Chapter 16

Biological Cycling of Inorganic Nutrients and Metals in Soils and Their Role in Soil Biogeochemistry

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Chapter Contents

I INTRODUCTION

Microorganisms play a major role in element cycling in terrestrial systems (Ehrlich and Newman, 2009; Gadd et al., 2012). Microbes inhabit a wide range of environmental niches in soils, populating oxygen gradients from fully aerobic in aerated surface soils to entirely anaerobic conditions within soil aggregates or in sediments and deep subsurfaces. They tolerate a broad range of moisture content, salinities, temperatures, and chemical compositions, providing 80-90% of the life support systems for many different types of soil (Silva et al., 2013). Most microbes inhabit soil surfaces, are able to access nutrients in the soil solution and in mineral and organic substrates, and are then able to interconvert them. They can also form multispecies biofilms on these surfaces in

Soil Microbiology, Ecology, and Biochemistry. http://dx.doi.org/10.1016/B978-0-12-415955-6.00016-5

which organisms with different capabilities can interact. The resulting redundancy of this microbial function helps ensure that nutrient transformations and soil fertility are resilient to environmental stresses (Griffiths and Philippot, 2013). The importance of soil microorganisms in C and N cycles has been explored in other chapters of this volume. This chapter will focus on the role of prokaryotes and fungi on cycles of other elements in soil systems.

II NUTRIENT NEEDS OF SOIL MICROORGANISMS

Because microbes provide the lowest trophic level in soil foodwebs, their elemental composition provides a clear indication of their production dynamics, and the biogeochemical cycles in which they are involved. Microbial cells are predominantly made up of C and N. They also show structural requirements for phosphorus (P) and sulfur (S), which are used for the production of nucleic acids/phospholipids and proteins. These elements are enriched in the microbial biomass above their content in the soil (Table 16.1). A recent work claimed that arsenic could replace P in a bacterium (Wolfe-Simon et al., 2011). However, this hypothesis was soon rejected as it was shown that the studied bacteria were arsenate resistant and could grow in the presence of very low available P concentrations (Erb et al., 2012).

The preferred forms of P and S for microbial growth are free inorganic phosphate and sulfate, respectively, although almost all of the P and S are sequestered either as organic compounds or are strongly sorbed on soil surfaces. The microbial preference for the simple anionic forms of these nutrients is clearly seen in the networks of "P-starvation-induced" and "sulfate-starvation induced" genes that control the uptake and assimilation of alternative P- and S-containing compounds (Hsieh and Wanner, 2010; Kertesz, 1999).

A compilation of studies that evaluate the organic P and S contents of nearly 1000 different soils from around the world revealed that: (1) C:N ratios varied between 9.8 and 17.5 (598 soils), (2) C:P ratios lay in the range 44-287 (408 soils), and (3) C:S ratios were between 54 and 132 (527 soils; Kirkby et al., 2011). In this chapter, the transformations of these elements have been subdivided into those carried out by prokaryotes (bacteria and archaea) and those carried out by fungi, in part because the ratios of these elements are fundamentally different for the various groups of organisms. Soil bacteria have a C:P ratio of about 59.5, whereas fungi contain much less P, with C:P in the range of 300-1190 (Kirchman, 2012). These differences reflect different biological roles for these organisms in the soil environment and are so pronounced that the C:N ratios have been successfully used to help predict the bacterial:fungal ratios in microbial communities of soils over a range of habitats and environments (Fierer et al., 2009).

Other elements are also essential for microbial growth, yet are required in smaller amounts than for major biogenic elements. The primary role they play is in biochemical processes, and they may be needed for functioning of specific enzymes or for redox processes. The most important of these elements is iron (Fe), which is abundant in the earth's crust and is required in small amounts in microbial cells, mainly in electron transfer reactions. Additional roles for other

TABLE 16.1 Elemental Composition of a Representative Soil Bacterium, *Pseudomonas putida* (Passman and Jones, 1985), of a Representative Soil Archaeon, *Methanosarcina barkeri* (Scherer et al., 1983), and of a Representative Soil Fungus, *Glomus intradices* (Olsson et al., 2008)

