400–900 nm, respectively (wider ranges of wavelengths are available on some instruments). The two regions of the spectrum are used very differently in soil analysis, although they are commonly found together in UV–Vis (ultraviolet–visible) spectrometers. The ultraviolet region can be used to obtain spectra of soil extracts; however, it is most commonly used as a detector for high-performance liquid chromatograph (HPLC). The visible region is used for colorimetric analysis, discussed below.

Common solvents, such as water, acetonitirile, and heptane, are transparent over both the ultraviolet and visible regions. Because these represent both hydrophilic and hydrophobic solvents, a very broad range of compounds can be easily analyzed by UV.

Adsorption in both regions involves promotion of electrons in double or triple bonds or nonbonding pairs of electrons in elements such as those on oxygen or nitrogen, to higher energy levels. Conjugated double bonds and nonbonding pairs conjugated with double bonds absorb at longer wavelengths than do isolated double bonds or nonbonding electron pairs. The more highly conjugated a system the longer the wavelength of the maximum absorption called the *lambda max* (λ_{max}) (see Figure 8.6). This means that all aromatic compounds have strong ultraviolet adsorption bands as well as terpenes and other conjugated systems. Unconjugated aldehydes, ketones, amides, acids, esters, and similar compounds also have ultraviolet absorptions, although they are not as strong.

Any extract of soil containing large amounts of organic matter may have multiple absorption bands in the UV–Vis region of the spectrum. A UV–Vis spectrum of a simple aqueous, low-organic-matter, soil extract is shown in Figure 8.6. As seen in this spectrum, even a simple soil extract can have significant absorptions that can mask other potential absorptions of interest. Absorptions can also result in interferences with an ultraviolet and/or visible determination of a component or the derivative of a component.

An interesting application of this region of the spectrum is the determination of nitrate and nitrite in soil extracts. Nitrate is very soluble in water and can be extracted using a simple water extraction procedure. Nitrate absorbs at 210 nm and nitrite are 355 nm and can be quantified using these absorption maxima (λ_{max}). Other materials extracted from soil as noted above, however, can obscure this region, and thus caution must be exercised to determine whether there are any interfering components in the extract [11,12,15–17].

8.6.1. Ultraviolet Sample Preparation

Samples are analyzed dissolved in a solvent, commonly acetonitrile, heptane, hexadecane, or water. A scan of the pure solvent in the sample cell, typically made of quartz (*A* in Figure 8.7), is first obtained. The dissolved sample is

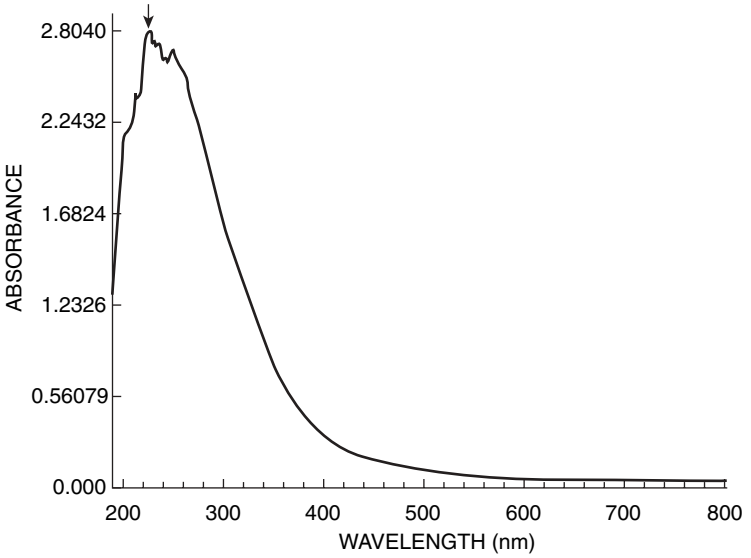

Figure 8.6. UV–Vis spectrum of simple deionized water extract of soil. Absorption maximum is indicated by an arrow. Note that the absorbance is above 1, and therefore the solution should be diluted before attempting to interpret the spectrum.

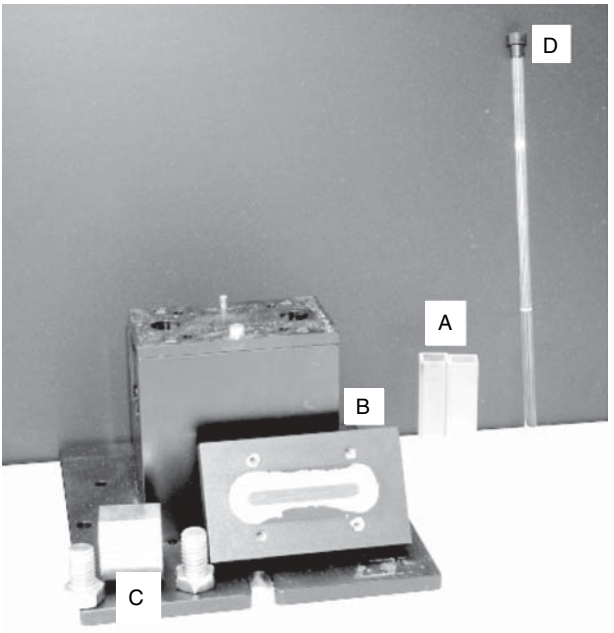

Figure 8.7. Sample holders for various spectrophotometric measurements: (*A*) UV–Vis cells; (*B*) infrared attenuated total reflection (ATR) plate and stand—plate is attached to stand behind it when in use; (*C*) simple KBr pellet maker using the two bolts and the center dye; (*D*) a sample tube for NMR spectroscopy.

placed in the sample cell, which is placed in the sample compartment of the instrument and a spectrum obtained. Many compounds have large molar absorptivities,[1] and thus only small amounts are needed to obtain a spectrum. Most UV–Vis spectrophotometers function best between 0 and 1 absorbence. Often a sample will have an absorbence above 1, as is the case illustrated in Figure 8.6. In these cases it is usually most useful to dilute the sample by a factor of 10 to bring the absorption below 1 before using the absorbence data.

8.7. THE VISIBLE REGION

The visible region of the spectrum, with wavelengths 400 nm (violet) and 900 nm, is used extensively in soil analysis in the colorimetric determination of components extracted from soil (see Section 8.8). Once extraction is complete and the extract has been filtered and otherwise cleaned as needed, it is analyzed for the components of interest by treating it with a reagent to produce a colored product. The amount of color is directly related to the amount of component present.

An excellent example of this type of analysis involves the determination of phosphate in soil extracts. Soil is extracted with an appropriate extractant and added to a solution of acid molybdate, with which the phosphate reacts to produce a purple or blue, phosphomolybdate solution. Standard phosphate solutions are prepared and reacted with acid molybdate, and the intensity of the phosphomolybdate produced is measured. A standard of calibration curve is prepared from which the intensity of the color is directly related to the concentration of phosphate in the extract.

For this type of analysis to be accurate, three characteristics must exist:

1. The soil extract must not contain any components that absorb light at the same wavelength as the phosphomolybdate; this includes suspended material that will refract light. Refracted light does not get into the detector and so is recorded as being absorbed by the sample, thus giving an inaccurate result.
2. The color producing reagent must not react with any other commonly occurring component in the extract to form a similarly colored product.
3. The soil must not contain any compound that inhibits or interferes with the production of the colored compound.

As described in previous chapters, soil contains many different inorganic and organic elements, ions, and compounds plus both inorganic and organic colloids. Thus it cannot be assumed that the soil being investigated does not contain any of the types of interference mentioned above. Some soils high in

[1] *Absorbtivity* is defined as $A = \varepsilon bC$, where A = absorbance, ε = molar absorption coefficient (liter mol^{-1} cm^{-1}), b = pathlength of radiation through sample (cm), and C = molar concentration.

organic matter may have dark-colored components that are released into the extracting solution and cause interference with the spectrophotometric analysis. Thus extracts must be analyzed to make sure that they are free of interference.

8.8. COLOR MEASUREMENT: THE SPECTROPHOTOMETER

Color measurement is based on the Beer–Lambert law, which can be expressed as follows:

$$A = \log_{10} \frac{I}{I_{ex}}$$

In this equation A is the absorbence also called the optical density, I is the intensity of the bean of light entering the sample, and I_{ex} is the intensity of the beam of light exiting the sample. The absorbance is proportional to both the concentration of absorbing species and the pathlength of the light through the sample. Normally standard-size sample cells are used, and so the pathlength is constant for all samples.

This type of analysis involves colored solutions; thus the spectrophotometer need function only in the visible range of the spectrum. A visible light source is used to provide light that passes through the sample in a cuvette, to a grating where it is dispersed into its respective wavelengths. The analytical wavelength, that is, the wavelength absorbed by the colored compound, is isolated and directed to the detector, where the amount of light at this wavelength is measured and displayed on the readout. A diagram of a spectrometer is shown in Figure 8.8. Note that there are many different ways to design a spectrophotometer and its light path. The diagram given is simply a general representation of how a spectrophotometer works.

Cuvettes used with the spectrophotometer are not simply test tubes. They are specially made tubes and are often matched such that a set of tubes will all have the same absorbence characteristics. Cuvettes should never be used as test tubes; they must be kept clean at all times, and care must be taken not

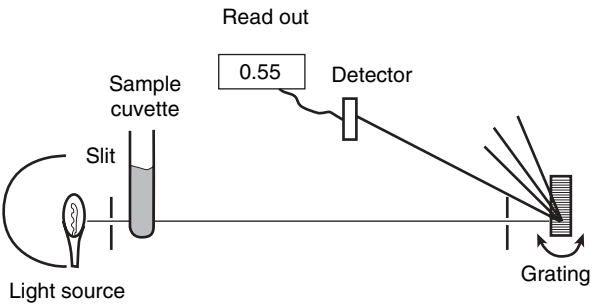

Figure 8.8. Diagram of the light path in a spectrophotometer.

to scratch them. When using cuvettes that have not been used before, they should be tested to make sure that they are all the same. This is accomplished by inserting them into the spectrophotometer and noting their absorbence. All should be the same. Keep in mind that empty cuvettes will have a higher absorbence than when filled with water. This is because light is refracted at each surface and when filled with water there is less refraction at the surfaces.

8.8.1. Zeroing and Blanks

When using a spectrophotometer for a colorimetric analysis, both the 0 (∞A) and 100% (0A) readings must be set. Once the instrument has warmed up, with nothing in the sample compartment, the readout is set to 0. A blank, a solution containing all the components used in the analysis except the analyte being measured, is placed in a cuvette and placed in the sample compartment, and the instrument is adjusted to 100%. This procedure is intended to account for all interferences that may be introduced into the measurement by components other than the analyte of interest. Once the instrument is adjusted, determination of the absorbence of standards and samples can be made.

8.8.2. Relating Component Concentration to the Original Sample

Performing an analysis requires the preparation of a standard or calibration curve. A series of standard solutions, a minimum of three, containing known amounts of the component of interest are prepared. There are two primary restrictions on these solutions: (1) they should be over the range of the expected concentrations of the component of interest and (2) all results for the extracted component must be higher than the lowest standard solution and lower than the highest standard. If a result is beyond these limits, its concentration cannot be determined. In the table of data for Figure 8.9, Unk1 (unknown 1) is 0.05, which is below the lowest standard, specifically, 0.15, and so its value cannot be determined. Likewise, the Unk2 value is 0.97, which is above the highest standard and also cannot be determined.

If, as with Unk 2, the value is above the highest standard, then the sample may be diluted and retested. In this case the concentration must be corrected for the amount of dilution. Similarly, in some cases when the concentration of an unknown is lower than the lowest standard, the solution can be concentrated and reanalyzed.

Furthermore, standards must be over a range where there is a direct or straight-line relationship between the amount of color produced and the amount of component present. It is common for data points beyond the standard curve to be part of a different straight line or simply not measurable because the solution is too light or too dark. In Figure 8.9 the distance between points 0,0 and 10,0.15 may form a line very different from the one shown in the graph. We know nothing about this region. The same can be said about the region above points 90,0.95. With many instruments very low absorbances are

Concentration	Optical Density
10	0.15
30	0.3
60	0.65
90	0.95
Unk1	0.05
Unk2	0.97

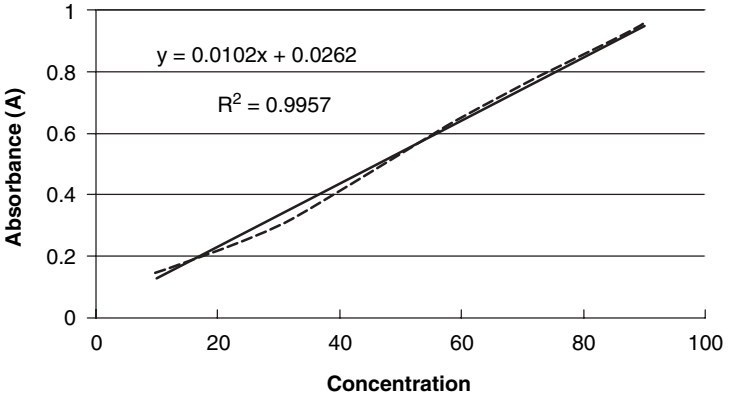

Figure 8.9. Standard or calibration curve for data at top.

easily measured; however, absorbences approaching or above 1 are seldom reliable.

A blank is often used as a 0,0 point in preparing a calibration curve as in Figure 8.10. This is an acceptable practice as long as the calibration curve is sufficiently accurate. If it is not, or if the inclusion of the 0,0 point produces an irregular curve, its removal from the curve is warranted. In Figure 8.10 the inclusion of the 0,0 point gives a slightly better r^2 value (see Section 8.9) and thus can be included in the curve. Also note that when this is done, the Unk1 value can be determined because the number is within the range of the standards used.

