

Chapter 15

Transformations of Sulfur

James J. Germida

... sulfur is a devilish substance. ... discharged from the bowels of the earth, by volcanoes or evil-smelling hot springs ... surely the effluent of Hell itself. ...

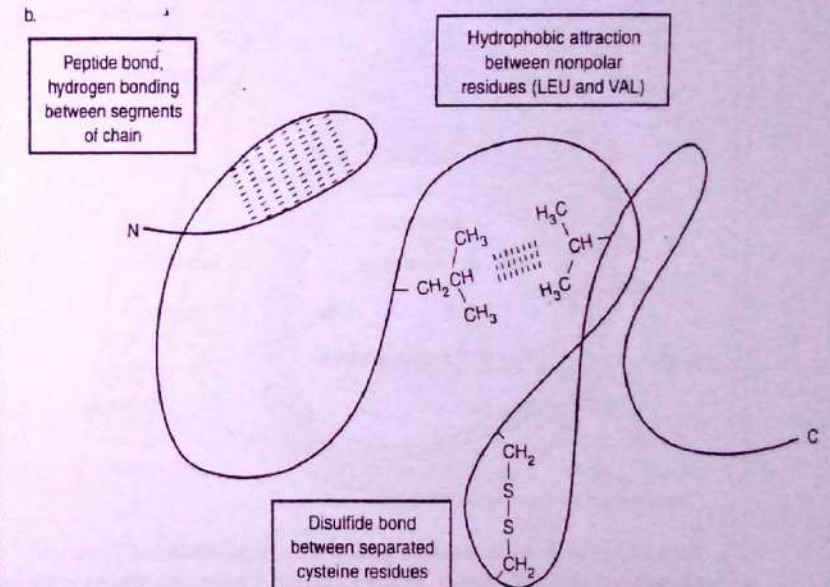
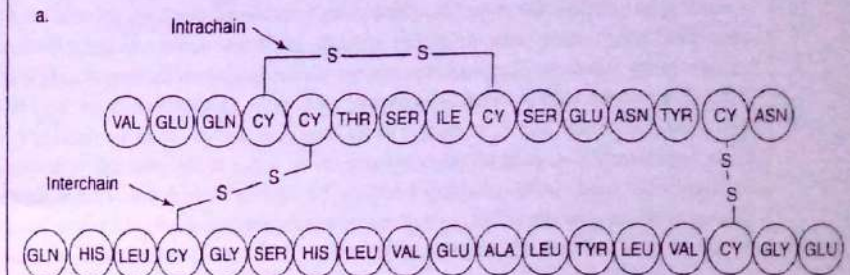
J.R. Postgate

Sulfur (S) is an essential element for the growth and activity of all living organisms. It is one of the ten major bio-elements required by organisms in relatively high concentrations (i.e., $> 10^{-4}M$). Sulfur is required for the synthesis of the amino acids cysteine, cystine, and methionine. It plays an active role in plants, animals, and microorganisms as an important constituent of vitamins, hormones, and structural components, and for other metabolically important molecules such as coenzyme A. For example, the **disulfide bond** formed between cysteine residues helps stabilize the tertiary structure of proteins (Box 15-1).

Sulfur is an important source of metabolic energy for many bacteria. For example, certain chemoautotrophic bacteria obtain energy for cell growth and division by oxidizing reduced sulfur compounds. In fact, some exotic ecosystems, such as hot sulfur springs and hydrothermal vent communities, are driven by energy generated in the oxidation of sulfur, which they use to fix carbon (Box 15-2).

This chapter introduces the basic principles of the sulfur cycle and the microorganisms that drive it. This information will give one an appreciation of how important it is to understand the biogeochemistry of sulfur and other elements (Box 15-3).

Disulfide Bonds Stabilize Protein Structure. A disulfide bond is a covalent linkage between two sulfur atoms. This type of linkage is commonly found in polypeptides and protein molecules and may occur as an interchain or intrachain bond between sulfur-containing amino acids. This type of bond is not common to all proteins, but is usually critical to those that do possess it.

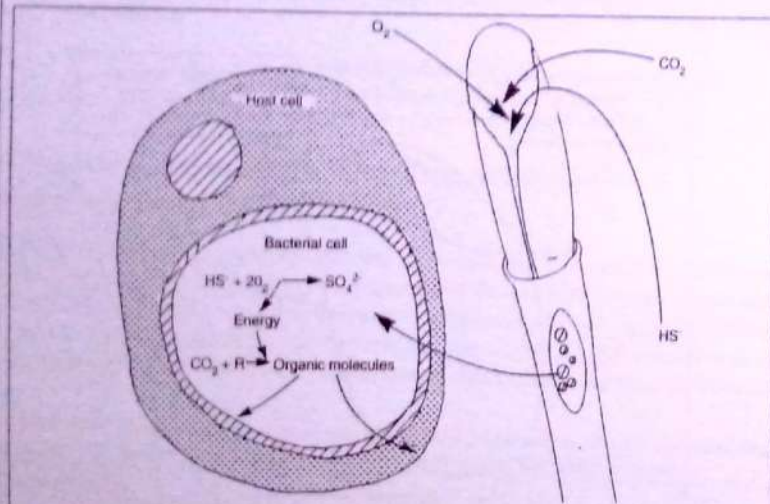


(a) Amino acid sequence of two polypeptide chains illustrating how intrachain and interchain disulfide covalent bonds form between cysteine residues. (b) Diagrammatic representation of how disulfide bonds and other stabilizing forces confer a unique spatial conformation on proteins. Not all types of forces or their frequency of occurrence are shown.

Box 15-2

Life Without Light: The Importance of Chemosynthesis. All life requires energy. Furthermore, it is clear that the sun's energy, trapped through photosynthesis, is the driving force for **primary productivity** in all ecosystems. Almost all, that is! About 20 years ago scientists made one of the most startling discoveries of twentieth-century biology. They found bizarre, exotic biological communities living in the deep ocean where hydrothermal fluids rise through the earth's crust. The communities at these deep sea thermal vents consist of giant tubeworms and masses of large clams. Life here is sustained by primary productivity based on microbial sulfur oxidation (Jannasch and Mottl, 1985).