Element	*Pseudomonas putida* (% w/w)	*Methanosarcina barkeri* (% w/w)	*Glomus intradices* (Young Hyphae) (% w/w)	*Glomus intradices* (Spores) (% w/w)
C	51-53	37-44	nm	nm
N	41-42	9.5-12.8	nm	nm
P	1.3-2.2	0.5-2.8	0.24-0.96	0.13-0.8
S	0.5-0.54	0.56-1.2	0.26-0.92	0.008-0.02
Mg	0.26-0.53	0.09-0.53	nm	nm
Na	0.16-0.39	0.3-4	nm	nm
K	0.18-0.3	0.13-5	1.2-3.6	0.09-0.28
Fe	0.012-0.023	0.07-0.28	0.06-0.13	0.03
Ca	0.19-0.33	0.0085-0.055	0.57-2.2	0.23-0.6
Zn	0.006-0.013	0.005-0.063	0.07	0.008-0.04
Cu	0.002-0.003	0.001-0.016	nm	0.004
Mo	nm	0.001-0.007	nm	nm
Mn	nm	0.0005-0.0025	0.02	0.003-0.04

nm, not measured.

TABLE 16.2 Major and Trace Elements Used by Bacteria, Archaea, and Fungi with Examples of Their Roles

Element	Inorganic Form Taken Up	Examples of Element's Role in Cell Metabolism
Major elements		
C	HCO_3^-	All organic compounds (bacteria/archaea/fungi)
N	N_2, NO_3^-, NH_4^+	Proteins, nucleic acids (bacteria/archaea/fungi)
P	PO_4^{3-}	Nucleic acids, phospholipids (bacteria/archaea/fungi)
S	SO_4^{2-}	Proteins, coenzymes (bacteria/archaea/fungi)
K	K^+	Cofactor for enzymes (bacteria/archaea/fungi)
Ca	Ca^{2+}	Intracellular signaling (bacteria/archaea/fungi)
Si	$Si(OH)_4$	Diatom frustules
Trace elements		
Fe	Fe^{3+} and Fe^{2+} complexes	Electron transfer systems (bacteria/archaea/fungi)
Mn	Mn^{2+}, MnO_2	Superoxide dismutase (bacteria/archaea/fungi), oxygenic photosynthesis in cyanobacteria
Mg	Mg^{2+}	Chlorophyll (bacteria/archaea)
Ni	Ni^{2+}	Urease, hydrogenase (bacteria/archaea/fungi)
Zn	Zn^{2+}	Carbonic anhydrase, alkaline phosphatase, RNA/DNA polymerase (bacteria/archaea/fungi)
Cu	Cu^{2+}	Electron transfer system, superoxide dismutase (bacteria/archaea/fungi)
Co	Co^{2+}	Vitamin B_{12} (bacteria/archaea)/antimicrobial compounds (fungi)
Se	SeO_4^{2-}	Formate dehydrogenase, betaine reductase, sarcosine reductase (bacteria/archaea)
Mo	MoO_4^{2-}	Nitrogenases, sulfite oxidases, nitrite reductase (bacteria/archaea/fungi)
Cd	Cd^{2+}	Carbonic anhydrase in diatoms
W	WO_4^{2-}	Hyperthermophilic enzymes (bacteria/archaea)
V	VO_4^{3-}	Nitrogenases (bacteria/archaea/fungi)

essential elements include (1) copper in some redox enzymes, (2) zinc in alkaline phosphatase and carbonic anhydrase, (3) molybdenum in the enzyme that catalyzes N fixation in bacteria, and (4) nickel present in urease and in several enzymes that are important in anaerobic environments, including hydrogenase, methyl coenzyme M reductase, and C monoxide dehydrogenase. Other microbes require a range of even less common metals, including tungsten and vanadium for specific enzymes (Table 16.2).

Attempts have been limited for both defining "typical" elemental compositions of bacteria and archaea *in vitro* and then linking them to the compositions observed in natural environments. This is largely because the actual composition of cells *in situ* reflects both the biochemical requirements of the cell under the given conditions and the availability of particular nutrients in the soil environment. A corollary of this is that by measuring the ratios of C to N and P in soil microbial biomass using fumigation techniques, it should be possible to extract direct information about the nutrient limitations that microbes experience in soils (Cleveland and Liptzin, 2007). However, showing nutrient limitation for microorganisms would require, in addition to information on nutrient content and ratio, an indication of microbial activity (Ehlers et al., 2010). In principle, fumigation techniques could be extended to minor and trace elements, which to date has not yet been attempted. However, there are known sources of error for this technique, which limit its usefulness (Ross, 1990).