8.9. REGRESSION ANALYSIS

The calibration curves in Figures 8.9 and 8.10 were prepared using Excel (*xy* scatter chart), which has automatic features for adding a trend line (the straight line), calculating an equation for the line, and calculating r^2. The r^2 is particularly useful in that it tells how close the line obtained is to a straight line. If the data produced an absolutely straight line, the r^2 would be 1. When

Data

Concentration	Optical Density
0	0
10	0.15
30	0.3
60	0.65
90	0.95

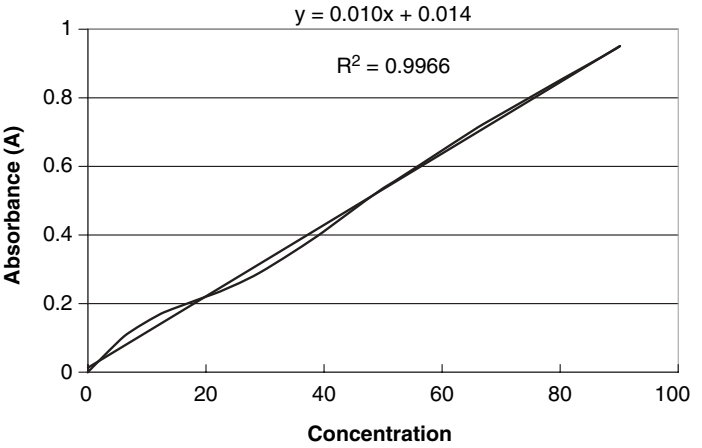

Figure 8.10. Calibration curve including 0,0 as a point.

it is less than 1, this means that some of the error in measurement has not been accounted for. In Figure 8.9 the r^2 is 0.9957 and can be interpreted as accounting for all the error except for 0.0043. This can also be expressed as a percentage. Thus the r^2 is 99.57%, and we have accounted for all except 0.43% or the error.[2]

Regression analysis, sometimes referred to as *least-squares analysis*, is a standard statistical analysis, which is available in statistical packages as well as Excel, and statistics text books [19]. In all analytical work using calibration curves an r^2 of 0.99 or higher is essential.

Once the calibration curve has been prepared and is of sufficient accuracy, the extracted samples can be analyzed and the calibration curve used to relate the results of the analysis of the unknown to the amount of component of interest present in them [19].

[2] Strictly speaking, we have used more significant figures than are warranted by the original data.

8.10. RELATIONSHIP TO ORIGINAL SAMPLE

At this point the original sample has been extracted, and the amount of component of interest has been determined. But it has been determined for only a portion of the extract and sample or area sampled. It is essential to relate this amount back to the amount originally present in the sample taken in the field and ultimately the field itself. This is then a process of working backward. Thus, one must first find the total amount present in the whole extract and then determine how this relates to the amount of sample originally taken and then to the field sampled.

If a 1-g soil sample is extracted with 10 mL of extractant, then the component extracted is evenly distributed throughout the 10 mL. This means that the final result will need to be multiplied by 10 because the component was diluted to 1 : 10. This then is related back to the volume or mass of soil in the original sample. It may also be necessary to apply other conversion or correction factors, such as the percent water present, depending on the procedure used.

8.11. INFRARED SPECTROSCOPY

The next portion of the electromagnetic spectrum most often used for analysis is the midinfrared region. In the infrared spectroscopy region, changes in the vibration, bending, and rotation of bonds in molecules results in the absorption of radiation. These changes from one mode to another higher-energy mode are caused by the absorption of infrared light.

In infrared spectroscopy the wavelengths become long, and so the spectrum is usually reported in reciprocal centimeters, which is a measure of frequency rather than wavelength. In the older literature this region will be from 4000 to 250 cm^{-1} (2.5–50 μm). Today the infrared region is divided into three distinct regions[3]; the near infrared (NIR), 1000–4000 nm; the midinfrared (MIR), 4000–250 cm^{-1}; and the far infrared (FIR), less than 250 cm^{-1}. Instrumentation for both the NIR and FIR regions is not as readily available as that for the MIR, and so these regions are not as commonly used. However, there has been some substantial development of NIR for use in the determination of components in soil, food, and feed such as oils and protein [20,21].

Generally, infrared spectroscopy, infrared spectrometers, accessories, and water do not mix, although there are techniques for sampling aqueous systems for IR analysis. Salts of alkali and alkaline-earth metals and halogens, such as sodium chloride and potassium bromide, are transparent to MIR and are used in the optical systems of spectrometers. Large amounts of water will dissolve parts of the optical system, rendering it inoperable. Small amounts, such as in high-humidity air, will lead to optical components being fogged and thus less transparent or opaque. These same salts are used for sample cells, sample preparation, and analysis, and thus samples containing water will be deleterious or destroy cells. Water has many strong broad adsorptions in the MIR, and

[3] These are typical instrument ranges.

these make analysis of some components mixed with water difficult or impossible.

Infrared spectroscopy is most frequently used for the identification of organic compounds. Some organic functional groups, particularly alcohol, acid, carbonyl, double bonds, triple bonds, and amines, have unique, strong, and easily identifiable absorptions in the IR spectrum. Both methyl and methylene groups are insensitive to their environments and absorb strongly in narrow frequency ranges, making them easily identifiable and useful for quantification (see discussion of hydrocarbon analysis below). However, since these groups are almost always present in organic compounds, their absorptions are seldom useful in identifying specific compounds. Carbonyl groups also have a strong, sharp absorption that is usually the strongest in the spectrum, which also makes their absorption behavior useful for identification.

A correlation chart giving the important and unique absorptions of various organic functionalities can be found in many books on infrared spectroscopy. These charts are useful, but the user needs to be familiar with the shape and size of typical functional group absorptions before using them with any degree of accuracy. A broad absorption between 2500 and 3650 cm^{-1} shows the presence of —OH. However, the absorption of alcohol —OH is very different from that of acid, as can be seen in Figure 8.11. Another example is the carbonyl absorption, which occurs in the 1700 cm^{-1} region of the spectrum. It is important to know not only that these are sharp absorptions but also that they are the strongest absorptions in the spectrum of aldehydes, ketones, and acids as seen in Figure 8.11. If there is an absorption in this region but it is not the strongest absorption in the entire spectrum or is not sharp, then it is not from a typical aldehyde, ketone, or acid but may be from a double bond, although this is not common. Table 8.1 lists some important functional group absorptions for the MIR region of the spectrum.

Infrared is not typically used in common soil analytical procedures, although it has been used to investigate various soil components, the most common of which are humus and its various subcomponents (see Figure 8.12). It has also been used to identify soil clays, both crystalline and amorphous. Spectra can be accessed on the Web at `http://speclab.cr.usgs.gov` particularly see `splib04a`.

There is one case, in environmental work, where the methyl and methylene absorptions are both useful and used. This is in the USEPA method for the determination of total recoverable petroleum hydrocarbons (TRPHs) in a soil or other extract. Here a supercritical carbon dioxide extraction of a hydrocarbon-contaminated soil is made (USEPA Method 3560) (see also Chapter 7). The extracted hydrocarbons are dissolved in tetrachloroethylene, which is transparent in the region of the IR spectrum where methyl and methylene groups absorb. The infrared —CH_3, —CH_2—, absorptions in the 2800–3000 cm^{-1} region are related to the amount of hydrocarbons in the soil extract through standard curves constructed, as described above, for the method [20–26].

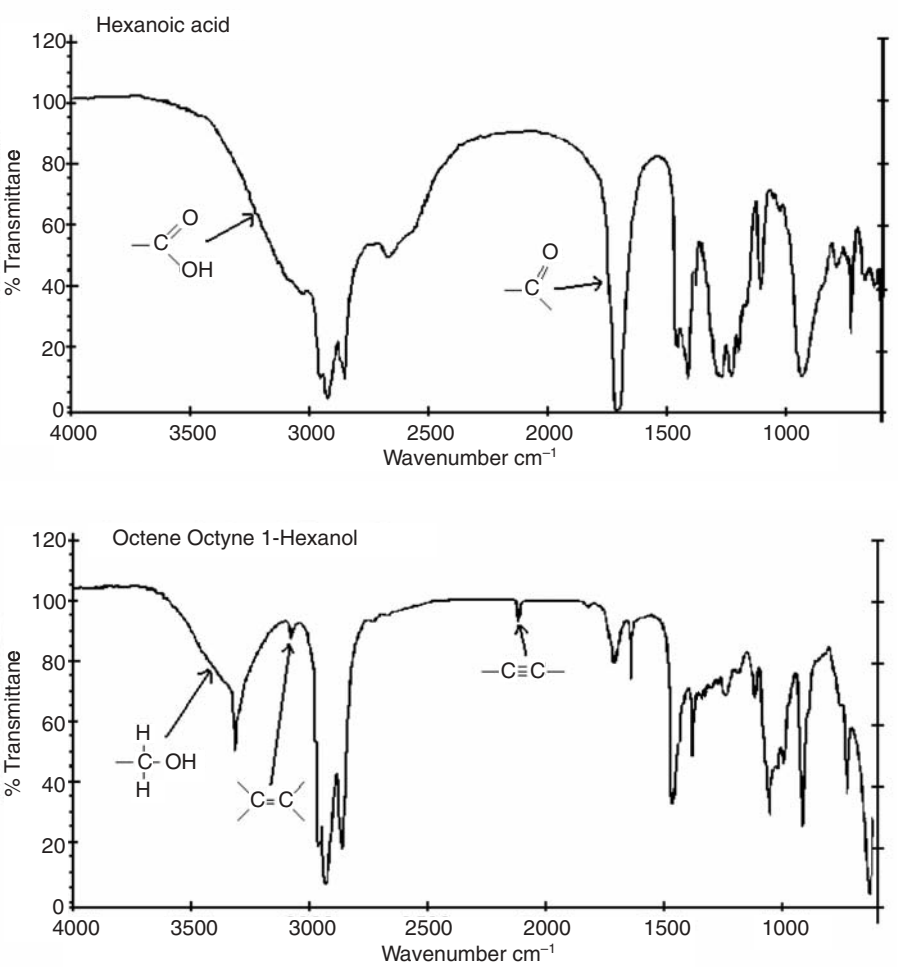

Figure 8.11. Infrared spectra showing common absorption. Both the OH of acids and alcohols are hydrogen-bonded, lending to broadening of the absorption. Note that the OH absorption of hexanol is unusually sharp for a hydrogen-bonded alcohol.

8.11.1. Infrared Sample Preparation

All compounds have absorptions in the midinfrared region of the spectrum. Most common solvents contain groups that absorb in this region of the spectrum, which are important for identification of unknowns and thus are not used. Carbon tetrachloride, carbon disulfide, and mineral oil (often called *nujol*) are exceptions, in spite of the fact that all three have significant infrared absorbences. Mineral oil, for example, has strong absorbences due to —CH_3, —CH_2— in its molecules. This obscures regions that may be of analytical

Table 8.1. Important Diagnostic Absorptions of Major Organic Functionalities in Mid-Infrared, ^{1}H NMR, and ^{13}C Spectra

Functionality	Mid-Infrared (cm^{-1})	^{1}H ppm	^{13}C ppm
—**CH_3**	3000–2900 and 2925–2825 **S–N**	0–1 **S**	0–40 **S**
—**CH_2**—	2950–2850 and 2875–2800 **S–N**	1–2 **S**	10–50 **S**
H–**C**=**C**–**H**	3100–3000 **W–N**	5–7 **W–N**	100–170 **S**
—**C**≡**C**–**H**	2250–2100 **W–N**	2.5–3.1 **W–N**	60–90 **S**
—**C**–**OH**	3500–3100 **B–S**	0.5–6.0 **S**	50–90 **S**
—**C**(=**O**)–**H**	2700–2900 (H) **W–N** 1650–1750 (C=O) **S–N**	9–10 **W**	190–220 **S**
—**C**(=**O**)—	1650–1750 (C=O) **S–N**	**NA***	190–220 **S**
—**C**(=**O**)–**OH**	3300–2900 (—OH) **B–N** 1675–1775 (C=O) **S–N**	10–13 **W**	160–185 **S**
—**C**–**NH**—	3500–3200 **W–N**	1.0–5.0 **W–N**	35–50 **S**
	3100–3000 **W–N**	6.5–8.5 **S–N–SS**	100–170 **S**

* **NA** not applicable, **S** strong, **N** narrow, **B** broad, **W** weak, **SS** split.

importance (see discussion of petroleum hydrocarbon analysis above). Neither carbon tetrachloride nor carbon disulfide is used today because of toxicity, difficulty in handling, and detrimental environmental effects.

Methods are available to remove the interferences due to solvents, such as subtraction and compensation. These may be accomplished manually or electronically depending on the instrumentation and how they are configured. However, the preference is always to obtain a spectrum without the interference of a solvent!

Perhaps the most widely and commonly used method for liquid sampling, used with FT-IR spectrophotometers, is attenuated total reflection, the cell for which is shown in *B* in Figure 8.7. A liquid sample is placed in the trough, the bottom of which is a crystal situated such that the incident IR beam is totally reflected inside the crystal and exits into the instrument. When reflected, the beam appears to slightly pass into the sample in contact with the crystal, allowing the sample to absorb energy, producing a spectrum. Another common method is to place the pure liquid between two salt windows. A drop may be placed between two salt windows or in a sealed cell, where the space between the windows is predetermined and held constant. These types of cells can be used where quantitation of the component is desired or where a volatile solvent or solution of a compound is being analyzed.