Hydrogen sulfide is the most abundant compound in the deep sea thermal vent fluid. This compound is extremely toxic to higher animals, but some sulfur-oxidizing bacteria can use it as an energy substrate. Considerable energy is released when hydrogen sulfide is oxidized. Through a process called **chemosynthesis**, analogous to photosynthesis, vent bacteria employ chemical rather than light energy to fix inorganic carbon to make organic compounds. More surprisingly, the giant tubeworms and clams living at the vent are in a symbiotic relationship with these sulfur-oxidizing bacteria that live in their tissue. This is truly a remarkable example of how life adapts to extreme environments and is found where least expected. In fact, it now appears that microbial life exists in all environments with an oxidizable energy source and favorable conditions for microbial life.



Bacteria found in symbiosis with tube worms at hydrothermal vents use chemosynthesis to fix carbon to make organic compounds. Shown is the breathing organ attached to the body of the worm, *Riftia pachyptila*, which contains the symbiotic association. The bacteria use the hydrosulfide ion (HS^-) and oxygen (O_2) to generate chemical energy (chemosynthesis). The hydrosulfide ion serves as the electron donor; the energy is released when HS^- and O_2 are combined, driving a series of reactions called the Calvin cycle. In the Calvin cycle, organic compounds are formed as the carbon dioxide (CO_2) present in both vent water and sea water is fixed. In this illustration, R stands for the auxiliary compounds, including CO_2 -fixing enzymes, involved in the cycle. Note that sulfuric acid is produced as a by-product. The process is analogous to photosynthesis, in which green plants use light energy to fix carbon dioxide and form organic compounds.

Redrawn from Tunncliffe (1992). Used with permission.

Box 15-3

Sulfur Can Be Good or Bad for the Environment. Although sulfur is considered a macronutrient, its availability to the biota can be limiting in some ecosystems; then it must be applied as a supplement or fertilizer. However, in other instances, too much sulfur is a pollutant. For example, burning high-sulfur-content (i.e., bituminous and lignite) coals produces gaseous sulfur emissions that can become components of acid precipitation. Microbial oxidation of reduced sulfur minerals associated with coal seams and the subsequent leaching of the resulting oxyanions (e.g., SO_4^{2-}) gives rise to acid mine drainage that can pollute streams or acidify reclaimed soils. Finally, sulfur is important for many industrial processes; it can be used, for example, in the production of chemicals, concrete, and asphalt. As one reads this chapter, it will become evident that sulfur is an essential, versatile, and economically and ecologically important element.

The Sulfur Cycle in Agroecosystems and Terrestrial Environments

The sulfur cycle bears many similarities to the nitrogen cycle. Both of these elements exist in a number of oxidation states and undergo similar types of chemical reactions and biological transformations, including volatilization. The majority of sulfur is found in the **lithosphere** (Table 15-1). Most nitrogen on earth is also in the lithosphere; however, dinitrogen in the atmosphere is the major pool of biologically available nitrogen. Only a small portion of the sulfur pool is found in the atmosphere, and most sulfur that cycles through the atmosphere is because of human activities. In fact, since the Industrial Revolution, increased burning of fossil fuels has almost doubled the rate of sulfur entering the atmosphere to approximately $1.5 \times 10^{11} \text{ kg S yr}^{-1}$. The volatilization

Table 15-1 Estimated quantities of sulfur in major sulfur pools.

Pool	Mass (kg S)
Atmosphere	3.6×10^9
Hydrosphere	1.3×10^{18}
Oceans	1.3×10^{18}
Marine organisms	2.4×10^{10}
Fresh waters	3.0×10^{12}
Ice	6.0×10^{12}
Lithosphere	24.1×10^{18}
Igneous rocks	5.0×10^{18}
Metamorphic rocks	11.4×10^{18}
Sedimentary rocks	7.7×10^{18}
Evaporites	5.1×10^{18}
Shales	2.0×10^{18}
Limestones	0.1×10^{18}
Sandstones	0.3×10^{18}
Soils	2.6×10^{16}
Soil organic matter	1.0×10^{14}
Biosphere	7.6×10^{12}

Table 15-2 Amounts and distribution of sulfur in some world soils.

Location	Type of soil	Total sulfur ($\mu\text{g g}^{-1}$)
Saskatchewan, Canada	Agricultural	88-760
British Columbia, Canada	Grassland	286-928
	Forest	162-2,328
Iowa, U.S.	Organic	1,122-30,430
	Agricultural	214-438
Carolinas, U.S.	Agricultural	57-618
Hawaii, U.S.	Tidal marsh	3,000-35,000
Eastern Australia	Volcanic	180-2,200
	Agricultural	38-545
Nigeria	Agricultural	25-177
Brazil	Agricultural	43-398

Modified from Paul and Clark (1989).

of sulfur as hydrogen sulfide, carbonyl sulfide, and dimethyl sulfide, for example, from marine algae, marsh lands, mud flats, plants, and soils also contributes to the global circulation of sulfur through the atmosphere.

The nature and quantities of the various sulfur pools in surface soils are the basis for sulfur cycling in terrestrial environments. These sulfur pools are influenced by pedogenic factors such as climate, regional vegetation, and local topography. For example, the total sulfur content of soils ranges from 0.002 to 10%, with the highest levels in tidal flats, and in saline, acid sulfate, and organic soils. The impact of pedogenic factors on the total sulfur concentrations in the surface is clear when comparing values for soils from diverse geographic areas, as shown in Table 15-2.

Nature and Forms of Organic and Inorganic Sulfur in Soil

Organic sulfur constitutes more than 90% of the total sulfur present in most surface soils. However, the precise nature of the organic sulfur compounds in soil cannot be clearly identified. Thus, organic sulfur is grouped into two broad categories, organic sulfates and carbon-bonded sulfur. Examples of these organic compounds are given in Figure 15-1. Organic sulfates (R-O-S) include sulfate esters (C-O-S), sulfamates (C-N-S), and sulfated thioglycosides (N-O-S). Organic sulfates constitute 30 to 75% of total organic sulfur in soil. Carbon-bonded S (C-S) includes the sulfur present in amino acids, proteins, polypeptides, heterocyclic compounds (e.g., biotin and thiamin), sulfonates, sulfones, sulfonates, and sulfoxides. A large portion of carbon-bonded sulfur present in soil has yet to be identified; however, in some cases the carbon-bonded sulfur of amino acids may constitute up to 30% of the organic sulfur in soil.