Several inorganic nutrients are used by a range of bacteria to provide energy for metabolism and growth. Different compounds of S (sulfate, sulfide, sulfite, and elemental sulfur) can be used by a range of organisms, either as electron donors or as terminal electron acceptors, in redox pathways that supply reducing equivalents to the cell. Metals such as Fe and Mn are used as energy sources by chemoautotrophic bacteria and also as terminal electron acceptors by a range of heterotrophic bacteria. The transformations of these metals between different redox states often have a significant effect on their solubility and hence on both their mobility within the soil environment and their bioavailability to microbes in the soil and to plants. Ferric (Fe^{3+}) compounds, for example, are much less soluble and bioavailable than ferrous (Fe^{2+}) compounds. Ferrous iron may also be said to be the preferred form of Fe for soil microbes (Cartron et al., 2006), despite the fact that Fe^{3+} predominates in all aerobic environments, and levels of Fe required for optimal microbial growth (10^{-7} to 10^{-5} molar, Loper and Buyer, 1991) are rarely present. Many microbes use extracellular reductases to convert Fe^{3+} to Fe^{2+} for uptake. In addition, Fe^{2+} is the active form controlling iron metabolism within the cell.

III EFFECT OF MICROORGANISMS ON ELEMENT CYCLES

A Phosphorus Cycle

Phosphorus ultimately derives from phosphate-containing minerals in the bedrock, such as apatite, which are progressively released into the soil by chemical

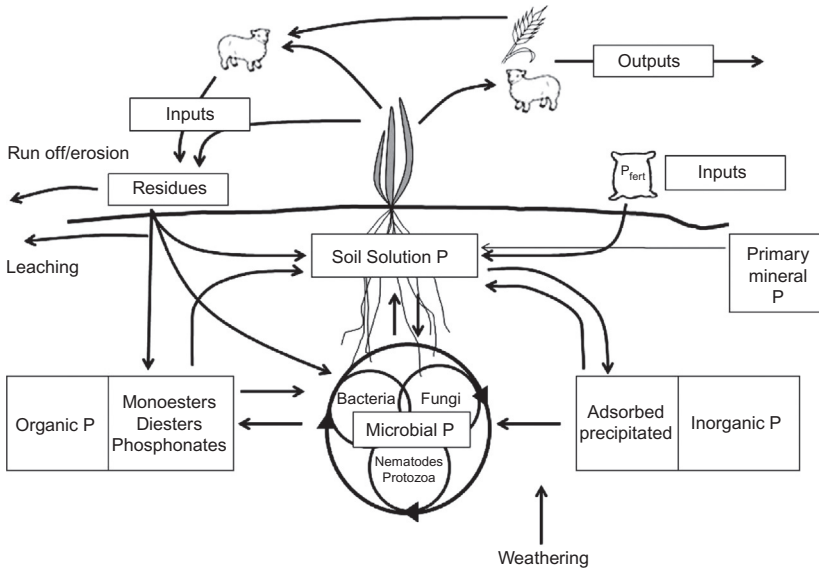

FIG. 16.1 Major components of the phosphorus cycle in agricultural systems. Transformations between major P components are indicated. *Adapted from Richardson et al. (2009).*

and biological weathering. The levels of P found in soils vary considerably. Young soils on bedrocks containing high levels of apatite are often quite rich in total P, whereas many highly weathered tropical soils are low in total P and are particularly deficient in soluble P. The total P content of agricultural soils ranges from 150 to 2000 μg P g^{-1}. Chemical weathering releases inorganic phosphate (Pi) into the soil solution (Fig. 16.1), but the levels of soluble orthophosphate present are very low in most soils (often below 0.1 mg P L^{-1} and in highly weathered soils below 0.01 mg P L^{-1}, Randriamanantsoa et al., 2013) because it sorbs to soil surfaces and forms precipitates with Ca salts in alkaline soils. These processes also affect phosphate applied to the soil as chemical fertilizer, with generally only 10-30% of applied fertilizer P taken up by crop plants in the year following application (Doolette and Smernik, 2011), and the remainder transferred to less soluble pools.