Solid samples or solid extracts can be mixed and ground with potassium bromide (KBr), pressed to form a transparent pellet and a spectrum obtained

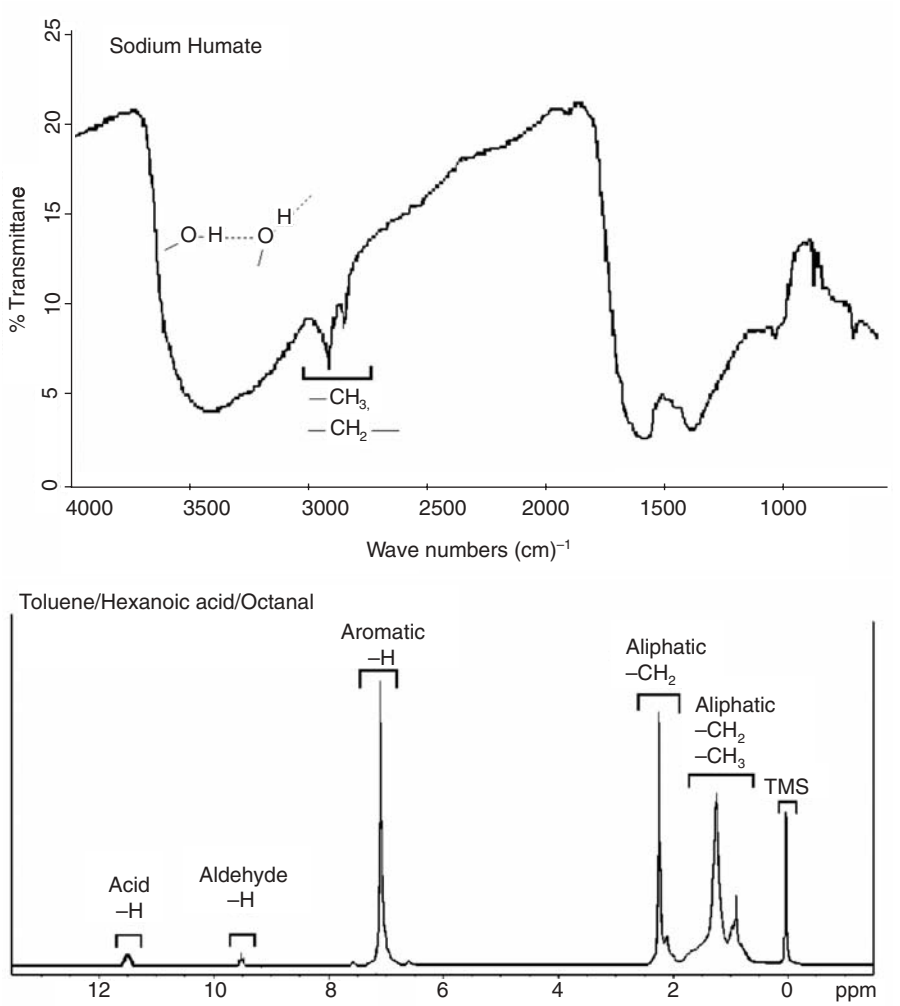

Figure 8.12. Infrared spectra of a KBr pellet of 3% sodium humate (Aldrich) and an NMR spectrum of a mixture of toluene, hexanoic acid, and octanal. The functionalities responsible for the absorptions are labeled.

from the pellet (see *C* in Figure 8.7). There are gas cells for obtaining spectra of gases, and many other methods for obtaining spectra from liquid and solid samples are available but not as frequently used as these [22–26].

8.12. NUCLEAR MAGNETIC RESONANCE

Although some useful and interesting data have been obtained by extracting various components from soil and analyzing them by nuclear magnetic reso-

nance (NMR) spectroscopy, it is not generally useful in the in situ analysis of soil or soil components. This is because NMR analysis depends on a stable uniform magnetic field in the sample. Most soils contain enough iron to alter the characteristics of the magnetic field, thus interfering with the analysis. Components extracted from soil can be analyzed by NMR; humus and phosphate are two examples.

Nuclear magnetic resonance is an extremely powerful method for observing the environment of an atom of interest. Most commonly the element studied is hydrogen-bonded to another element, usually carbon, and is referred to as NMR spectroscopy (sometimes called H NMR, ^{1}H NMR, P NMR or proton NMR). The second is carbon, specifically ^{13}C, attached to both other carbons and hydrogen. Other elements commonly measured include fluorine and phosphorus.

Details of NMR spectroscopy will not be dealt with here but can be found in sources cited in the Bibliography. NMR is most often carried out on liquid samples or solutions of pure compounds in deuterated solvents, although MNR of solids can also be obtained. Different organic functional groups, methyl, methylene, phenyl, the hydrogen adjacent to the carbonyl carbon in aldehydes, and organic acid group hydrogens absorb at different frequencies and thus can be easily identified. Similarly, different ^{13}C environments result in different absorptions; for instance, ^{13}C aromatic and carbonyl carbons have unique absorptions.

There are also vast number of powerful NMR experiments that yield detailed information about the structure of pure organic molecules. However, as with other regions of the spectrum, mixtures produce spectra that are mixtures of the components present in the ^{1}H NMR spectrum, such as the mixture of toluene, hexanoic acid, and octanal as shown in Figure 8.12. This spectrum shows the unique absorptions of each of these functional groups and also illustrates that mixtures of compounds give spectra containing the absorptions of all the components. Table 8.1 gives some important diagnostic adsorptions for ^{1}H and ^{13}C NMR spectroscopy.

One place where there is potentially more use for NMR in soil and environmental analysis is in speciation of soil components. Using a broadband NMR instrument, many different elements can be analyzed and used to differentiate between species, for instance, PO_4^{3-}, MPO_4^{2-} and $M_2PO_4^{-}$ (where M represents a metal). This potential, however, has not been exploited to any great extent [27–30].

8.12.1. Nuclear Magnetic Resonance Sample Preparation

Because most common solvents, including water, contain protons and most analysis involves the measurement of protons, a solvent without protons is generally used in NMR spectroscopy. Commonly solvents in which hydrogen has been replaced with deuterium (i.e., solvents that have been duterated) are used; the most common is deuterochloroform. In addition, an internal standard, most commonly tetramethylsilane (TMS), is added to the sample in the

NMR sample tube (see Figure 8.7, *D*) and all absorptions are recorded relative to the absorption due to TMS.

8.13. MASS SPECTROSCOPY

Mass spectroscopy as the name implies is a method by which the mass of a molecule, that is, its molecular weight, of a pure compound is determined. A sample is introduced into the instrument, ionized, and fragmented during the ionization process. It and its fragments are accelerated down a path that can be one of several types, separating ions and fragments on the basis of their mass and charge, and finally the ions are detected and recorded. Because the method depends on the ions moving unimpeded through the path, the inside of the instrument must be under high vacuum at all times.

The ionization–fragmentation process can be accomplished by bombarding the sample with electrons or with a chemical species. This process removes an electron from the compound and fragments it, producing a positive species. Accelerating plates at high negative voltage attract the particles that pass through a hole or slit in the plate and move down the path. The path may be a straight tube at the end of which is a detector. The time it takes an ion to travel the length of the straight tube will depend on its mass and charge. This is called a *time-of-flight* (TOF) mass spectrometer.

In a magnetic sector mass spectrometer the path of the ions is through a curved tube with a magnet at the curved portion. The path of the charged species will bend in the magnetic field depending on the strength of the magnetic field and the mass and charge of the ions. After passing the magnet, the charged species will impinge on a detector and be recorded. When the strength of the magnet is changed, the masses of the ions reaching the detector will change and be recorded.

In a quadrupole mass spectrometer the ions pass into a path between four rods attached to an electric circuit that can induce a range of frequencies in the rods. Ions will resonate in the quadrupole until a certain frequency, which depends on their mass and charge, is reached, and then the ions exit the quadrupole and are measured. A diagram of a quadrupole mass spectrometer is given in Chapter 9, Figure 9.5.

As with all the spectroscopic methods, this method is best suited to measurement and elucidation of the characteristics of pure compounds. For this reason, MS is often used as a detector for gas chromatographs. The MS of choice for this use is the quadrupole mass spectrometer. For this reason it will be discussed again in Chapter 9, Section 9.2.4.

Using mass spectrometry it is possible to determine the molecular weight of the compound being analyzed. It is also possible to distinguish between isotopes of elements. Thus ^{14}N and ^{15}N can be separated and quantified using mass spectrometry.

Much work has been done on the nitrogen cycle and fate of nitrogen compounds in the environment using ^{15}N mass spectrometry. Fertilizer or other

nitrogen containing material, enriched in ^{15}N, is applied to soil or a crop and samples taken after various periods of time. The samples are digested using kjeldahl methods as discussed earlier in Chapter 6, and ammonia is collected and then decomposed before being injected into the mass spectrometer. A species enriched in ^{15}N is a measure of the partitioning of nitrogen and its movement in the environment. Other stable isotopes can be used in a similar way to follow their movement through the environment in general and in soil in particular [9,10,31–33].

8.14. CONCLUSION

Matter interacts with all forms of electromagnetic radiation, and these interactions are used to gain information about the matter with which it interacts. Thus, X-ray, atomic absorption, and many other spectroscopic methods are available for the investigation of soil. X rays are used to determine the structure of clays and identify the elements present in soil via X-ray fluorescence. Atomic emission, adsorption, and ICP are routinely used to determine metals extracted from soil. Ultraviolet and infrared spectroscopies along with mass spectrometry are used to unequivocally identify compounds. NMR is extremely powerful in determining the structure and species of various compounds and species. A common method of analysis is colorimetry, in which the component of interest is reacted with a reagent, which produces a colored compound. Using the visible region of the spectrum, the intensity of the color, as measured by a spectrophotometer, is directly related to the amount of component present in the sample using a calibration curve. The results are related back to the original sample.

PROBLEMS

8.1. Describe two ways to distinguish between an analytical signal and noise in the output of an instrument.

8.2. Describe some major limitations of X-ray fluorescence as a method for determining metals in soil.

8.3. What is spectral overlap, and how may it affect analysis of metals extracted from soil?

8.4. List three common solvents useful in UV–Vis spectroscopy. Which of these might be more useful in analysis of soil extracts?

8.5. Describe two ways in which spectra may be used to identify compounds.

8.6. Explain how infrared and nuclear magnetic resonance spectra are different.

8.7. Soil commonly contains water. Why are water and infrared spectroscopy not compatible?

8.8. Explain why water is rarely used as a solvent in NMR spectroscopy.

8.9. Mass spectroscopy is useful because it can determine what important characteristic of a compound?

8.10. Explain, giving at least two reasons, why using UV–Vis or mass spectroscopy as a detector for chromatography might be beneficial.

8.11. A researcher randomly picks test tubes and uses them in the spectrophotometer and obtains confusing results. Explain how this can happen.

8.12. Explain what a calibration curve is and how it is used.

8.13. Two researchers prepare calibration curves; one researcher's curve has an r^2 of 0.9977 the other, an r^2 of 0.9845. Which is the better calibration curve? Explain how you came to this conclusion; be specific.

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CHAPTER

9

CHROMATOGRAPHY

Chromatography and its derivatives, such as solid-phase extraction, discussed in Chapter 7, is a powerful, essential tool for the analysis of soils. Of the many forms of chromatography that have been developed, gas and high-performance liquid chromatography are the most commonly used, particularly in the analysis of soil extracts. Chromatographic methods, as are spectroscopic methods, are almost exclusively referred to by their acronyms: CG—gas chromatography, LC—liquid chromatography, HPLC—high-performance liquid chromatography,[1] TLC—thin-layer chromatography, CE—capillary electrophoresis. Also, because there are subdivisions to each type of chromatography, the acronyms are often lengthened, such as GLC—gas–liquid chromatography (liquid because there is a liquid coating the stationary phase).

Chromatographic methods are also often used as so-called hyphenated methods, where their output is used as the input for an identification method such as mass spectroscopy. These "hyphenated" methods are also most often referred to by their acronym, for example, GC/MS—gas chromatography/mass spectroscopy and HPLC/MS—high-performance liquid chromatography/mass spectroscopy. Note that although UV–Vis is hyphenated, it is not a hyphenated method in that it does not consist of two different methods of analysis.

9.1. FUNDAMENTALS OF CHROMATOGRAPHY

All chromatographic methods function on the same principle, which is the partitioning of components of a mixture between two phases: (1) a stationary phase, which may be a solid, liquid, or gel, and (2) a mobile phase, which may be gas, liquid, solution, or a varying mixture of solvents. When a mixture is introduced to a chromatographic system, its components are alternately adsorbed and deadsorbed, that is, partitioned between, the stationary and mobile phases. Partitioning is caused by different polarities of the stationary and mobile phases and the compounds being separated. Compounds in the mixture have different affinities for the phases and will move at different rates in the chromatographic system and thus be separated.

[1] High-performance liquid chromatography is also called *high-precision* and *high-pressure liquid chromatography*.