Inorganic forms of sulfur account for less than 25% of the total sulfur in most agricultural soils. Sulfur exists in a number of forms with a wide range of oxidation states (Table 15-3). Sulfide, elemental sulfur, sulfite, thiosulfate, tetrathionate, and sulfate are the main forms of inorganic sulfur in agricultural soils. Sulfate is the most common form of inorganic sulfur found in well-aerated agricultural soils, whereas sulfides account for less than 1% of total sulfur and measurable quantities of thiosulfate and tetrathionate are usually detected only in soils treated with sulfur fertilizer or those receiving pollutants.

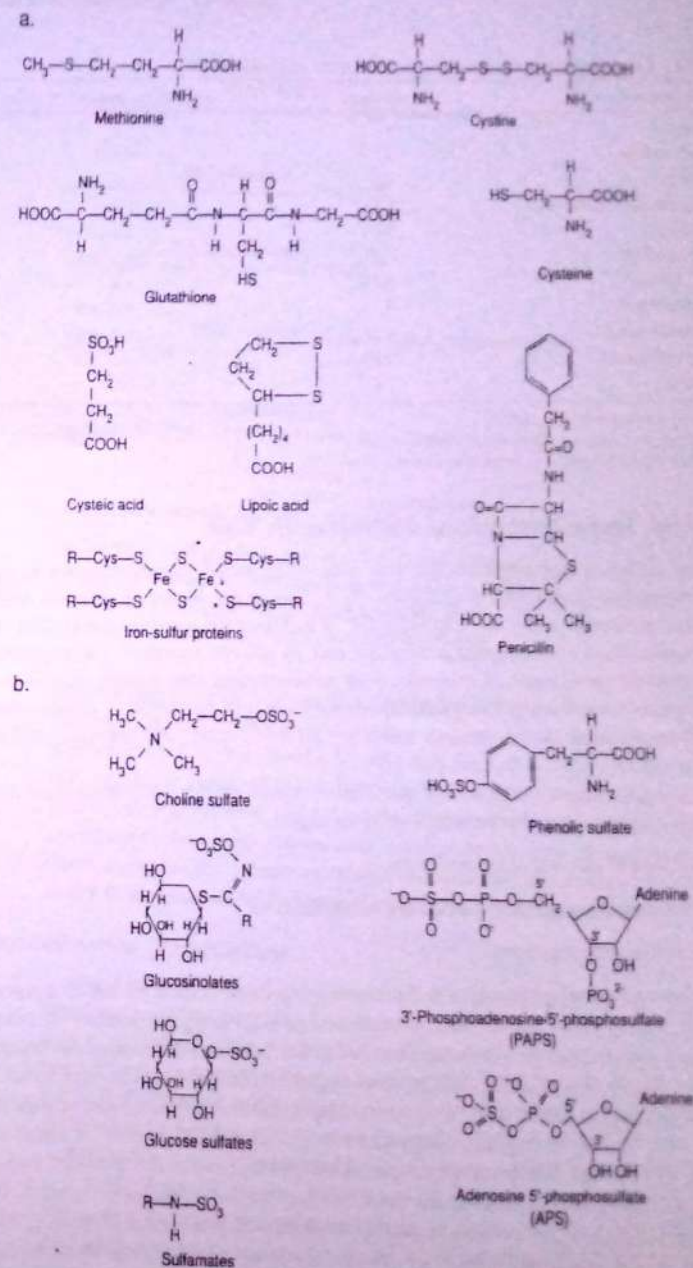


Figure 15-1 Examples of some of the organic sulfur compounds found in soils. (a) Amino acids and other compounds containing carbon-bonded sulfur. Note that lipoic acid also has a disulfide bond. (b) Compounds that possess ester sulfate bonds (e.g., C-O-S and C-N-S). This class of compounds is sometimes used by microorganisms to store sulfur, although some bacteria are able to store elemental sulfur. From Paul and Clark, 1989. Used with permission of Academic Press, Inc.

Table 15-3 Important forms of sulfur and their oxidation states.

Compound	Formula	Oxidation state(s) of sulfur
Sulfide	S^{2-}	-2
Polysulfide	S_n^{2-}	-2, 0
Sulfur*	S_8^0	0
Hyposulfite (dithionite)	$S_2O_4^{2-}$	+2
Sulfite	SO_3^{2-}	+4
Thiosulfate†	$S_2O_3^{2-}$	-1, +5
Dithionate	$S_2O_6^{2-}$	+6
Trithionate	$S_3O_6^{2-}$	-2, +6
Tetrathionate	$S_4O_6^{2-}$	-2, +6
Pentathionate	$S_5O_6^{2-}$	-2, +6
Sulfate	SO_4^{2-}	+6

From Narasimharthy et al. (1993).

*Occurs in an octagonal ring in crystalline form.

†Outer S has a valence of -1; inner S has a valence of +5

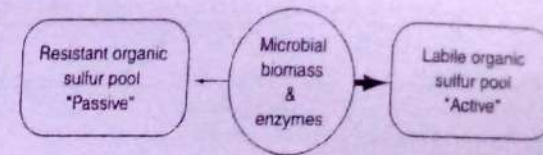
Microbial Transformations of Sulfur in Soil

The sulfur cycle—emphasizing soil and plant sulfur transformations in agroecosystems—is illustrated in Figure 15-2. The major forms of sulfur in soil include elemental sulfur (S^0), sulfides (S^{2-}), sulfates (S^{6+}), and organic sulfur compounds. Most of this sulfur enters the soil as soluble inorganic forms produced during the weathering of minerals, from fertilizers and atmospheric deposition, or as soluble organic and inorganic forms from the decomposition of organic matter. Losses of sulfur occur through leaching, surface runoff, volatilization, and crop removal.

Soil microorganisms drive the sulfur cycle. Hence, sulfur undergoes many microbially mediated transformations in soil, including:

- oxidation and reduction reactions,
- mineralization and immobilization reactions, and
- volatilization reactions.