Soils also contain a large pool of organic forms of P. These are partly derived from biological compounds released into the soil as animal waste, plant litter, and microbes. The exact nature of organic P compounds is difficult to assess, but they have been classified (in decreasing order of abundance) as: (1) monoester phosphates, such as phytate (myo-inositol-hexakisphosphate); (2) diester phosphates, such as nucleic acids; and (3) phosphonates containing a direct C-P bond, probably derived from phosphonolipids, which replace phospholipids in some microbes (White and Metcalf, 2007). In marine systems, methylphosphonate is synthesized by the Thaumarchaeota (Metcalf et al., 2012), a major group in soils for which the functions are not yet well defined (Pester et al., 2011).

Phosphorus speciation has largely been conducted in recent years by two spectroscopic methods, nuclear magnetic resonance (NMR) and X-ray absorption near edge spectroscopy (XANES). Each of these characterizes the chemical environment surrounding the P nucleus to provide information about the nature of the P atom. NMR may be performed either on solid samples or soil extracts and is particularly useful for differentiating organic P forms, whereas XANES is usually done with solid samples, providing useful information on inorganic P forms (Kizewski et al., 2011).

Phosphorus availability is mediated by mineralization and immobilization from organic fractions, whereas sorption/desorption and precipitation/solubilization processes are mediated from the inorganic fractions (Frossard et al., 2000). According to McGill and Cole (1981), organic P can be mineralized either biologically, when microbes mineralize organic compounds in search of C and thereby release P associated with C, or biochemically, when microbes specifically scavenge P through the release of phosphatase enzymes. Work by Bünemann et al. (2012) suggests that the release of P through enzymatic hydrolysis constitutes a major part of net organic P mineralization in a pasture soil.

Phosphatase enzymes catalyze the hydrolysis of the phosphate ester bonds to release inorganic phosphate. These phosphatases are specific to particular forms of phosphate esters, many of which may be specific to certain compounds. Thus, phosphomonoesterases cleave phosphate from monoester forms, such as phospholipids or nucleotides. Phosphodiesterases release phosphate from diester forms, such as nucleic acids, and phytases release phosphate from inositol phosphates (Keller et al., 2012). To mobilize the amounts of phosphate needed for cell growth, most of these enzymes are synthesized by the microbial cell with a signal sequence that targets them for extracellular localization. Inorganic phosphate is therefore released outside the cell and taken up by specific high-affinity phosphate transporters in the cell membrane. This allows the cell to scavenge phosphate from a broad range of phosphate-containing substrates without the need to synthesize specific transporters for multiple different compounds, many of which are of high molecular weight. By contrast, phosphonates are almost certainly degraded within the bacterial cell, as the enzymes responsible, the C-P lyases, are unstable and require several cofactors (Zhang and van der Donk, 2012). Because the cells also require other nutrients for growth, the rate of incorporation of the released phosphate into microbial biomass depends on the availability of C and N in the soil. Generally, if the C:P ratio is $>\sim$300:1, net immobilization of P into microbial biomass occurs, whereas for C:P ratios $<\sim$200, microbial growth yields a surplus of P, and net mobilization of orthophosphate into the soil solution is observed.

Although the process described immobilizes P into the soil microbial biomass, this P is also readily remineralized. Bacterial cell turnover in the soil can be rapid, especially in the regions surrounding plant roots (the rhizosphere), where there is organic C for growth. Microbial cells die both through predation by protists and by viral attack, releasing their cell contents into the soil solution. In addition, the

processes of soil drying and rewetting cause changes in cell turgor, which can both release cell contents during the drying process and cause cell lysis through osmotic shock on rewetting (Bünemann et al., 2013). This leads to the release of a significant proportion of the P from microbial biomass during events such as rainfall after a dry period (Blackwell et al., 2010). Similar effects are also caused by cycles of freezing and thawing, although the quantity and form of biomass P that is released will change over time as these environmental stresses lead to selective changes in the microbial community (Blackwell et al., 2010).