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

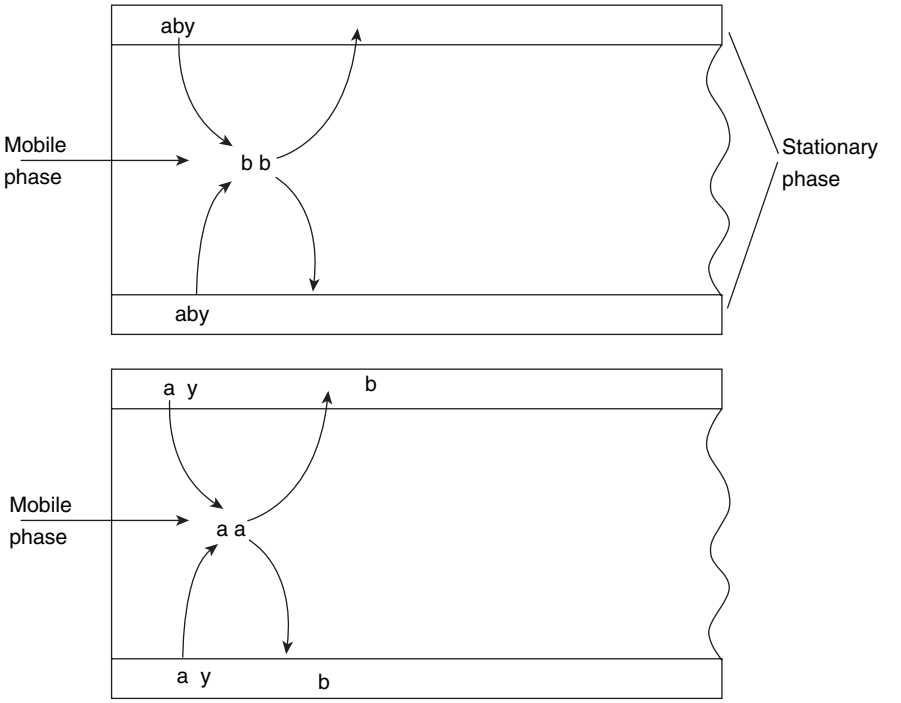

Figure 9.1. Partitioning of the components of a mixture in a chromatographic column.

This partitioning–separation process is diagramed in Figures 9.1 and 9.2. In Figure 9.1 a mixture is introduced into a chromatographic column. The components are adsorbed on the stationary phase, and the component more soluble in the mobile phase moves into this phase and is swept along the column, where it is adsorbed again. The less soluble component then moves into the mobile phase and is again readsorbed by the stationary phase. In Figure 9.2 a mixture is introduced into a chromatographic column, and as its components move down the column, they are separated into discrete "packets" of the same compound and on exiting the column, they are detected.

Table 9.1 summarizes the common chromatographic methods and the characteristics that differentiate them from other methods. In this list HPLC and GC are most commonly used in analyzing soil extracts, particularly when they are used in combination with mass spectral methods.

9.2. GAS CHROMATOGRAPHY

Gas chromatography is a powerful, rapid method for separation of mixtures of gases and compounds with boiling points below ~400°C.[2] The sample, when

[2] The temperature value 400°C is not an absolute upper limit of GC but is near the upper limit of most GC columns.

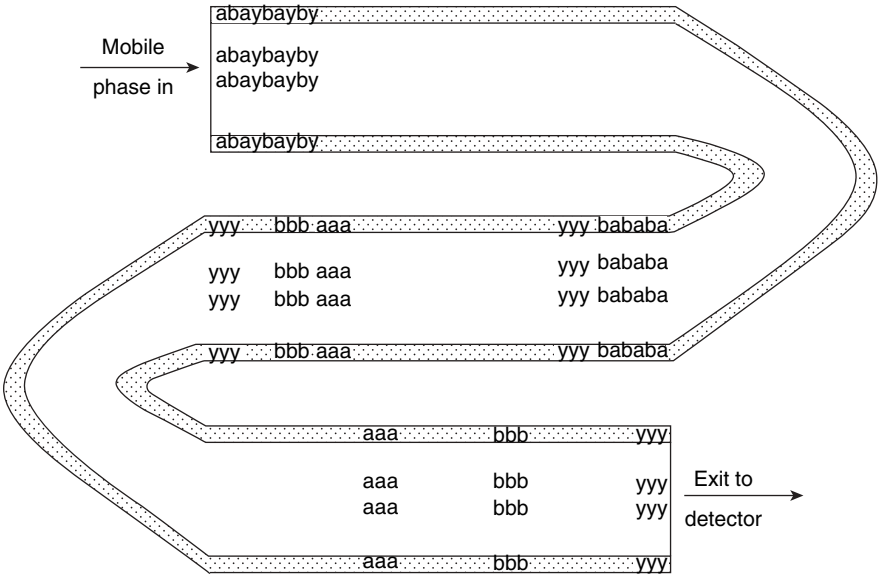

Figure 9.2. Separation of a complex mixture by chromatography. The top segment shows the column just after application of mixture. As the sample progresses through the column, separation of mixture occurs as a result of the chromatography. The first component exits the column and is detected, followed by the other two components.

introduced into a gas chromatograph, must either already be a gas or be immediately turned into a gas on injection. A complete chromatogram is usually obtained in less than 20 min, although some analysis can take much longer.

9.2.1. Sample Introduction

For gas chromatography a 1 to 0.1 μL syringe, shown in Figure 9.3, which has the entire sample contained in the needle, is frequently used for the introduction of the sample into the injection port. The needle must be of the correct type and length for the gas chromatograph being used, or poor results will be obtained. Injection is effected through a septum made of special rubber; Teflon-backed septa are also available and must be in good condition for accurate chromatograms to be obtained. Injection ports without septa are also available. When a new septum has been placed in the injection port, piercing it with a syringe needle will be harder, and care must be taken not to bend the needle or syringe plunger. When the needle is fully inserted, the plunger is depressed and a recording of the detector output is started. This is the 0 time used in calculating the retention times (R_t, described below) of emerging peaks. Needle insertion and depression of the plunger must be done in one continuous motion to obtain a good chromatogram. GCs can be fitted with automatic samplers that take and inject the sample automatically.

Table 9.1. Fundamental Types of Chromatography

Type of Chromatography	Stationary Phase	Mobile Phase	Types of Compounds Amenable to Separation
Gas (also GLC[a] and GSC[b])	High-boiling liquid	Gas	Compounds to be separated must be in gaseous phase
HPLC[c]	Solid	Solvent, aqueous, aqueous solution; solvant, or mixture of solvants	Compounds or ions must be soluble in mobile phase and have appreciable attraction to solid phase
TLC[d]	Solid	Solvent, aqueous, aqueous solution; solvant, or mixture of solvants	Compounds or ions must be soluble in mobile phase and have appreciable attraction to solid phase
Electrophoresis	Solid or semisolid in a buffer solution	Curerent	Compounds or ions must be ionized to be separated

[a] Gas–liquid chromatography.
[b] Gas–solid chromatography.
[c] High-performance liquid chromatography.
[d] Thin-layer chromatography.

9.2.2. Mobile Phases

In gas chromatography common mobile phases are hydrogen, argon, helium, and nitrogen gases and air; helium and nitrogen are the most commonly used. Because gas chromatographic detectors are extremely sensitive and it is desirable to keep the noise level as low as possible, it is always advisable to use very high-purity gas as the mobile phase.

9.2.3. Stationary Phases

Chromatographic stationary phases are either solids or liquids coating or bonded to solids. Common stationary phases are listed in Table 9.2. For gas chromatography the solids are activated carbon of various sorts, polymers, molecular sieves, or similar materials, and the chromatography may be referred to as *gas–solid chromatography* (GSC). This type of stationary phase

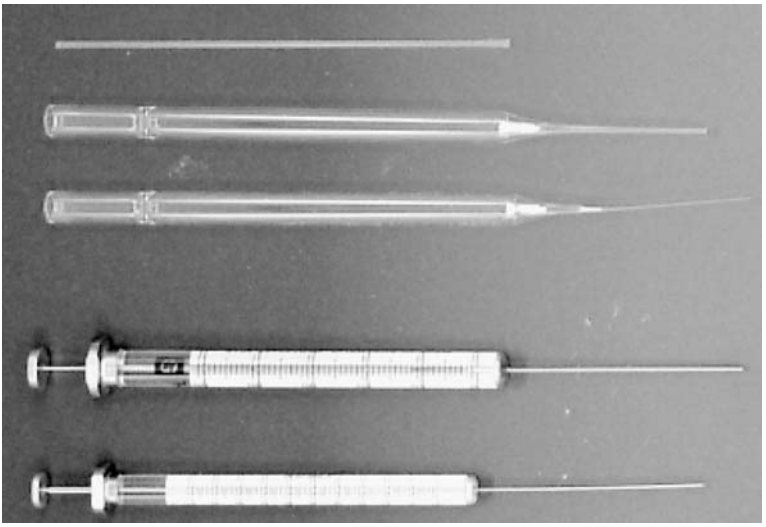

Figure 9.3. Sample application tools from top to bottom, glass capillary, Pasteur pipette with tip drawn out, syringe for HPLC, and syringe for GC.

Table 9.2. Common Stationary and Mobile Phases Used in Chromatography

Chromatographic Method	Stationary Phases	Mobile Phases
Gas chromatography	Solid or inert solid covered by high-boiling liquid[a]	Gas, usually helium, nitrogen, argon, or hydrogen
High-performance liquid chromatography	Fine porous silica and alumina; particles are finer, increasing efficiency but requiring high pressure to push elutant and sample through column[b]	Same as liquid chromatography but at high pressure
Thin-layer chromatography (applies also to paper chromatography)	Thin layer, commonly 250 μm thick, of silica gel, alumina, cellulose,[b] or a sheet of chromatographic paper	Aqueous and solvent solutions
Electrophoresis	Solid or semisolid absorbant saturated with buffer	Current

[a] Oil can be varied such that the stationary phase has varying degrees of polarity.
[b] Surface modifications commonly involve phases to change the surface polarity such as attaching a long-chain hydrocarbon, usually 18 carbons in length. This would be called a *reverse-phase C18-impregnated stationary phase*.

and chromatography is commonly used for compounds that are gases at room temperature or are low-boiling compounds.

There are two types of gas chromatography columns that employ a liquid phase. Packed columns, usually 3 or 6 mm (~$\frac{1}{8}$ or $\frac{1}{4}$ inch) in diameter and 2 m (~6 ft) long, are filled with an inactive solid coated with a high-boiling grease or oil or has a similar compound bonded to it. Also, compounds having various functionalities and polarities can be bonded to the stationary phase to produce packings with specific separation capabilities. The second type is a capillary column with an inner diameter of 1 mm or less and a length of either 30 or 60 m, although some are much longer. The inside of the capillary is commonly coated with the same stationary-phase materials as used in packed columns. It is this layer of organic material, which acts as the adsorbant. In either case the layer must have a sufficiently high boiling point or be attached such that it will not be lost when the column is heated during use.

Most columns are usable to 250–300°C and in some cases even higher. A capillary gas chromatographic column (*A*) and a packed column (*B*) are shown in Figure 9.4.

Some GC packings or stationary phases are sensitive to either or both oxygen and water. Because soil always contains both, it is important that care be taken with soil extracts to make certain that they do not contain any components that will degrade the chromatographic column, which may cost between $500 and $1000.

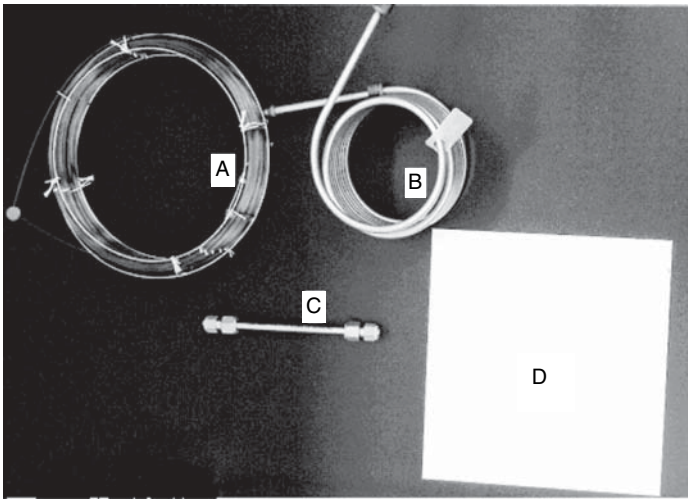

Figure 9.4. Columns used in chromatography and a thin-layer sheet. Columns *A* and *B* are a capillary column and a packed column, respectively. Column *C* is for HPLC. Column *D* is a thin-layer sheet with plastic backing such that it can be cut into smaller pieces as needed.

Table 9.3. Common Detection Methods for Chromatography

Gas	Thermal conductivity (TC or TCD)
	Flame ionization (FID)
	Mass spectrometry (MS or GC/MS)
High-precision-liquid	Ultraviolet or visible (UV–Vis)
	Refractive index
	Conductivity
Thin-layer	Fluorescence-indicator-impregnated
	Visualization reagents
	Charring with acid and heat
Electrophoresis	Staining
	Ultraviolet or visible (UV–Vis)
	Conductivity

9.2.4. Detection

There are three main types of detectors used in gas chromatography: thermal conductivity (TC), also known as a hot-wire detector, flame ionization (FID), and quadrupole mass spectrometer (MS) (Table 9.3). The TC detector consist of coils of high-resistance wire in a detector block where the carrier gas from the gas chromatograph exits the column and flows over the wire. The coils of wire are arranged in a Wheatstone bridge with two arms receiving gas exiting the column and other two receiving the pure carrier gas. The heat capacity of the carrier gas changes when a compound in the carrier gas is exiting the column; this changes the resistance characteristics of the coil, which are recorded. TC is a universal detector but it is also the least sensitive of the common detectors.

The flame ionization detector has a small hydrogen flame into which the carrier gas exits. There is a voltage across the flame, which is nonconducting. When an organic compound exits the column, it is burned and produces ions, which conduct electricity. The electrical signal thus produced is recorded. FID is a highly sensitive detector although not the most sensitive. It will respond only to organic compounds, or compounds that will burn, and exiting compounds are thus destroyed in the process.