The soil microbial biomass is the key driving force behind all sulfur transformations. This biomass acts as both a source and sink for inorganic sulfate, whereas microbial activity regulates both the fluxes of sulfur between different pools (inorganic sulfate, labile organic sulfur, and resistant organic sulfur) and the losses of sulfur from these pools (e.g., conversion of complex organic sulfur compounds into mobile forms that may be lost by leaching). Most of the sulfur in soil (75 to 90%) is found in organic complexes. These complexes are either stable, passive fractions that turn over very slowly or active, dynamic fractions that are readily transformed or metabolized. Actually, the continuum of sulfur organic complexes in soil ranges from very old, stable (e.g., organic sulfur found in soil humus) fractions to very young, short-lived (e.g., organic sulfur found in the amino acid cysteine) fractions. The microbial biomass is the engine for the conversion of passive fractions into active fractions, and vice versa. This is illustrated in the following diagram, where the relative flux of sulfur between pools is reflected in the size of the arrows depicting microbial conversion:



Understanding the sources, sinks, and transformations of sulfur in soil is crucial to ensure adequate supplies of sulfur for the biota of various ecosystems and to protect the environment from the detrimental effects of too much sulfur. Because microorganisms are so important for the conversions between the active and passive organic matter pools, any factor that disturbs or otherwise has an impact on

Atmosphere

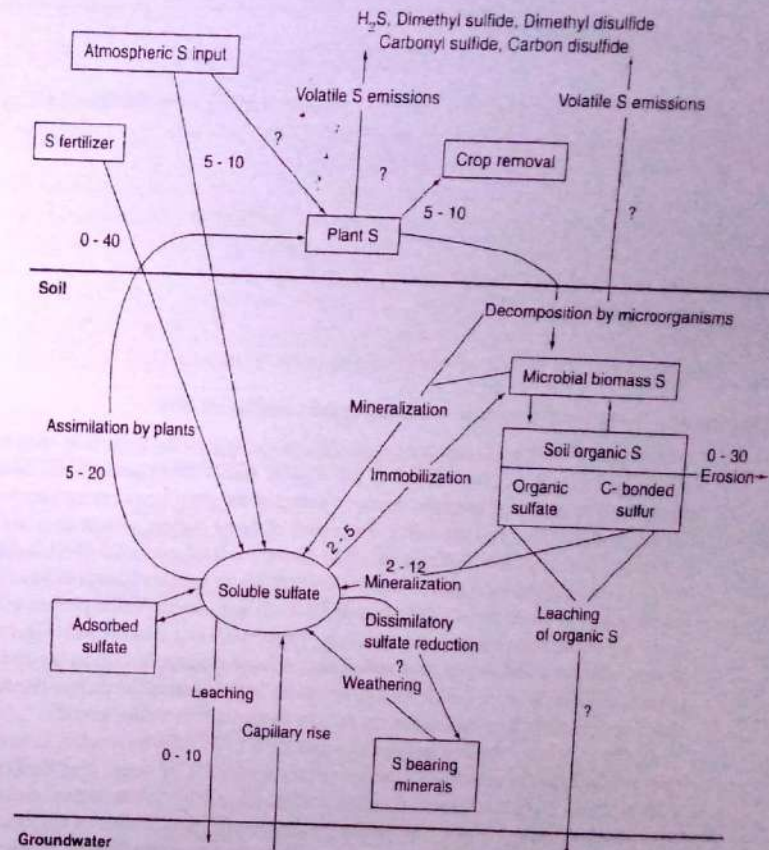


Figure 15-2 Conceptual sulfur cycle in agroecosystems. Numbers represent flux estimates ($\text{kg ha}^{-1} \text{yr}^{-1}$) for sulfur transformations in western Canadian soils. Adapted from Schoenau and Germida (1992). Used with permission.

Table 15-4 Total sulfur and ester sulfate content of selected microorganisms grown in culture with varying sulfur concentrations.

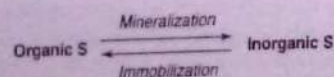
Organism	Total sulfur ($\mu\text{g g}^{-1}$ cells)			Ester sulfur (%)		
	S1*	S2	S3	S1	S2	S3
<i>Arthrobacter globiformis</i>	1,626	1,706	1,850	23	10	14
<i>Bacillus licheniformis</i>	ND	1,667	1,700	ND	6	8
<i>Bacillus</i> sp., soil isolate	1,142	1,054	ND	19	10	ND
<i>Micrococcus flavus</i>	2,398	1,500	1,950	7	8	9
<i>Pseudomonas cepacia</i>	2,477	ND	ND	16	ND	ND
<i>Fusarium solani</i>	ND	4,750	4,900	ND	13	21
<i>Penicillium nalgioeensis</i>	ND	1,815	2,450	ND	45	45
Soil isolate J-20	2,800	3,764	4,017	45	14	27
Soil isolate P-44	ND	5,527	6,400	ND	25	32
<i>Streptomyces</i> isolate 3-4L	ND	3,043	3,072	ND	14	16

From Gupta (1989)

*S₁ = Complex organic medium; S₂, S₃ = mineral salts medium containing 16 and 32 $\mu\text{g S ml}^{-1}$, respectively.

the microbial biomass influences sulfur cycling. For example, crop rotations and soil cultivation typically increase sulfur cycling in soil. When a soil is cultivated, it is mixed and churned and broken up into smaller pieces. This increases soil aeration and exposes soil particles and organic matter that were previously "hidden" from the soil microflora. Fresh organic matter, containing different ratios of carbon, nitrogen, phosphorus, and sulfur, is mixed into the soil. Some microbial biomass is activated due to the flush of available nutrients, and some of the biomass might be killed. For example, fungal hyphae help to hold soil aggregates together, as cultivation breaks up these aggregates, the hyphae are broken, resulting in dead biomass. This dead biomass is now available to be mineralized. As new microbial biomass is formed during decomposition of the newly exposed or added "active" organic material, nutrient elements (carbon, nitrogen, phosphorus, sulfur, and micronutrients) are cycled back and forth between active and passive states as organic and inorganic forms of the element. The overall consequence is increased nutrient cycling.

Similarly, interactions among different microbial groups, such as predation and parasitism, tend to increase the turnover rate of microbial biomass sulfur and hence affect sulfur fluxes. One can think of this in the following way. Bacteria and fungi store sulfur in the organic and inorganic sulfur compounds that comprise their cells (Table 15-4). When predators, such as soil amoebae, eat bacterial cells or pieces of fungal hyphae, that biomass material (labile or active sulfur pool) is broken down into smaller, nonmetabolized sulfur containing organic fragments resistant to further metabolism by the amoebae, which are excreted into the soil, along with any inorganic sulfur or metabolizable organic sulfur not needed by the amoebae for growth. The excess inorganic sulfur or metabolizable organic sulfur is now available for plants and other organisms to use. This is an example of the processes of **mineralization** and **immobilization** of nutrients.