Bacteria play an important role in both the solubilization of phosphate from the precipitated inorganic fraction of soil P and in weathering of minerals to release P (Uroz et al., 2009). This process is mediated by the release of organic acids from the cell, which causes a localized decrease in the soil pH. Acids that are released in this way include gluconic, oxalic, malonic, succinic, lactic, isovaleric, isobutyric, and acetic acids (Rodriguez and Fraga, 1999). The "phosphate-solubilizing bacteria" (PSB) that mediate this process include many strains of *Pseudomonas*, *Bacillus*, and *Rhizobium* (Rodriguez and Fraga, 1999), the best-characterized of which is *Bacillus megaterium*, which has been commercially applied in biofertilizers. The PSBs are readily isolated from the soil by screening for the ability to solubilize Ca or Fe phosphates in an agar-plate test. Specific genes have also been linked with their ability to solubilize bound phosphate (Gyaneshwar et al., 2002). These genes are also found in many bacteria that do not catalyze the phosphate solubilization process *in vitro*, and their presence is therefore not a reliable diagnostic test for phosphate solubilization ability. In addition, supplementation of soils with PSBs does not reliably increase soluble soil P or increase plant P-uptake (Gyaneshwar et al., 2002), so it is not yet clear which bacteria are most important for this process *in situ*.

All fungi take up P as orthophosphate ions using both low- and high-affinity transporters (Plassard and Dell, 2010; Plett and Martin, 2011). Glomeromycota take up mostly orthophosphate ions from the soil solution and have limited direct effect on soil P solubility (Smith and Smith, 2011). The presence of P-solubilizing bacteria around the hyphae can allow the uptake of P that was not originally available, although the importance of this remains to be clarified. Ectomycorrhizal fungi and other saprotrophs can release phosphatase enzymes able to cleave Pi from organic P sources. For instance, some fungi (e.g., *Aspergillus fumigatus*) release high quantities of phytase that cleave Pi from phytate, which can be present in high concentration in soils (Jennings, 1995; Plassard et al., 2011). Fungi can also release low-molecular-weight organic acids, such as oxalate, which help the release of P from inorganic forms as the result of acidification and chelation of the cations bonding P by the organic acids (Plassard et al., 2011). Smits et al. (2012) demonstrated that *Paxillus involutus* in symbiosis with *Pinus sylvestris,* growing under low available P conditions, was able to deliver plant C to apatite grains and to accelerate the rate of P release from these grains. Similarly, *Aspergillus niger*, which is known to produce large amounts of citric acid, can dissolve large amounts of apatite (Bojinova et al., 2008). Finally, siderophores produced by

fungi for Fe acquisition can also significantly increase phosphate solubility (Reid et al., 1985), probably by displacing phosphate associated with Fe. Phosphate taken up in excess to growth is stored in the fungal vacuole as polyphosphates. Compared to plants and bacteria, fungi contain more polyphosphate and less diester P (Bünemann et al., 2011). In a recent review, Richardson et al. (2009) suggest that fungi (Glomeromycota, but also *Penicillium* spp. or *Aspergillus* spp.) could be used to improve the use efficiency of soil P for agricultural plants, yet their successful application in the field on a large scale remains to be demonstrated.

B Sulfur Cycle

Most global reserves of S are tied up in the lithosphere, from which they are slowly released by weathering processes. Weathering provides sulfate input to the oceans (the inorganic sulfate concentration in seawater is *ca* 27 mM) and to soils. Agricultural soils contain 2-2000 μg S g^{-1}, but in contrast to marine environments, inorganic sulfate usually makes up only a small proportion of the total S, with organic forms making up >90% of total soil S. Uncultivated soils, such as forest and grassland, contain different proportions of organic S compounds, but inorganic sulfate content is similar to that of farmed soils (Autry and Fitzgerald, 1990; Chen et al., 2001). Unlike phosphate, inorganic sulfate is relatively soluble at soil pH values, and ground water can contain considerable sulfate concentrations (Miao et al., 2012). Available levels of inorganic sulfate in soil therefore vary considerably throughout the year with changes in the water table.

The bulk of the S bound to soil organic matter (SOM) is found in the high-molecular-weight fraction (MW > 100,000 kDa; Eriksen, 2009). Much of this is physically protected within soil microaggregates and therefore not readily mobilizable in the short term (Eriksen et al., 1995). The precise chemical structure of the larger S-containing molecules in SOM is not known, but the chemical environment of the S atoms has been defined in a number of ways. Historically, organic S has been classified according to its chemical reactivity with reducing agents (Freney et al., 1975). Two major groups of organic S compounds can be distinguished: those that yield H_2S on reduction with hydriodic acid predominantly contain sulfate ester-S bonds (C—O—S), whereas those that are unreactive to this treatment contain a direct C—S linkage. Further treatment of the nonextracted soil S with Raney-Ni releases H_2S from amino acid S (cysteine/methionine/peptides), and the residual, unreduced S is considered to be predominantly sulfonate-S, containing a —SO_3H moiety (Autry and Fitzgerald, 1990). More recently, S speciation within SOM has been defined in terms of the S oxidation state, using X-ray spectroscopy (K-edge XANES). This can be done by using either bulk soils directly (Prietzel et al., 2011) with humic extracts of the soils (Zhao et al., 2006) or with organic S compounds extracted from soil with acetylacetone (Boye et al., 2011). This provides specific information on S functional groups within the sample, distinguishing

between reduced S, oxidized S (sulfonates and sulfate esters), and S with intermediate redox state, such as sulfones and sulfoxides (Prietzel et al., 2011).