The quadrupole mass spectrometer detector or mass filter, as it is sometimes called, is extremely sensitive and allows the identification of compounds exiting the gas chromatograph. The compounds are destroyed as they exit the chromatograph and enter the mass spectrometer (shown in Figure 9.5), where they are ionized, fragmented, and analyzed. Other sensitive but also more selective detectors are available and may be called for in certain analyses.

9.2.5. Gas Chromatography Applied to Soil Analysis

Soil extracts are most commonly characterized by gas chromatography and gas chromatography–mass spectrometry. For example, the composition of soil

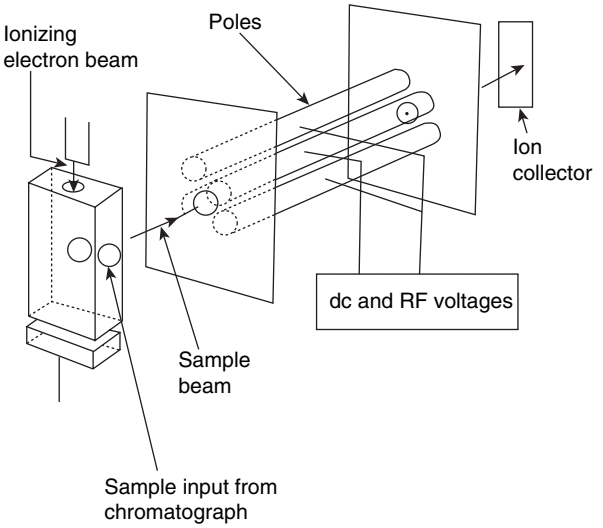

Figure 9.5. Diagram of quadrupole MS used as detector for gas chromatography.

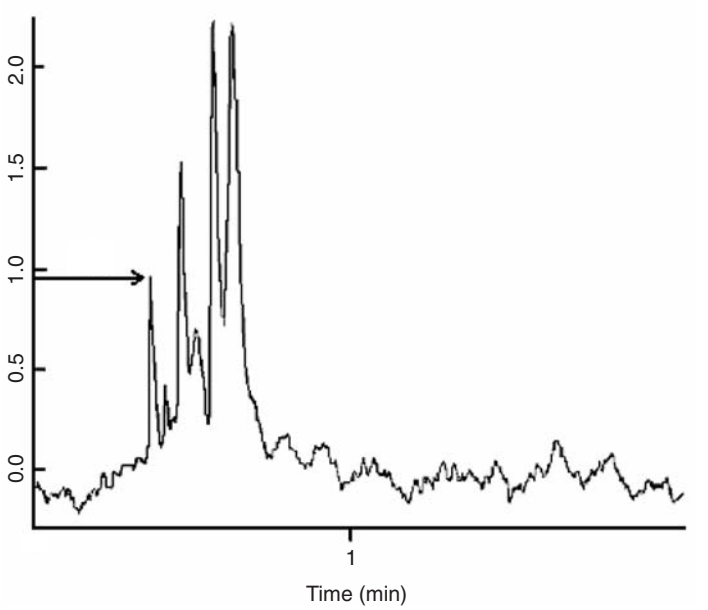

Figure 9.6. Gas chromatograph of soil air taken in Figure 9.7. R_t is the time it takes the first peak to come out. R_t values can be found for all peaks if needed.

air can easily be analyzed using gas chromatography. A gas chromatograph of the soil air in a container with wheat growing in it is shown in Figure 9.6. A sample of the soil air for this analysis was obtained by simply inserting a small Phillips head screwdriver into the soil to make an access hole for a syringe needle. The syringe needle is then inserted into the hole and the plunger with-

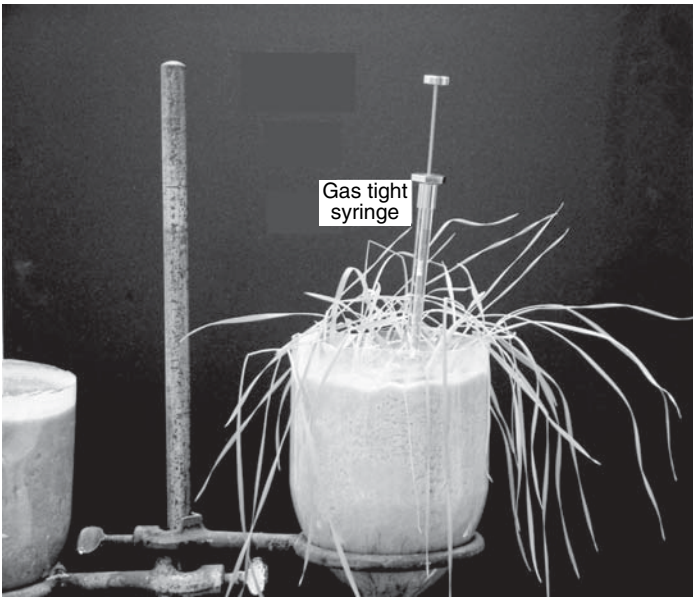

Figure 9.7. Gastight syring used to sample soil air; the same chromatograph is shown in Figure 9.6.

drawn to move soil air into the syringe (see Figure 9.7). This syringe has a valve in the needle hub such that the sample is sealed in the syringe between taking the sample and injecting it into the chromatograph. The GC used for this analysis is equipped with a flame ionization detector such that each peak must represent an organic compound. If standards with the same retention times are injected into the gas chromatograph under these same exact conditions, then the "identity" of the compound would be assumed. A better approach, however, would be to use a GC/MS that would give a true identification of these components.

It is most common to extract a soil, using any of the many extraction procedures described in Chapter 7, with an organic extractant, dry, clean, concentrate, if necessary, and inject the extract into the gas chromatograph or GC/MS. Although such extraction procedures can and are used to study natural organic components in soil, the greatest use of GC and GC/MS in soil analysis is in two cases: (1) to assess the fate of a herbicide or insecticide once they have been applied to a field or in the case of a spill of these materials and (2) to determine the condition of soil contaminated by an industrial organic material such as from a spill or as a byproduct of a manufacturing process. There are no plant nutrients other than carbon dioxide and water, which are volatile, and so GC is not used to investigate soil fertility.

A number of techniques have been developed to directly volatilize soil components and contaminants and introduce them directly into the carrier gas of a gas chromatograph. Such procedures avoid the time and cost of extrac-

tion, cleanup, and concentration and also the introduction of contaminants during extraction, drying, cleanup, and other procedures. However, these methods are not universally applicable to all soil analysis procedures, and caution must be used in application to new or untested analysis or analytical procedures [1,2].

9.2.6. Gas Chromatography–Mass Spectroscopy

Gas chromatograph–mass spectroscopy (GC/MS) consists of a gas chromatograph where the chromatographic column exits into a mass spectrometer (see Figure 9.5) that acts as a detector for the GC. As the GC carrier gas exits the GC column, the carrier gas is stripped from separated components that enter the MS, where it is ionized, fragmented, and scanned and the mass of the component it contains along with its fragmentation pattern is recorded. The fragmentation pattern is used to identify the compound using a computer to match the pattern found with standard fractionation patterns for known compounds. The computer then produces a list of compounds that come close to matching the unknown along with a percentage (%) that indicates how well the patterns match.

Caution must be exercised with these types of matching pattern computer programs as they can easily misidentify compounds, especially if the program is set up for the wrong analysis or assumed composition of the unknown [2–4].

9.3. HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY

In high-performance liquid chromatography (HPLC) the mobile phase is a liquid, in which the sample must be soluble, and detection is most often accomplished by ultraviolet absorption. It is generally a slower process than gas chromatography; however, its advantage is that the compounds to be separated are not limited by their boiling points, although low-boiling compounds are almost never separated by HPLC. Solid mixtures, as long as they are soluble in the mobile phase, can be chromatographed.

9.3.1. Sample Introduction

For HPLC the injector is a valve. In the charge position a 50-μL syringe is used to fill the sample loop that holds a specific volume of sample solution. The valve is switched to the RUN position, and the elutant carries the sample out of the sample loop and into the column. A recording of the detector output is automatically begun and produces a chromatogram of the separated components.

9.3.2. Mobile Phases

In HPLC the mobile phase is a liquid or a mixture of liquids. The common elutants are water, aqueous solutions, acetonitrile, and methanol. Almost any other common solvent, compatible with the column packing and the detector, may be used. In some cases the HPLC instrument will be capable of making a mixture of elutants or changing the mixture of elutants during chromatography. If this is done, care must be taken to make sure that the elutant mixture is compatible with the detector.

9.3.3. Stationary Phases

In HPLC columns (Figure 9.4, *C*) the most common packing is a solid with an organic group attached to it. For instance, the solid may have a hydrocarbon containing 18 carbons attached to it, making it hydrophobic. This type of column would be called a C18 column or reverse-phase column. Columns can be made with varying polarity and functionality and thus be used to carry out a wide variety of separations.

9.3.4. Detection

In HPLC four different types of detectors are common: ultraviolet, refractive index, conductivity, and mass spectrometry. The ultraviolet detector is an ultraviolet source that passes a specific wavelength of UV light through the sample as it exits the chromatographic column. The absorbence of the compound is then recoded. The source of ultraviolet light may be a deuterium lamp with a filter to remove all except the desired wavelength of light. Alternatively, it may be designed like a spectrometer such that the analytical wavelength of light being used can be changed. In the most sophisticated cases the whole spectrum of the compound may be taken as it exits the column. In this case the ultraviolet spectrum may be used to identify the compound.

Refractive index and conductivity detectors are much simpler but cannot identify compounds eluting from the chromatographic column. The refractive index of the elutant will change as its composition changes. Thus, as compounds elute from the column, the refractive index (RI) changes, and this is recorded to obtain a chromatogram. The conductivity detector is used when water is used as the elutant and the materials being separated are ionic. When no ions are present, the conductivity of water is very low; when ions emerge from the column, the conductivity of the water increases and is recorded, producing the chromatogram. Chromatographic procedures that involve changing the elutant during the chromatography will not generally be suitable for these detectors.

The quadrupole mass spectrum detector is used in the same way as it is with gas chromatography with the same sensitivity and ability to identify compounds. In use most or all the solvent is removed before the sample enters the MS.

Retention times can be used for "identification" of eluting compounds in the same way as with CG [4–6].

9.3.5. High-Performance Liquid Chromatography Applied to Soil

In soil analysis HPLC is used much like GC in that soil is extracted and the extract, after suitable cleanup and concentration, is analyzed. One major difference between them is that HPLC does not require that components be in the gaseous phase. They must, however, be soluble in an elutant that is compatible with the column and detector being used. Another difference is that both a syringe and an injector are used to move the sample into the elutant and onto the column. Detection is commonly by UV absorption, although both refractive index and conductivity are also commonly used. Conductivity or other electrical detection methods are used when analysis of ionic species in soil is carried out [2,6,7].

9.4. THIN-LAYER CHROMATOGRAPHY

Thin-layer chromatography (TLC) is carried out on a thin layer of adsorbant on a glass or plastic support (other supports have been used). It has sometimes been referred to as *planar* chromatography since the separation occurs in a plane. Paper chromatography, which is carried out using a piece of paper, usually filter paper, is very similar to thin-layer chromatography and will not be covered here.

9.4.1. Sample Application

In thin-layer chromatography a 10-μL or larger syringe is often used to place sample spots on the thin layer before development. Alternatively, a glass capillary tube or Pasteur pipette may be heated in a burner and pulled to obtain a fine capillary suitable for spotting (see Figure 9.3).

9.4.2. Mobile Phases

Development of the thin-layer chromatogram is accomplished by placing a small amount of elutant in the bottom of a suitable container then placing the spotted thin layer in the container, sealing it, and allowing the elutant to ascend the layer through capillary action.

9.4.3. Stationary Phases

A thin layer of adsorbant is applied to a support that may be a sheet of glass, metal, or plastic (Figure 9.4, *D*). Adsorbants are typically alumina, silica gel, or cellulose and may be mixed with gypsum to aid in adhering to the support.

They may also include a fluorescent indicator that aids in visualization once the plate is developed. These adsorbants may also have hydrocarbons attached to them such that reverse-phase TLC can be carried out.

9.4.4. Detection

> ***Caution*:** Spraying thin layer plates with visualization reagents should always be done in a functioning hood. These reagents may be caustic and toxic, as is the case with sulfuric acid.

Thin-layer plates, once developed and dried, are sprayed with a visualization reagent that allows the detection of the separated components. The visualization reagent produces a colored spot where each component of the mixture is on the plate. For example, a 0.1% solution of ninhydrin (1,2,3-indantrione monohydrate) in acetone can be sprayed on a plate that has separated amino acids on it. The amino acids will show up as blue to brown spots on the plate. Sulfuric acid can be sprayed on a silica or alumina plate and heated to show the position of organic compounds as charred spots (this reagent cannot be used on cellulose plates since the whole plate will turn black). Reducing compounds can be visualized by using a solution of ammonical silver nitrate, which will produce black spots where the silver is reduced by the reducing compound. This reagent is particularly sensitive for reducing sugars. There are a whole host of different visualizing reagent that can be found in the book by Hellmut et al. [8].

A method for detecting compounds that have UV absorbence is the use of thin-layer plates that contain a fluorescence dye or indicator. Plates are developed and then placed under an ultraviolet light. Compounds on the plate may show up as bright or dark spots depending on their interaction with the ultraviolet light and the fluorescence produced by the fluorescence indicator [8,9].

9.4.5. Soil Thin Layer

A variation on thin-layer chromatography is the use of soil thin layers to investigate the movement and degradation of organic compounds in soil. A soil is sieved using a number 200 or smaller sieve, and this soil is used to produce a suspension of soil in distilled water that is then spread on a glass sheet to produce a soil thin layer. An organic component, often a herbicide, is spotted on the dried sheet, which is developed using deionized water. The movement and degradation of the organic compound under these conditions can then be related to its expected movement and degradation in soil in the field [10].