Biological mineralization and immobilization are processes that occur concurrently and exhibit a strong relationship with the soluble sulfate pool in soil.

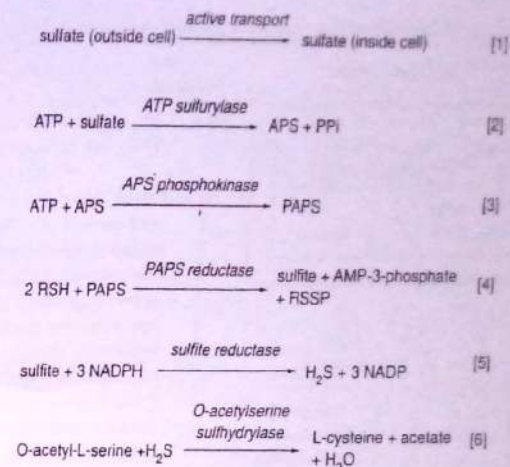


Figure 15-3 Assimilatory reduction of sulfate and formation of cysteine. RSH stands for thioredoxin; its reduced form is regenerated from the oxidized form through reduction by NADPH. Redrawn from Gotschalk (1979). Used with permission.

Immobilization occurs as a result of the microbial assimilation of nutrients that are then rendered unavailable for further plant or microbial uptake until the cell dies and is remineralized. Immobilization of sulfur may also involve precipitation as metal sulfide, especially pyrite, as in salt marshes. Because these transformations are mediated by microorganisms, soil factors that influence the growth and activity of microorganisms (e.g., pH, temperature, and moisture) also affect the rate of sulfur transformations. To estimate or predict the available sulfur status of soils, it is necessary to understand the factors that influence these processes.

Immobilization (Assimilation)

Microbial assimilation and conversion of inorganic sulfate into organic sulfur through the *assimilatory sulfate reduction pathway* leads to temporary immobilization of sulfur from plant or microbial availability. This process involves ATP sulfurylase and two energy-rich sulfate nucleotides, APS (adenosine 5'-phosphosulfate) and PAPS (3'-phosphoadenosine-5'-phosphosulfate). The overall reaction of $\text{SO}_4^{2-} \rightarrow \text{S}$ incorporation into amino acids is shown in Figure 15-3.

Most of the sulfur accumulated by microorganisms is in the form of amino acids in proteins; however, microorganisms also accumulate sulfate esters, sulfonates, vitamins, and cofactors. Some microorganisms, such as fungi, accumulate especially large amounts of sulfate esters (Table 15-4; Fig. 15-1). This is important because organic sulfates (e.g., sulfate esters and thioglucosides) are considered to be the most labile form of organic sulfur in soil and may comprise up to 30 to 70% of the organic sulfur in surface soils. The relative proportion of fungal biomass to bacterial biomass in soil (approximately 2:1) underscores the potential importance of microorganisms accumulating ester-sulfur compounds.

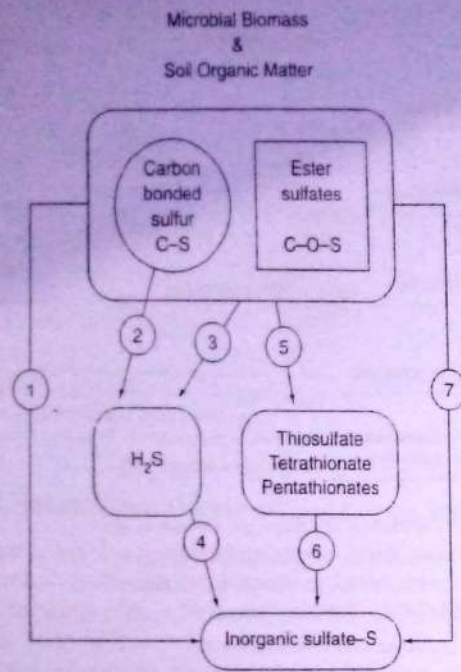


Figure 15-4 Known pathways for mineralization of organic sulfur compounds in soil: (1) biological (direct) mineralization during the oxidation of carbon as an energy source; (2) hydrolysis of cysteine by cysteine desulfhydrase; (3) anaerobic mineralization (desulfurization) of organic matter; (4) biological oxidation of hydrogen sulfide to sulfate through elemental sulfur and sulfite; (5) incomplete oxidation of organic sulfur into inorganic sulfur compounds; (6) biological oxidation of tetrathionate to sulfate through sulfide; and (7) biochemical (indirect) mineralization when sulfate esters are hydrolyzed by sulfatases. Adapted from Laurence (1987).

Typically, the addition of inorganic $\text{SO}_4^{2-}\text{-S}$ to soil leads to its quick incorporation into the organic sulfur fractions via microbial assimilation. The rate and magnitude of this immobilization is increased in the presence of an energy source, such as metabolizable organic matter or addition of easily degradable carbon sources like glucose. Later, much of this accumulated sulfur is found in the fulvic acid fractions, especially as organic sulfates.

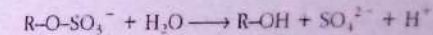
Mineralization

Mineralization of organic sulfur in soil is largely mediated by microbial activity. The various known pathways of sulfur mineralization are summarized in Figure 15-4. Carbon-bonded sulfur is mineralized either through oxidative (aerobic) decomposition or desulfurization (anaerobic) processes, whereas various sulfatases are involved in the mineralization of sulfate esters (Box 15-4). The mineralization process may be direct (i.e., cell mediated), involving viable microbial cells, or indirect (i.e., cell-free, enzyme mediated), involving enzymes such as sulfatases. In the case of direct mineralization, elements such as nitrogen and sulfur in direct association with carbon are mineralized as microorganisms oxidize the organic carbon compounds to obtain energy. Heterotrophic soil microorganisms decompose organic sulfur compounds to grow, as the carbon-sulfur bond is broken, the sulfur is released, usually as sulfide. Because this process involves actively growing microorganisms, their requirement for sulfur may meet or even exceed the sulfur supplied by the substrate. Thus net mineralization of sulfur by this process may not be reflected by in-

creases in the sulfate-sulfur pool in soil. In the case of indirect mineralization, those elements that exist as sulfate esters are hydrolyzed by intracellular or extracellular enzymes. This process, also known as *enzymatic mineralization*, occurs mainly outside the cell and may be regulated by end product inhibition, or the sulfate level. Direct mineralization is controlled by the microbial need for carbon and energy sources, whereas indirect mineralization is controlled by factors influencing enzyme synthesis, activity, and kinetics.