Inorganic sulfate makes up <5% of the S in most aerobic soils, with the organic fraction comprised of sulfate esters (30-75% of total S) and sulfonates (20-50% of total S). There is considerable variation in the organic S composition between different types of soil, but on the whole, pastures and grasslands tend to be higher in sulfate esters, and forest soils contain more of the sulfonated fraction. Organic S in the soil is derived from plant litter and animal waste inputs, such as: (1) sheep urine containing 30% of its S as sulfate esters, (2) sheep dung containing about 80% of its S as carbon-bound-S (Williams and Haynes, 1993), and (3) 60-90% of the S in decaying plant material is C-bound S (Zhao et al., 1996), much of it derived from sulfolipid, a common lipid in the thylakoid membranes of the plant chloroplast (Benning, 1998). Carbon-bound S also enters soil as methanesulfonate during rainfall, derived in part from atmospheric dimethylsulfide (Kelly and Murrell, 1999).

Sulfur-containing compounds in aerobic soils undergo a range of transformations as part of the soil S cycle, including immobilization of inorganic sulfate and organic S compounds into microbial biomass and SOM and mineralization of soil organic S by means of sulfatase and sulfonatase enzymes in the soil (Fig. 16.2).

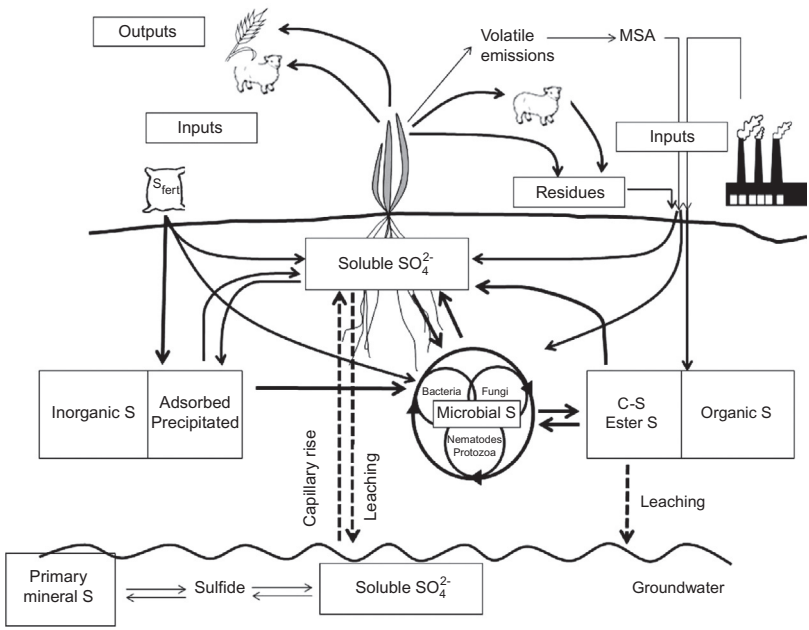

FIG. 16.2 Schematic diagram of the sulfur cycle in agricultural systems. Note that the S cycle is remarkably similar to the P cycle, but with the key differences of enhanced sulfate exchange with groundwater and the redox transformations of S compounds.

Immobilization of sulfate into organic matter has been studied primarily by experiments with ^{35}S-sulfate, which is initially incorporated into the sulfate ester pool and then slowly transformed into C-bonded S (Ghani et al., 1993b). This immobilization of sulfate is microbially mediated because it is stimulated by preincubation under moist conditions to stimulate bacterial growth (Ghani et al., 1993b) and also by addition of C or N in the form of glucose, organic acids, or model root exudates (sugars, organic, and amino acids; Dedourge et al., 2004; Vong et al., 2003). Incorporation of sulfate into the organic S pool is also dramatically increased by the addition of cellulose as a C source (Eriksen, 1997).