9.5. ELECTROPHORESIS

As with other chromatographic methods, there are an number of electrophoretic methods, including paper, gel, and capillary. Electrophoresis uses an electric current to move ionic species, either simple ions, amino acids, or complex proteins, through a medium or a capillary [i.e., capillary electrophoresis (CE)]. During this process, the ionic species typically move at different rates and are thus separated (CE is somewhat different, as discussed below).

9.5.1. Sample Application

In paper or gel electrophoresis, a sample may be applied with a syringe or a micropipette similar to the application of samples to thin layer plates. In some cases there may be "wells" in the gel that accept the solution containing the species to be separated. In capillary electrophoresis samples may be applied using electromigration, hydrostatic, or pneumatic injection.

In all cases the ions to be separated must be soluble in and compatible with the stationary phases and buffers used.

9.5.2. Movement of Species

For electrophoresis the paper or gel is saturated with the required buffer at the desired pH. The ends of the paper or gel are placed in a buffer reservoir that contains the buffer with which the paper or gel is saturated and that also have electrodes connecting one end to the positive DC terminal and the other to the negative terminal of the power source. It is the electrical current that causes the movement of ionic species through the medium.

In capillary electrophoresis a high voltage is used to produce electroosmotic flow, and both electricity and the buffer flow through the capillary, with the buffer flowing toward the cathode. Both carry the sample through the capillary, and because of the flow of the buffer, both charged and uncharged species are separated.

9.5.3. Stationary Phases

Electrophoresis can be carried out using paper or a gel as the supporting medium. Typically it can be carried out only in media compatible with water since buffers or salt solutions are required to carry the electric current required for separation. Capillary electrophoresis is carried out in a fused-silica capillary filled with buffer.

9.5.4. Detection

Once an electrophoretic separation has been accomplished, the paper or gel is sprayed or dipped in a visualizing solution similar to that used in the visu-

alization of components on a thin-layer plate. Detection methods similar to those used in HPLC are used in capillary electrophoresis. The type of separation, ion, organic ion, or ionic biomolecule will determine which detection method is best.

9.5.5. Electrophoresis Applied to Soil

Gel electrophoresis has been applied to soil DNA and RNA extracts using procedures similar to those used in DNA testing used for forensic analysis. Capillary electrophoresis has also been applied to the analysis of ionic species extracted form soil. While these processes show promise for the elucidation of valuable information about soil, neither is used for common, routine soil analysis [11–13].

9.6. IDENTIFICATION OF COMPOUNDS SEPARATED BY CHROMATOGRAPHIC PROCEDURES

There are three ways of "identifying" compounds once they are separated. The simplest is that for which chromatography is named. If a mixture of colored compounds is separated, and then where and when they elute or are found on a thin layer or gel is identified by searching for the color. After separating and visualizing a colorless component, it must be identified. This can be done in one of two ways. The first and simplest is by R_f or R_t, which are the distance, relative to some fixed point, that compounds move during a chromatographic procedure or between the time they enter and exit the chromatographic column [see equation (9.1) and Table 9.4 for further details on the use of R_f and R_t]. The formulas for R_f and R_t are:

$$R_f = \frac{\text{distance spot moved}}{\text{distance elutant moved}} \tag{9.1}$$

$$R_t = \text{Time from injection to top of peak}$$

Table 9.4. The R_f Values of Spots on Thin-Layer Sheets Shown in Figure 9.6[a]

Spot	Calculation	R_f
A_1	3/15	0.20
A_2	9/15	0.60
B_1	2/15	0.13
B_2	9/15	0.60
B_3	12/15	0.80

[a] The distance that the solvent front moved is 15 cm; therefore, this is the denominator for all calculations.

R_f is applied to thin-layer chromatography, while R_t is applied to GC and HPLC. For identification purposes the R_f values of pure samples of all the compounds to be found in the unknown are determined and are used to "identify" these same compounds in an unknown mixture. The R_f of a compound is determined by placing a spot of sample on a thin layer sheet slightly above the bottom, placing the TLC sheet in the developing chamber with the appropriate elutant and allowing the elutant to raise the thin layer until it is close to the top (but never at or over the top). The distance from the site where the spots were placed on the plate to top of the plate where the elutant stopped is recorded. This number is then divided into the distance the spot moved from the beginning as shown in Figure 9.8.

The distances should always be measured from the starting point, which is where the original mixtures were spotted both from and to the midpoint of the spots. The determination of the R_f values of five spots on a thin-layer chromatogram is given in Figure 9.6. The R_f values of the spots are listed in Table 9.4. The material in spots A_2 and B_2 are expected to be the same since they have the same R_f and the chromatogram was run under exactly the same conditions.

R_t is used in the same manner as is R_f except that it is the retention time of components in the GC or HPLC column (see Figure 9.6). The time can be measured from the time of injection to the time when the top of the peak elutes from the column. Another approach is to include a compound that is not retained by the solid or liquid phase in the injection and its peak used are the starting time for measuring the time for each peak to exit the column. Two compounds exiting the column at the same R_t will be assumed to be the same compound. Often R_t values are used for identification, although only a spectroscopic method can truly be an identification method.

Another method is to chromatograph the unknown and determine the R_f values of the components present. When a compound is "identified" by its R_f,

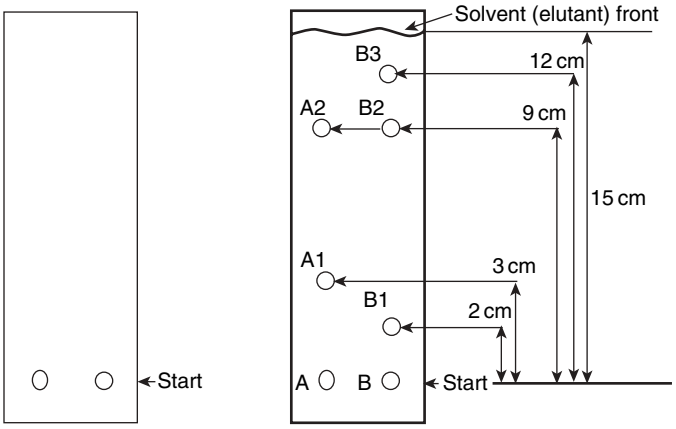

Figure 9.8. Thin layer at start (left) and after development (right). Right chromatogram shows the distance that the elutant and spots moved during the development of the chromatogram.

then a pure sample of that compound can be added (this is called "spiking") to the unknown, and a sample of this mixture analyzed. If only one larger peak is obtained at the specific R_f values of both the unknown and the known, they are the same (R_t can be handled similarly). When mixtures of similar composition are to be analyzed repetitively by a chromatographic method, it is common to prepare a list of the compounds that are expected to be present along with either their R_f or R_t values. In this way these components can be "identified."

9.7. QUANTIFICATION

GC and HPLC allow for ready quantification of the components exiting the column in that the area under the peak in the chromatograph is proportional to the amount of component present. However, to make a quantitative analysis, it is essential to have a calibration curve for each component of interest. This means making solutions of differing concentration, injecting them, and finding the relationship between peak area and amount of material present in a manner similar to that described in Chapter 8, Sections 8.8 and 8.9, for colorimetric analysis. In many cases the software that control the chromatograph can be set up to automatically do this analysis.

For TLC quantitative analysis is more difficult, although it can be accomplished in some instances. If such an analysis is undertaken, great care must be taken to ensure that there is an excellent relationship between the spot characteristic measured and the amount of material present.

9.8. CONCLUSION

Chromatography in its various forms is extremely important in the isolation and identification of complex mixtures found in the environment and in soil. It is applied to components that are isolated from soil by extraction. The extraction medium is important in that it must be compatible with the chromatographic method and analyte detection method. Samples for gas chromatography must be volatile in that they must be in the gaseous state for analysis. Samples for HPLC and TLC must be soluble to some minimal extent in the elutant being used. Detection of components after separation is performed by either optical, spectrophotometric, or electrical methods. In the case of TLC plates, separated components may be found by spraying the plates with a reagent that reacts with the component(s) present to produce a spot where the component has eluted. By chromatographing known compounds, a list of R_f values can be prepared and used to "identify" the components in an unknown mixture. In the case of GC and HPLC, the same thing can be accomplished by preparing a list of R_t values for compounds and used to "identify" components of an unknown mixture. Identification by R_f and R_t values is not

sufficient. True identification occurs when HPLC and GC are coupled to a spectrophotometic method such as MS and/or UV–Vis.

PROBLEMS

9.1. Explain how all chromatographic methods are similar.

9.2. Explain how the basic chromatographic methods are different.

9.3. What physical characteristics must components have to be separated by gas chromatography?

9.4. What do the terms R_f and R_t refer to? Explain how they are used in determining what components are likely to be present in a mixture.

9.5. Describe detection methods used in GC, HPLC, and TLC.

9.6. What general characteristic must a component have to be separated by electrophoresis?

9.7. How are soil thin-layer plates used in environmental investigations?

9.8. Explain why it is advantageous to have separated compounds exiting a GC or HPLC column analyzed by MS.

9.9. What is the difference between the mobile phase and the stationary phase in chromatography?

9.10. The chromatograms from two different injections have the following components (peaks) with the indicated retention times. Injection 1: peak 1, $R_t = 0.55$; peak 2, $R_t = 1.25$; peak 3, $R_t = 2.44$; peak 4, $R_t = 5.65$. Injection 2: peak 1, $R_t = 0.22$; peak 2, $R_t = 1.00$; peak 3, $R_t = 5.65$; peak 4, $R_t = 6.74$. Which of the peaks in the two chromatograms is likely to be the same compound?

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CHAPTER

10

SPECIATION

The term *speciation* for many people brings to mind oxidation forms of metals and nonmetals; however, this is much too restrictive an idea of speciation, especially when applied to soil. *Speciation* can broadly refer to the form of an element or molecule present under a set of environmental conditions as illustrated in Figure 10.1. This would include metal cation and oxyanion oxidation states, cation and anion associations, ionization states of organic compounds, and organic and inorganic associations. It will also include association of metals, nonmetals, organic ions, and compounds with both inorganic and organic components of soil, especially colloids.

Several different types of species commonly discussed in soil science are illustrated in Figure 10.1. The potassium cation (K^+) at the top is separated from the soil surface by a water molecule and would thus be considered an outer-sphere species. At the bottom the potassium cation is directly connected to the soil particle by an ionic charge and thus would be an inner-sphere species. Above this is an inner-sphere phosphate directly bonded to a soil surface aluminum. Also shown are potassium cations attached (inner sphere) to colloidal clay (CC) and colloidal soil organic matter (COM). Each of these is a different species.

Colloidal associations are particularly important in soil because they allow for movement of otherwise immobile or slowly mobile species. They can also lead to confusing analytical results in that atoms, ions, or molecules may appear to be soluble above their solubility limit when in reality they actually represent a different species.

Organic compounds might also be regarded as having species, although they are seldom discussed in this manner. Organic compounds are capable of existing in various conformations, some of which are easily changed, while others are more or less fixed by steric, hydrogen bonding, or other atomic and molecular interactions. In addition, compounds containing double bonds and chiral centers can exist as different optical isomers. For instance, fatty acids contain double bonds, which can be either *cis* or *trans*. Amino acids can be either *S* or *R* (D or L) optical isomers. These various "species" are illustrated in Figure 10.2. It is well documented that the various conformations and optical isomers have dramatically differing biological activities. An especially

Introduction to Soil Chemistry: Analysis and Instrumentation, By Alfred R. Conklin, Jr.

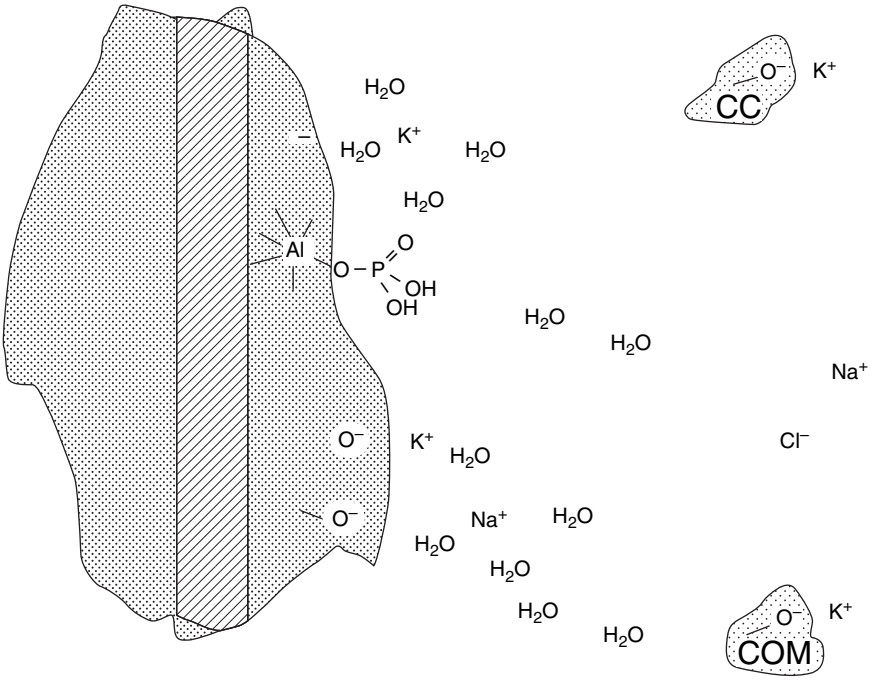

Figure 10.1. Species around soil particles (CC = clay colloid; COM = colloidal organic matter).