Box 15-4

Enzymes Drive Mineralization of Sulfate Esters in Soil. Sulfatases (technically referred to as sulfohydrolases EC 3.1.5) are enzymes that hydrolyze sulfonic acid esters, where the linkage with sulfate is in the form of R-O-S and R represents a diverse group of organic moieties. This reaction can be shown as follows:



Arylsulfatase is by far the most studied enzyme involved with sulfur cycling in soil. This is because organic sulfates are abundant in soil, and hence arylsulfatase may play an important role in the mineralization of organic soil sulfur. The enzyme may be extracellular or associated with cell debris, and its activity is easily determined based on colorimetric assays.

Factors Affecting the Mineralization of Sulfur in Soil

Mineralization is generally measured as net mineralization, either the amount of $\text{SO}_4^{2-}\text{-S}$ accumulated during the period of study or the difference between gross mineralization and assimilation. Thus, for higher net mineralization to occur, the mineralization—assimilation balance has to be driven toward mineralization. A break-even point for mineralization and immobilization can be calculated based on the carbon-to-sulfur ratio of the substrate, the decomposing organisms, and the yield coefficient. For example, if we consider the decomposition of crop residues, net mineralization will generally occur with a carbon-to-sulfur ratio of 200 or less, whereas net sulfur immobilization will occur when the ratio is greater than 400/1. Because microbial activity is the driving force for mineralization and immobilization, these processes are significantly influenced by all factors affecting microbial metabolism, such as:

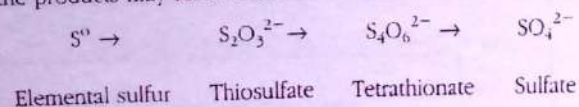
- energy and nutrient supply,
- carbon-to-sulfur ratio,
- abundance of organic sulfur,
- water availability,
- pH,
- temperature, and
- redox potential.

For example, actively growing plants may significantly increase sulfur mineralization in soils. Plants supply energy sources to the rhizosphere in the form of root exudates that increase microbial growth and activity, thus increasing sulfur mineralization. However, the re-assimilation of inorganic sulfates released by the growing microorganisms may result in no increase in the sulfur pool available to plants and may even result in reduction of that pool when the microbial demand exceeds the rate of sulfur mineralization. Different plants excrete different types and amounts of root exudates and require different amounts of sulfur for growth. Hence, crop rotations can have a significant impact on sulfur cycling in soil.

Microbial Oxidation of Inorganic Sulfur Compounds

Chemoautotrophic and Chemoheterotrophic Sulfur Oxidation

The abiotic oxidation of reduced sulfur compounds can occur to a limited extent in soils, but microbial reactions clearly dominate the process. For example, the biological oxidation of elemental sulfur in soils apparently takes place primarily via the following sequence of reactions (i.e., those most common to heterotrophs), although some of the products may result from abiotic side reactions:

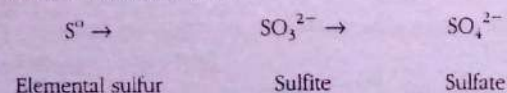


Many different microorganisms are important for the oxidation, reduction, and cycling of sulfur in soil and other ecosystems (Table 15-5). In the case of sulfur oxidation, the microorganisms can be divided into:

- chemoautotrophs (lithotrophs), including species of the genus *Thiobacillus*,
- photoautotrophs, including species of purple and green sulfur bacteria, and
- chemoheterotrophs (organotrophs), including a wide range of bacteria and fungi.

The chemoautotrophs and chemoheterotrophs are largely responsible for oxidizing sulfur in most aerobic, agricultural soils.

Many chemoautotrophic bacteria (e.g., thiobacilli) are capable of oxidizing reduced inorganic sulfur compounds. The biochemistry of sulfur oxidation by thiobacilli growing *in vitro* has been extensively reviewed (Postgate and Kelly, 1982; Pronk et al., 1990). For acidophilic thiobacilli, the most common sequence of reactions involved in sulfur oxidation is:



A great variety of thiobacilli can be isolated from natural habitats. They include obligate acidophilic chemoautotrophs, facultative chemoautotrophs (thiobacilli that grow autotrophically with reduced inorganic sulfur compounds as energy sources, but are also capable of heterotrophic growth), and **mixotrophs**, which can use mixtures of inorganic and organic compounds simultaneously. The thiobacilli differ in

Table 15-5 Sulfur-using bacteria occurring in soil and aquatic habitats.

Group	Sulfur conversion	Habitat requirements	Habitat example	Examples of Genera
Heterotrophs that use oxidized S species as electron acceptors	$SO_4^{2-} \rightarrow HS^-$ $S_2O_3^{2-} \rightarrow HS^-$ or S^0 $S^0 \rightarrow HS^-$ $SO_3^- \rightarrow HS^-$	anaerobic; organic substrates available; light not required	anoxic sediments and soils	<i>Desulfomonas</i> <i>Desulfotribrio</i> <i>Desulfotomaculum</i> <i>Desulfurmonas</i> <i>Campylobacter</i>
Obligate and facultative autotrophs that use reduced S as an energy source	$HS^- \rightarrow S^0$ $S^0 \rightarrow SO_4^{2-}$ $S_2O_3^{2-} \rightarrow SO_4^{2-}$	$H_2S - O_2$ interface; light not required	mud; hot springs; mine drainage; soils	<i>Thiobacillus</i> <i>Thiomicrospira</i> <i>Achromatum</i> <i>Beggiatoa</i>
Phototrophs that use reduced S as an electron donor	$HS^- \rightarrow S^0$ $S^0 \rightarrow SO_4^{2-}$	anoxic; H_2S ; light	shallow water; anoxic sediments; metalimnion or hypolimnion; anoxic water	<i>Cblorobium</i> <i>Chromatium</i> <i>Ectothiorhodospira</i> <i>Thiopelis</i> <i>Rhodospirillum rubrum</i>
Heterotrophs that use organic S compounds as energy sources or that hydrolyze esters	$org\ S \rightarrow HS^-$ $org\ S_1 \rightarrow volatile\ org\ S$ $estes\ SO_4 \rightarrow SO_4^{2-}$	source of organic S compounds	sediments; soils; water column	Many
Microorganisms that use SO_4^{2-} or H_2S in biosynthesis	$SO_4^{2-} \rightarrow protein$ $HS^- \rightarrow protein$ $SO_4^{2-} \rightarrow DMSP^*$	nonspecific	sediments; soils; water column	Many

From Cook and Kelly (1992). Used with permission.