Early studies on mineralization of sulfate from the organic S fraction (Freney et al., 1975) suggested that sulfate ester S and C-bonded S contributed about equally to S mineralization. However, other chemical-speciation studies (Ghani et al., 1992, 1993a) showed that almost all the S released in short-term incubation studies was derived from C-bonded S. This agrees with spectroscopic (XANES) data, which show a good correlation between S mineralization and the amount of peptide-S and sulfonate-S in the soil humic fraction and a less clear relationship with sulfate ester-S (Zhao et al., 2006). Nonetheless, soil enzymes, such as sulfatases, are thought to be important in soil S transformations. These enzymes catalyze the hydrolysis of sulfate esters and are common and easily measured in soils. Arylsulfatase activity has been widely used as a measure of soil health and soil microbial activity (Taylor et al., 2002). Until recently, arylsulfatases have been thought to be largely extracellular enzymes (Gianfreda and Ruggiero, 2006). However, the arylsulfatase of one common soil genus, *Streptomyces,* has now been shown to be located in the cell membrane (Cregut et al., 2012), and the arylsulfatase genes in several *Pseudomonas* species do not appear to encode a signal peptide for extracellular secretion (Kertesz et al., 2007). Arylsulfatase activity is correlated with soil microbial biomass and the rate of S immobilization (Vong et al., 2003), along with other factors that affect microbial activity, such as pH and organic C levels (Goux et al., 2012). The most common cultivable, sulfatase-producing bacteria in agricultural soils belong to the *Actinobacteria* and *Pseudomonas* clades (Cregut et al., 2009).

Sulfonate-S comprises up to 80% of total organic S in some soils, with a large proportion of soil and water bacteria able to mineralize low-molecular-weight sulfonates (King and Quinn, 1997). The desulfurization reaction is catalyzed by a family of flavin-dependent monooxygenase enzymes, which cleave the sulfonate moiety to yield sulfite, which can then be assimilated into bacterial biomass. The key groups of bacteria responsible for this process in agricultural and grassland soils are members of the *Variovorax* and *Polaromonas* genera and *Rhodococcus* species, which have been studied using molecular methods to assess *in situ* diversity of the sulfonatase genes (Schmalenberger et al., 2008, 2009, 2010). The sulfonatase enzymes are thought to be entirely intracellular in location due to the mechanistic requirement for flavin and nucleotide cofactors. It is not clear how high-molecular-weight humic substances carrying sulfonate moieties can enter the cell for mineralization to occur.

Unlike P, S is subject to a range of different oxidation and reduction transformations in soil environments, almost all of which are mediated by bacteria. The main microbially catalyzed oxidation sequence for inorganic S compounds involves the sequential conversion of sulfide to sulfate as follows:

$$\underset{\text{Sulfide}}{S^{2-}} \rightarrow \underset{\text{Sulfur}}{S^{0}} \rightarrow \underset{\text{Thiosulfate}}{S_2O_3^{2-}} \rightarrow \underset{\text{Tetrathionate}}{S_4O_6^{2-}} \rightarrow \underset{\text{Sulfate}}{SO_4^{2-}}$$

Oxidation of these S-containing compounds is catalyzed by two main groups of microbes. Chemoautrophic bacteria use the S-substrates as a source of energy, while using inorganic C sources (lithotrophs). These are primarily bacteria of the genera *Thiobacillus* and *Acidithiobacillus*, most of which are obligate aerobes and use oxygen as a terminal electron acceptor for growth. However, *Thiobacillus denitrificans* is also able to grow anaerobically using nitrate as an electron acceptor. In addition, some of the thiobacilli are able to oxidize inorganic S compounds while growing either heterotrophically or autotrophically (facultative heterotrophy), or with a mixture of organic and inorganic C sources (mixotrophy). Chemoautrophic growth with inorganic S compounds is best characterized for organisms isolated from environments with low organic C (organic C $<<<$ inorganic S), such as hot spring sediments; many of which are thermophiles. However, the levels of organic C in soil are usually much higher (organic C $>>>$inorganic S), with bacterial populations being dominated by heterotrophs. A wide range of heterotrophic bacteria are also able to oxidize inorganic S compounds, including *Arthrobacter*, *Bacillus*, *Micrococcus*, and *Pseudomonas* species, many of which only carry out a partial oxidation (e.g., oxidize S to thiosulfate, or thiosulfate to sulfate, rather than the complete oxidation of S to sulfate). The role of these organisms is unclear because they do not appear to gain any energy from the oxidation process, with the possibility that the transformation is entirely cometabolic. On addition of elemental S to agricultural soils, populations of both thiobacilli and heterotrophic S-oxidizers are strongly stimulated, but the increase in thiobacilli population is only transient, suggesting that heterotrophic S-oxidation is more important in these environments (Yang et al., 2010).