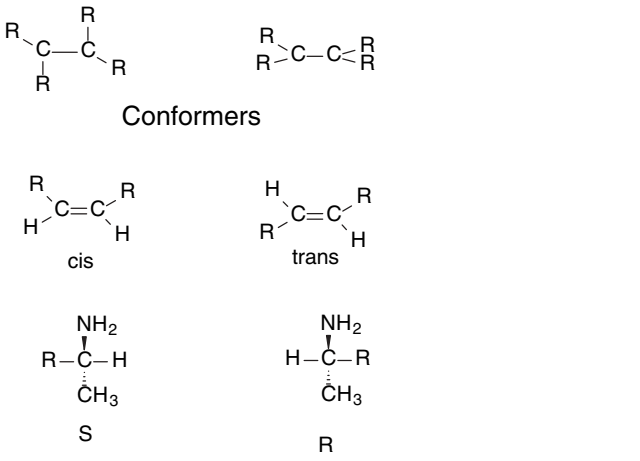

Figure 10.2. Common forms of organic compounds that have the same number and type of elements and bonds arranged in different orientations.

vivid example of this is thalidomide, which exists in two optical isomers, one relieving nausea the other causing birth defects.

Inorganic and metallorganic compounds also have conformations and isomers, including optical isomers. It is expected that these various conformers and isomers will also have varying biological activities.

There are many other reasons why speciation might be important. Among these are plant nutrient availability, biological contamination by metals, human or animal toxicity, movement of species in the environment, biological accumulation or amplification, obtaining data needed for models, understanding a species' role in the reactions and fates of various environment components, and development of a basic understanding of a species' chemistry. Any one of these by itself is an important reason to understand speciation, and often several of these reasons will be important at the same time.

Why might one be specifically interested in the species of components in soil? From a positive perspective it is desirable to provide plants with nutrients in forms that are available and yet are not present in concentrations high enough to cause environmental harm. From a negative aspect, some species of certain components are more and some less toxic. Some species are biologically available; some are not. Some species may accumulate in biological tissue; others will not. When a species is detected in the environment, it is important to know which of the categories described above it falls into.

A first step in deciding on an analytical procedure or a species to look for is to understand that the species of interest may be in one of, four "compartments" in soil (see Figure 10.3): the solid (both inorganic and organic), the liquid (soil solution), the gaseous (soil air), or the biological (living cells) compartment. It is important to remember that species are constantly moving both between compartments and between species.

Once the compartment is decided on, then the species, as illustrated in Figure 10.1, to be analyzed for can be decided on. In this case it is important

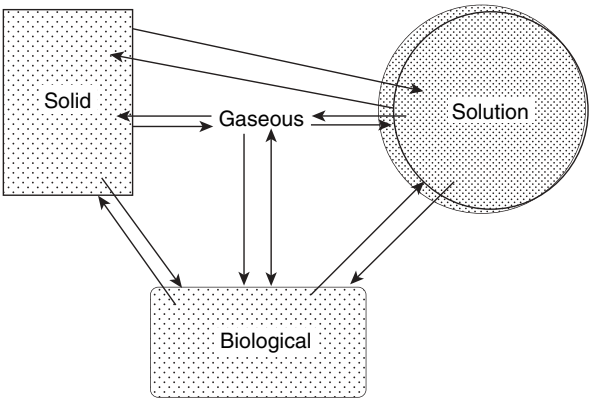

Figure 10.3. Soil compartments.

to consider all possible species that might be present in the compartment. It is also important to consider the changes that a species is likely to undergo during the sampling and analytical procedures to which it will be subjected. This topic is discussed further in Section 10.5 [1,2].

10.1. CATIONS

Any positive chemical species can be considered a cation or as being in a cationic form. Some will be present as relatively simple, single-oxidation-state cations, while others may be more complex in that they may have several oxidation states and the cations may have oxygens or hydroxy groups associated with them.

In all cases cations will associate with water molecules. Thus it is common to find them expressed as hydrated or hydroxide species. Such a representation more closely resembles the actual condition of cations in soil, particularly if the discussion relates to cations in the soil solution. This brings up a troublesome question or problem. When a cation is removed (extracted) from a solid matrix into an extracting solution, a species change most likely occurs. In the solid matrix the cation may not be associated with water molecules, while in solution it most certainly will be. For hydrated species the amount of associated water and the activity will also change. This raises the issue of the actual species present in the solid matrix. When analyzing for a specific cation species or discussing cation speciation in soil, it is important to keep this issue and problem in mind.

Molybdenum should be discussed in this section because it is a metal; however, it is always present in soil as an oxyanion and so will be discussed in the next section [3,4].

10.1.1. Simple Cations in Soil

Simple cations are those that exist in only one oxidation state in soil and are associated with water, although they may also be chelated and form other associations with inorganic and organic components.

The alkali and alkaline-earth metals are examples of relatively simple cations that occur in only one oxidation state and are surrounded by waters of hydration in the soil solution. The most common in soil, in order of decreasing abundance, are calcium (Ca^{2+}), magnesium (Mg^{2+}), potassium (K^{+}), and sodium (Na^{+}). Sodium is typically present in small amounts in high-rainfall areas, whereas it may be relatively high in low-rainfall areas.

Although simple, these cations can occur as a number of different species. They will be present as exchangeable or in solution as hydrated cations. They can be part of the inorganic component structure, for example, as in isomorphous substitution, or be trapped between clay layers. They will also be associated with organic matter and with colloidal inorganic and organic matter,

and chelated species. In many cases these cation species are further grouped into those available to plants and those that are unavailable. If a more detailed grouping is needed, species can be further divided into readily, slowly, very slowly, and nonavailable species.

Another simple cation commonly occurring in soil is ammonium (NH_4^+). Because of the unique role and chemistry of nitrogen and nitrogen species in soil, it will be discussed separately in Section 10.3 [5,6].

10.1.2. Complex Cations in Soil

Complex cations are those that can occur in several oxidation states in soil and are often associated with oxygen and hydroxy groups while still carrying a positive charge. The periodic table contains more than 65 metals that may be present as complex cations. Of these, only a few are common in soil. Even those that are not common in soil will, if present, occur predominately in one oxidation state under normal soil conditions. No attempt will be made to cover all these cations; rather, a limited number of the more important will be presented to illustrate the chemistry of complex cations in soil.

Examples of cations, which are present in significantly lower concentrations than the simple cations, are iron, manganese, zinc, copper, nickel, and cobalt. Except for cobalt, these have multiple oxidations states in soil as shown in Table 10.1. Because of their multiple oxidation states they may be present as

Table 10.1. Common Important Ionic Species in Soil[a]

Element/Compound	Cations[b]	Anions
Nitrogen	NH_4^+	NO_3^-, NO_2^-[c]
Phosphorus	None	$H_2PO_4^-$, HPO_4^{2-}, PO_3^{3-}
Potassium	K^+	None
Sulfur	None	S^{2-}, SO_4^{2-}
Chlorine	None	Cl^-[d]
Carbon	None	HCO_3^-, CO_3^{2-}
Iron	Fe^{2+}, $Fe(OH)_2^+$, $Fe(OH)^{2+}$, Fe^{3+}	None
Manganese	Mn^{2+}, Mn^{3+} Mn^{4+}[e]	None
Copper	Cu^{2+}, $Cu(OH)^+$	None
Zinc	Zn^{2+}, $Zn(OH)^+$	None
Nickel	Ni^{2+}, Ni^{3+}	None
Boron	None	$H_2BO_3^-$
Cobalt	Co^{2+}	None
Molybdenum	None	MoO_4^{2-}, HMO_4^-

[a] Only elements and compounds commonly found in multiple oxidation states are listed.
[b] In some cases species in this column may exist in soil under certain conditions; however, they are not common.
[c] Nitrogen oxides and nitrogen gas from denitrification will also be present.
[d] Any halide found in soil solution will be present as its anion.
[e] Often found in mixed oxidation states.

many more species than the simple cations. Typically the higher oxidation states predominate under oxidizing conditions, while the lower oxidation states predominate under reducing conditions. However, it is common to find both or all oxidation states existing at the same time in either an aerobic or an anaerobic soil [7,8].

10.1.2.1 Iron

Iron and its various oxidation state species are very common components of the environment. In addition to the common simple oxides FeO add Fe_2O_3, it is found in minerals such as hematite, goethite, and ferrihydrite, and in a number of hydroxy and oxy compounds. Because of its common occurrence in the environment in general and in soil in particular, the total iron content of soil is seldom a useful piece of information.

Most commonly iron is discussed as being in either the ferrous (Fe^{2+}) or ferric (Fe^{3+}) state. Changes between these two depend on the soil's pH and Eh as discussed in Chapter 4. Acid conditions and low Eh values tend to lead to the production of ferrous ion, while high pH and high Eh values result in predominance of ferric ion. It should be noted that ferrous ion is more soluble than ferric ion and thus will be more available to plants.

Iron cations in both the ferrous and ferric states can act as exchangeable cations; however, ferric ion is generally unsoluble and thus not present on exchange sites as such while its other species involve other oxidation states, and compounds of iron, oxygen, and hydroxy groups tend to form other cationic species which may be exchangeable. There are still other species involving other ligands and ferrous and ferric ions that are chelated, thus forming yet other species. Any compound having atoms with electron pairs that can be shared with positive species will associate with iron cations in soil. Any iron species may become attached to soil components such as sand, silt, clay, and inorganic and organic colloids to form still more species.

Because of its common occurrence and biological importance, it is an essential micronutrient for most organisms, a number of analytical procedures (see Bibliography) for analysis of iron species have been developed and concentrate on biologically available species [9].

10.1.2.2 Manganese

Manganese in soil has many characteristics that are similar to those of iron; for instance, it exists in multiple oxidation states: Mn^{2+}, Mn^{3+}, and Mn^{4+}. Although manganese can exist in the laboratory in other oxidations states, these are the ones most common in soil. Manganese forms various oxide and hydroxide species and chelates with many components. Its low oxidation state (i.e., Mn^{2+}) is more soluble and more available than is its high oxidation state (i.e., Mn^{4+}).

Manganese does, however, have some unusual characteristics. It is very unusual to find soil situations where iron is toxic, whereas manganese toxicity is known. As noted above, iron is found in only two oxidation states while manganese can have three oxidation states. However, the situation is found to be much more complex than this when soil is analyzed for the species of manganese present. A simple analysis for manganese might indicate an oxidation state of +3.5, indicating that the material analyzed contains an unknown mixture of the common oxidation states of soil manganese. This then creates problems in understanding the chemistry of manganese and thus reactions leading to its deficiency, toxicity, biological availability, and movement in the environment. This is an issue and problem which has received significant attention and research [10].

10.1.2.3 Chromium

Chromium has numerous oxidation states, some of which are strongly oxidizing. The most highly oxidized species is Cr^{6+}, and the reduced ion is Cr^{3+}. In soil, because of its strong oxidizing characteristics and oxidizable species present, Cr^{6+} is rapidly reduced to Cr^{3+}. Chromium species of intermediate oxidation states can exist in soil; however, the +3 state is the most common. As with the other metals, all the possible combinations of species with other components are possible and must be kept in mind when carrying out an analysis for chromium species in soil [11–13].

10.1.2.4 Mercury

Mercury is unusual in that it is found in the environment as both oxidized mercury ions and reduced methyl mercury. The mercurous (Hg^{+}) ion is unstable and not likely to be found in soil, while mercuric (Hg^{2+}) ions and methyl mercury compounds are. All forms of mercury are of environmental concern, and mercury ions can form the same types of interactions with soil constituents as those described for other multi-oxidation-state metals. Mercury in all its forms is toxic and thus of concern; however, methyl mercury, which can form in soil under anaerobic conditions, is particularly dangerous because of its extreme toxicity.

Mercury has several other characteristics that render it of particular environmental concern and make it likely to be found as many different species. It is a natural constituent of soil, although it occurs at low concentrations. It is widely used in both industry and in the laboratory, making it a common contaminant of reference materials. Metallic mercury has a relatively high vapor pressure, which means that it can occur in measurable amounts in the soil atmosphere.

Analysis of mercury is difficult, and specialized sampling and instrumental techniques are generally required to carry out an accurate analysis. Although

atomic absorption is applicable, it requires specialized heating of the sample such as a graphite furnace and other specialized sample handling [14,15].

10.1.2.5 Aluminum

Aluminum deserves special attention because, although it is present in only one oxidation state, it is commonly associated with both oxygen and hydroxy groups and is an extremely important ion, particularly in acidic soils. Although it is not toxic to most animals, it is toxic to most plants and is regarded as being present only in the Al^{3+} oxidation state. However, Al^{3+} reacts with water, releasing protons into the soil solution. Under acid conditions aluminum is more soluble and thus some of the following reactions lead to additional acidity. Reactions of aluminum ions in soil solution with the release of protons are shown in equations below:

$$Al^{3+} + H_2O \rightleftharpoons AlOH^{2+} + H^+ \quad (10.1a)$$

$$AlOH^{2+} + H_2O \rightleftharpoons Al(OH)_2^+ + H^+ \quad (10.1b)$$

$$Al(OH)_2^+ + H_2O \rightleftharpoons Al(OH)_3 + H^+ \quad (10.1c)$$

Other reactions of aluminum lead to the formation of other species. In the solid inorganic compartment it is most commonly found octahedrally bonded to a combination of oxygen atoms and hydroxy groups. In solution it may be in any one of the species shown in reactions (10.1). It may also be bonded or associated with the colloidal inorganic and organic particles and the surfaces of other soil components.