*dimethylsulfoniopropionate

their physiological characteristics and in the reduced sulfur compounds used as energy sources (Table 15-6). The majority of these thiobacilli are obligate aerobes, although some, like *Thiobacillus denitrificans*, can grow anaerobically by using nitrate as a terminal electron acceptor. Other species of thiobacilli use electron donors such as ferrous iron (*T. ferrooxidans*) and thiosulfate (*T. thioparvus*) in addition to sulfur.

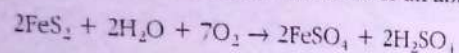
Although thiobacilli can oxidize sulfur to plant-available sulfate in some soils, this process is also evidently mediated by many different heterotrophic soil microorganisms. Bacteria, such as *Arthrobacter*, *Bacillus*, *Micrococcus*, and *Pseudomonas*, some actinomycetes, and a wide range of fungi are also capable of oxidizing elemental and reduced forms of sulfur. Many of these sulfur-oxidizing heterotrophs have been isolated from soil and may:

- oxidize sulfur, producing mainly thiosulfate,
- oxidize sulfur, producing sulfate, and
- oxidize thiosulfate to sulfate.

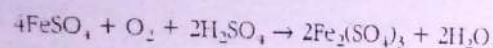
The pathway by which heterotrophic microorganisms produce these sulfur oxyanions has not been established, although several studies suggest that it is enzymatic in fungi. Apparently, no energy is derived by the organisms through these oxidations, and the transformations are incidental to the major metabolic pathways.

organic matter. The formation of metal sulfides during the mineralization of organic sulfur compounds is also possible, although little is known about this phenomenon.

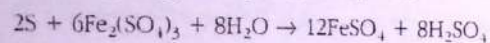
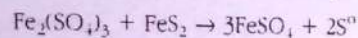
The oxidation of metal sulfides in soil involves both chemical and microbial processes and, as a result, is a more complex process than is sulfur oxidation. Chalcocite (Cu_2S), chalcopyrite (CuFeS_2), galena (PbS), pyrite (FeS_2), and nickel sulfide (NiS) are just a few examples of metal sulfides that are subject to microbial transformations. For example, the biological oxidation of pyrite follows a series of oxidation steps described in the following equations. These biotic oxidations are responsible for the formation of acid mine drainage and acid soil formation in surface mine spoils. First, ferrous sulfate is formed as the result of an abiotic oxidation step:



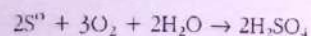
This reaction is then followed by the bacterial oxidation of ferrous sulfate, generally by *T. ferrooxidans*:



This reaction occurs chemically but can be accelerated 10^9 - to 10^8 -times by thiobacilli. Subsequently, ferric sulfate is reduced and pyrite oxidized by a strictly chemical reaction.



The elemental sulfur produced is finally oxidized by *T. thiooxidans* and *T. ferrooxidans*, and the acidity produced helps the whole process to continue.



Note the net production of 10 molecules of H_2SO_4 during the process.

Although several sulfur-oxidizing thiobacilli and heterotrophs can be isolated from acid sulfate soils in which pyrite is being oxidized, only *T. ferrooxidans* appears to play an important role in the process. The biological oxidation of sulfides and other reduced sulfur compounds can have severe consequences for the environment (Box 15-6). For example, acid mine drainage contaminates several thousand kilometers of streams in the Appalachian coal mining region of the United States.

Box 15-6

Problems Associated with Sulfur Oxidation. Oxidation of reduced sulfur compounds leads to the formation of acidic products. As a consequence, sulfur oxidation can have detrimental effects on the environment. This is especially true in the case of sulfide minerals such as pyrite (FeS_2). Some of the more serious problems associated with sulfur oxidation include:

- formation of acid mine drainage,
- formation of acid sulfate soils,
- corrosion of concrete structures, and
- corrosion of metals.

Microbial Reduction of Inorganic Sulfur Compounds

Bacterial Sulfate Reduction

The reduction of sulfate to hydrogen sulfide is mediated mainly by anaerobic, sulfate-reducing bacteria. This process may be significant in anaerobic, waterlogged soils, but is usually not important in well-aerated agricultural soils, except in anaerobic microsites. Nevertheless, sulfate reduction is a major component of the sulfur cycle in soils exposed to waterlogging or periodic flooding, especially where readily decomposable plant residues are present.

Microorganisms reduce oxidized sulfur compounds by either an assimilatory or dissimilatory process. Some use *assimilatory sulfate reduction* to meet their sulfur requirements. In *dissimilatory sulfate reduction*, bacteria use sulfate as a terminal electron acceptor, and large quantities of hydrogen sulfide (H_2S) are released. This process is analogous to the denitrification process discussed in Chapter 12. Like most denitrification, dissimilatory sulfate reduction is a strictly anaerobic process. In this case it is carried out by bacteria such as *Desulfovibrio* spp., *Desulfomonas* spp., and *Desulfotomaculum* spp. (Table 15-7). These bacteria use end products of other fermentations such as lactate, malate, and ethanol as electron donors.