Aluminum is always present in soil as it is a constituent of soil minerals, particularly clay minerals. As the pH of soil decreases, aluminum from various sources is brought into solution. At very low pH levels, the very fabric of soil begins to erode, which causes two things to happen:

1. The soil has very high buffering capacity at this point because the added acid is decomposing the inorganic components in soil. This means that a large amount of acid is needed to decrease the pH of soil to a point where metals are solubilized and can be leached out.
2. The soil itself is destroyed and hence at the end of the extraction process soil is no longer present, and what is left is a mixture of highly acid salts, which must be disposed of. It is for this reason that extraction or remediation methods that depend on the acidification of soil to low pH fail and should never be undertaken [4,16,17].

10.2. ANIONS

Simple anions are those that exist in only one oxidation state in soil and generally are associated only with water. Complex anions are typically oxyanions of nonmetals, although molybdenum occurs as an oxyanion [4].

10.2.1. Simple Anions in Soil

There is only one simple anion commonly found in soil, and that is chloride (Cl^-). Chloride is an essential nutrient for plants but is typically present in sufficiently high concentrations that deficiencies are never observed. If other halogens are present, they will also be simple anions. Most soils contain small amounts of bromide as the second most common simple anion; however, in some cases significant levels of fluoride and iodide may be present, although this is rare. Inorganic combinations of all these anions are soluble in water and thus this tends to be their predominate species. However, they may be combined with other components and so may be present as other species; for instance, fluorine is a component of phosphates and organic compounds, and chlorine and bromine are components of chloro and bromo organic compounds such as insecticides, dichloromethane, and other solvents. There are also other nonionic species of these elements that may be present [18].

10.2.2. Complex Anions in Soil (Oxyanions)

Many important soil components are not present as simple cations or anions but as oxyanions that include both important metals and nonmetals. The most common and important metal oxyanion is molybdate (MoO_4^{2-}). The most common and important nonmetal oxyanions are those of those of carbon [bicarbonate (HCO_3^-) and carbonate (CO_3^{2-})], nitrogen, [nitrate (NO_3^-) and nitrite (NO_2^-)], and those of phosphorus, [monobasic phosphate ($H_2PO_4^{2-}$), dibasic phosphate (HPO_4^{2-}), and tribasic phosphate (PO_4^{3-})]. The soil chemistry of oxyanions is complicated by the fact that some act as simple anions and move readily through soil while others react with numerous soil constituents, forming insoluble immobile constituents. Common oxyanions in soil and their chemical characteristics and mobility are summarized in Table 10.2.

Molybdate, although present in small amounts in soil is an essential nutrient for nitrogen fixation, specifically in the enzyme nitrogenase. The mobility of molybdate in soil is limited, and so this anion does not move readily through soil.

Of the nonmetal oxyanions, those of carbon have a role in soil different from those of nitrogen and phosphorus. Bicarbonate and carbonate can act as counterions to cations to keep the soil electrically neutral. They are also important because all pH changes in soil tend to involve either carbonate or bicarbonate, and thus they are both involved in soil pH and buffering.

Both nitrogen and phosphorus oxyanions are important because they are sources of nitrogen and phosphorus for plants and their potential for causing water pollution. Nitrogen oxyanions, nitrite and nitrate are of great interest because they are readily formed in soil from organic matter and inorganic nitrogen containing compounds, particularly ammonia (NH_3). Soil must be moist but not saturated, with a temperature above 20°C for rapid oxidation of ammonia to nitrite and nitrate. Both oxyanions are mobile in soil and so can be leached into groundwater and find their way into lakes, ponds, and drinking water. Nitrite and nitrate are readily available to plants and can move

Table 10.2. Common Oxyanions in Soil, Their Chemical Characteristics and Mobility

Oxyanion	Chemical Characteristics	Mobility
Carbonate	Forms insoluble carbonates with cations	Precipitates from solution
Bicarbonate	Forms slightly soluble bicarbonates with cations	Mobile in solution
Nitrate	Available to plants. Converted to N_2 gas under anaerobic conditions	Mobile
Nitrite	Readily oxidized to nitrate	Mobile
Monobasic phosphate	Stable at low pH	Considered immobile
Dibasic phosphate	Stable under neutral conditions	Considered immobile
Tribasic phosphate	Stable under basic conditions	Considered mobile
Molybdate	Reacts with various soil constituents	Some mobility
Borate	Acts as simple anion	Mobile

through soil to roots with water taken up by plants and are readily converted to nitrogen gas under the proper soil and environmental conditions, the most important of which are temperature; between 20 and 40°C is optimal and saturated soil.

Phosphorus oxyanions are entirely different from nitrogen oxyanions. The oxyanion species present is controlled by the pH; also, phosphate oxyanions are generally not mobile in soil; sandy soils and soils high in phosphorus are exceptions to this rule. Any soil, however, can loose phosphate by erosion, and this phosphate can cause environmental problems. Because of its unique chemistry, phosphorus will be discussed separately below.

Boron and arsenic are natural components of soil and are both present as oxyanions. Boron is present as boric acid or borate polymers, and arsenic is present as arsenate. While boron is weakly held by soil, arsenic is similar to phosphate in its interactions with soil constituents. Boron is an essential nutrient for plants; however, it is also toxic at relatively low levels. Arsenic is toxic. The laboratory chemistry of both of these elements is well understood but their environmental chemistry and speciation is less well understood [19–23].

10.3. NITROGEN

Nitrogen is unique because it is found in soil as both reduced and oxidized species, as part of organic molecules, and as oxidized gaseous species, as well as in the elemental form (i.e., nitrogen gas). Many nitrogen species are soluble in water and thus tend to be found in the soil solution as opposed to attached to soil particles. There are, however, three important exceptions to this rule. Ammonium having a positive charge can act as a cation and thus be attracted

to soil cation exchange sites and be an exchangeable cation. In the fine-grained mica-type clays, ammonia can be trapped between clay sheets. In this condition the ammonium is not available to plants and is seldom biologically available.

The third group is amino acids and proteins. While amino acids are soluble in water and thus are expected to move readily, they are also zwitterions (having both positive and negative charges) and thus may interact with charges on soil particles, depending on the pH, and move slowly or not at all. Proteins are polymers of amino acid and may or may not be soluble but can have charges as do amino acids. There are also a large number of other types of nitrogen containing compounds, for example, amino sugars and nitrogen containing lipids, present in soil, and some will be soluble while others are insoluble.

Otherwise, nitrogen moves between the various soil compartments easily, depending on the soil environmental conditions. These movements are often outlined as the nitrogen cycle (which can be found in the text by Coyne [24]). The slowest and most energy-intensive part of the cycle is the "fixation" of nitrogen gas (N_2) forming various nitrogen compounds with either oxygen, forming oxyanions (hydrogen), forming ammonia, or with carbon to form amino acids. Once these initial compounds are formed, subsequent changes in nitrogen species are fairly rapidly and easily accomplished.

Ammonium (NH_4^+) may be added as a fertilizer or released during organic matter decomposition. It is the chief species present immediately after the injection of anhydrous ammonium into soil (as a fertilizer). Ammonium will be present in solution, as a cation, attracted to the cation exchange sites, and it may also become trapped between clay layers. It is labile in soil in that, under aerobic, moist, moderate temperature conditions, it is rapidly oxidized to nitrite and nitrate. Ammonia may also be volatilized from the soil solution, particularly under basic soil conditions.

Oxidized species, chiefly nitrite and nitrate, of nitrogen occur in all soils in the soil solution. Although nitrite in the environment is of concern, its occurrence is usually limited because the oxidation of nitrite to nitrate is more rapid than the oxidation of ammonia to nitrite. Both nitrite and nitrate move readily in soil, and nitrate is available to plants as a source of nitrogen and can move to plant roots with water.

As a result of these reactions and volatilization, nitrogen species concentrations are expected to change during sampling and sample storage. This is particularly true if precautions are not taken to limit this eventuality [2,23,25–27].

10.4. PHOSPHORUS

To understand the occurrence of phosphate oxyanions in soil, the titration of phosphoric acid is instructive. Phosphoric acid is triprotic, and each of the protons has a different pK_a. Thus the titration curve has three inflections

reflecting the titration of each of these protons. At low (acidic) pH (~2), the chief form is phosphoric acid. Above this the primary form is $H_2PO_4^-$, which is called *monobasic phosphate*; at higher pH the chief form will be dibasic phosphate (HPO_4^{2-}), and finally at high pH the form will be tribasic (PO_4^{3-}). It might assumed that the species of phosphorus found in soil would be controlled solely by pH. This is not the case. The phosphate species found is dependent in large measure on pH but is also dependent on organic matter. Lower soil pH (i.e., 4–7) favors monobasic phosphate, while higher pH (i.e., 7–10) favors dibasic phosphate.

Phosphorus occurs almost exclusively as either monbasic or dibasic phosphate in the various soil compartments, including biological tissue. However, these two species react with a host of both inorganic and organic components, forming a multitude of other species. Often overlooked are the species that form where phosphate reacts with surface constituents on soil particles, including colloidal inorganic an organic particles. Species containing phosphorus, other than phosphate or compounds containing phosphate, can sometimes be found, but this is unusual.

Organic phosphorus associations can occur under both acidic and basic conditions. Phosphorus can form esters with organic alcohol functional groups and can be associated with amine groups in various ways.

Phosphate reacts and forms insoluble compounds with iron, aluminum, and calcium. Under acid soil conditions both iron and aluminum become more soluble, and thus as soil pH decreases, its "phosphate fixing power" increases. This means that iron and aluminum react with phosphate to form insoluble and plant unavailable iron and aluminum phosphate species. Under basic conditions, high concentrations of calcium exist and insoluble calcium phosphates form. Insoluble phosphates are formed with other metals that happen to be present; however, the three mentioned are generally present in the highest concentration, and so they represent the major reactants with phosphate. Iron, aluminum, and calcium phosphates can also occur as coatings on soil particles.

When analyzing soil for phosphorus, all these forms or potential forms must be kept in mind. It is to be expected that all soluble forms of phosphorus will be available to plants while all insoluble forms will not. However, precipitation processes will also play a role in phosphorus availability. Initial precipitation results in small crystals with high surface areas and thus greater reactivity and tendency to move into solution when the concentration of phosphorus in solution decreases as with plant uptake. On the other hand, as time passes, the crystals grow larger, thus decreasing surface area, reactivity, and availability [28].

10.5. SAMPLING, SAMPLE STORAGE, AND SPECIATION

The problem of species changes during sampling and storage can be ameliorated in three ways. First the soil component of interest can be measured in

situ, thus removing problems associated with sampling and extraction. This approach, although optimal, is feasibly in only a few instances. The second approach is to make the analysis as quickly after sampling as possible, preferably in the field, so as to eliminate storage problems. In this case minutes or seconds between sampling and analysis are desirable.

When these approaches are not possible, steps must be taken to account for changes occurring during sampling and storage. These can be in either direction; that is, the concentration of a species may increase or decrease or be converted to an entirely different species. It is not possible to give a prescription for sampling or storage that will guarantee the stability of a soil species over a period of time. Some species will be sensitive to oxidation while others will be changed by the lack of oxygen. In some cases species will be stable in air dry soil while others will be more stable in moist soil. In some cases refrigeration at 0°C is best while other species will require storage at –40°C or even lower temperatures. In yet other cases storage at room temperature will be sufficient. The solution to sampling and storage problems is to study the stability of the species of interest under various sampling and storage conditions and thus determine which is best.

Some generalized sampling and storage recommendations, however, can be made. During sampling and storage samples must be kept under the same oxygen concentration, temperature, and pressure conditions occurring in the field, if species integrity is to be maintained. Samples taken from anaerobic, low- or high-temperature, or high-pressure conditions should be maintained under these conditions during sampling, storage, and until actual analysis is carried out. A sample taken from the bottom of a lake will be under pressure and low-oxygen or anaerobic conditions. It may also be at a significantly lower temperature than surface water, air, or soil temperatures. To maintain sample integrity, it must be kept under these conditions [2].

10.6. CONCLUSIONS

This discussion is not intended to be a comprehensive discussion of all the elements, cations, anions, and organic species present in soil. It includes a range of such species that illustrate the common situations related to the analysis of cations, anions, and oxyanions in soil. Different species have different biological reactivity and availability, and thus it is important to know which species is present. Cations have water molecules surrounding them, and some cations react with water, forming oxy and hydroxy cations. Other cations can be present in multiple oxidation states and may be present in mixtures of these oxidation states. Some species added to soil will rapidly be converted to other species, such as the conversion of Cr^{6+} to Cr^{3+}, such that analysis for the original species may be fruitless, especially if the soil sample has been stored for any length of time. All species may be associated in various ways with the inorganic, organic, and colloidal components of soil to form species of interest.

Some oxyanions are soluble in the soil solution and readily move in soil, whereas other oxyanions are immobile and do not move in soil. In some cases species of interest may be combined into new compounds such as to be readily, slowly, or nonavailable to plants.

PROBLEMS

10.1. Give some examples of anions including oxyanions that are common in soil.

10.2. Give examples of cations including oxy and hydroxy cations common in soil.

10.3. Write reactions that illustrate how metal cations can lead to the release of protons into the soil solution.

10.4. Illustrate solution, cation exchange, and outer-sphere and inner-sphere species around a soil particle.

10.5. Compare the reactivity and movement of nitrogen and phosphate oxyanions in soil.

10.6. Explain why the occurrence of colloidal species is important in the analysis of soil for other species.

10.7. Compare the characteristics of manganese and iron in soil, and describe their similarities and differences.

10.8. What special roles do carbonate and bicarbonate play in the chemistry of soil?

10.9. Explain why analysis of soil for chromate +6 (Cr^{6+}) species is generally not an enlightening analysis.

10.10. Diagram the compartments in the soil environment and the movement of species between these compartments.

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