Factors Influencing Sulfate Reduction

When a soil is flooded, electron acceptors become reduced in an ordered sequence: first oxygen, followed by nitrate, nitrite, manganic, and ferric compounds, and finally sulfate and carbon dioxide. Although the reduction of one compound does not have to be completed before another is reduced, oxygen and nitrate must be removed before the reduction of ferric and sulfate ions can occur. Because of this reaction sequence, sufficient ferrous ions generally are available to react with any hydrogen sulfide produced, and as a result, free hydrogen sulfide is rarely liberated from soils. Sulfate reduction increases with the period of soil submergence and following the addition of organic matter. Sufficient organic substrates to stimulate the process are also

Table 15-7 Dissimilatory sulfate-reducing bacteria.

Genera	<i>Desulfobacter</i> , <i>Desulfobulbus</i> , <i>Desulfococcus</i> , <i>Desulfonema</i> , <i>Desulfosarcina</i> , <i>Desulfotomaculum</i> , <i>Desulfovibrio</i>
General characteristics	Strict anaerobes Grow at mildly acid to mildly alkaline pH Generally mesophilic, but some species thermophilic
Substrates	Most sulfate reducers will also reduce sulfite and thiosulfate Some species reduce elemental sulfur Organic matter utilization varies with genus and species As a group, capable of completely oxidizing fatty acids from C1 to C18, lactate, pyruvate, low-molecular-weight alcohols, and some aromatic compounds
Habitats	Anaerobic sediments of freshwater, brackish water, and marine environments, thermal regions, water-logged soils, and animal intestines.

From Trudinger (1986). Used with permission.

liberated from seeds and from roots into the rhizosphere, with the result that in paddy soils, blackening caused by ferrous sulfide deposits often occurs in the root region. There is evidence, however, that rice roots can aerate the soil sufficiently that ferric iron is observed on the root surface. In general, the rate of sulfate reduction increases with decreasing redox potential, with the optimum being a function of soil pH, around -300 mV at pH 7. Sulfate-reducing bacteria are active in soil, sediments, polluted water, oil-bearing strata, and shales. Their activity may be beneficial or detrimental to the surrounding environment and have serious economic consequences (Box 15-7).

Box 15-7

Consequences of Sulfate Reduction. The activity of sulfate-reducing bacteria and the problems they pose can be seen in many examples from our daily lives:

- Sulfate-reducing bacteria are a major cause of corrosion of underground iron pipes, costing between \$1.6 billion and \$5.0 billion in the United States in 1990.
- Turf managers find that sulfate-reducing bacteria can produce a black layer under golf course greens by using the organic matter in root exudates to reduce soil sulfates to ferrous sulfides.
- The water in the canals of Venice is polluted with hydrogen sulfide and, as a consequence, the gondolas of Venice turn black regardless of their original color.

Volatilization of Inorganic and Organic Sulfur Compounds from Soil

A number of sulfur gases are released from soils, marshes, peats, and sediments or from anthropogenic sources. These gases may be inorganic or organic and play an important role in the cycling of sulfur through the atmosphere (Chapter 23). Many different fungi and heterotrophic bacteria are responsible for the formation of these volatile compounds during the metabolism of organic sulfur compounds (Table 15-8).

Table 15-8 Biochemical origin of volatile sulfides produced in soils by microbial degradation of organic matter under aerobic and anaerobic conditions.

Volatile Name	Formula	Biochemical precursors
Hydrogen sulfide	H ₂ S	Proteins, polypeptides, cystine, cysteine, glutathione
Methyl mercaptan	CH ₃ SH	Methionine, methionine sulphoxide, methionine sulphone, S-methylcysteine
Dimethyl sulfide	CH ₃ SCH ₃	Methionine, methionine sulphoxide, methionine sulphone, S-methylcysteine, homocysteine
Dimethyl disulfide	CH ₃ SSCH ₃	Methionine, methionine sulphoxide, methionine sulphone, S-methylcysteine
Carbon disulfide	CS ₂	Cysteine, cystine, homocysteine, lanthionine, djenkolic acid
Carbonyl sulfide	COS	Lanthionine, djenkolic acid

From Andreae and Jaeschke (1992). Used with permission.

Environmental Aspects of Sulfur Pollutants

Acid Sulfate Soils

Acid sulfate soils contain sulfides, mainly in the form of pyrites, which may be oxidized to yield free and adsorbed sulfates. They are characterized by yellow mottling due to the formation of jarosites [AFe₃(SO₄)₂(OH)₆, where A = K⁺, NH₄⁺, Na⁺ or H₃O⁺], and have a pH typically below 4. Although these soils cover large areas of the tropics, they tend to be of only local importance in temperate regions. The acidification of these soils results from the abiotic and microbial oxidation of pyrite. Problems in producing crops on these soils occur because of aluminum and manganese toxicity rather than to the direct effects of acidity. Acid sulfate soils can be reclaimed by:

- controlling the water table through adequate drainage,
- adding lime,
- planting crops tolerant of aluminum, manganese, and iron, and
- improving soil fertility generally.

Deposition of Atmospheric Sulfur in Soils

Soils subject to atmospheric pollution receive sulfur from the atmosphere largely in the form of dilute sulfuric acid. Thus, sulfate is the main sulfur ion entering soils from the atmosphere; smaller quantities of sulfite and bisulfite may also contaminate these soils. Atmospheric pollution deposits, consisting largely of soot, may also be locally important sources of reduced sulfur compounds, particularly in areas adjacent to industrial plants, such as coking and steel works. Because sulfate is the major sulfur ion entering soil from atmospheric pollution, we expect that the major sulfur transformations that occur involve sulfur assimilation and sulfur reduction rather than sulfur oxidation. However, when reduced sulfur compounds in the atmosphere are deposited on soils, they are rapidly oxidized.

Summary

Sulfur is an essential element for all living organisms and is the basis for primary productivity in some exotic communities. It exists in a number of oxidation states as inorganic and organic compounds that undergo a number of biotic and abiotic transformations. These transformations can be beneficial or detrimental to ecosystems depending on the forms and fluxes of sulfur. The cycling of sulfur through aquatic, terrestrial, and atmospheric ecosystems is similar to that of other elements, such as carbon and nitrogen, and is influenced by natural and anthropogenic processes.

We currently have a basic understanding of the forms and amounts of sulfur in terrestrial ecosystems and the processes controlling the supply of sulfur to plants. The exchange of sulfur gases between the soil-plant system and the atmosphere is less well documented. Understanding how key processes in the sulfur cycle respond to environmental factors (e.g., construction of models of mineralization and volatilization processes that include temperature, moisture, substrate, and microbial