Whereas microbial oxidation of inorganic S compounds almost always requires oxygen, the dissimilatory reduction of these compounds only takes place under anaerobic conditions. It is therefore not usually an important process in well-aerated soils (except in the interior of soil aggregates), but becomes very significant in sediments and when soils are flooded, especially when large amounts of organic C are present in the form of, for example, plant residues. This converts sulfate to sulfide and is catalyzed by the anaerobic sulfate-reducing bacteria, which are organotrophic organisms that use other low-molecular-weight organic compounds (e.g., propionate, butyrate, or lactate) or H_2 as an electron donor and sulfate as terminal acceptor. Marine sediments are particularly high in sulfate-reducing bacteria due to the high levels of inorganic sulfate present in seawater. Dissimilatory sulfate reduction

is known for five major groups of bacteria and two archaeal groups (Muyzer and Stams, 2008), many of which are also able to reduce sulfite and thiosulfate. Among the bacteria, common sulfate-reducing genera include *Desulfovibrio*, *Desulfobacter*, *Desulfococcus*, and *Desulfotomaculum*, whereas the best-known sulfate-reducing archaeal genus is *Archaeoglobus*. The key genes involved in the dissimilatory sulfate reduction process are *dsrAB*, which encodes the dissimilatory sulfite reductase, and *aprA*, which encodes adenosine phosphosulfate reductase. Sulfur-reducing populations have been identified in hydrothermal vents, mud volcanoes, acid mine drainage at high pH values, soda lakes, oilfields, agricultural soils, plant rhizospheres, and waste water treatment plants (Muyzer and Stams, 2008). Because the *aprA* gene is an essential part of the S oxidation mechanism, molecular analysis of *aprA* diversity yields detailed information on the activity of both S oxidizing and reducing pathways in any given environment, without the need to cultivate the bacteria in the laboratory (Meyer and Kuever, 2007). The ability of sulfate-reducing bacteria (SRB) to adapt to a wide range of anaerobic environmental conditions has been investigated further using systems biology approaches and appears to be largely due to flexibility in their energy metabolism and their readiness to form syntrophic associations with other microbes, especially H_2-producers (Zhou et al., 2011).

There is little information on S in soil fungi. Studies carried out on *Neurospora crassa*, *Aspergillus*, and *Penicillium* spp. show the existence of two types of sulfate transporters for intracellular uptake (Jennings, 1995; Marzluf, 1997). When sulfate availability decreases, fungi can synthesize arylsulfatase, methionine permease, extracellular protease, and more enzymes of the two transport systems (Jennings, 1995). After uptake, sulfate is reduced stepwise to 3′-phosphoadenosine 5′-phosphosulfate (PAPS), thiosulfonate, and sulfide, from which it is incorporated into cysteine and homocysteine. Some fungi (*Puccinia* spp.) are not able to use sulfate and require the addition of methionine for growth. Some of the aforementioned reduction steps are probably blocked in these fungi. When sulfate is taken up in excess, it can be stored as sulfate, choline sulfate, or glutathione in the vacuole. Fungi can also oxidize inorganic S, but it is not known whether they gain sulfate or energy from oxidizing inorganic S (Jennings, 1995).

C Iron Cycle

Iron is the fourth most abundant element in the earth's crust. However, it is poorly available to microorganisms because it is either bound up in primary minerals (either as Fe(II) or Fe(III)) or is in a sparingly soluble form in Fe oxides and oxyhydroxides, which may be strongly sorbed to clays and organic compounds. In aerobic soils, these oxides form Fe(III), and their solubility is governed by equilibria related to $Fe(OH)_3 \rightleftarrows Fe^{3+} + 3OH^-$ and is therefore strongly dependent on the soil pH. At acidic soil pH values (~3.5), the concentration of Fe^{3+} generated by this equilibrium is about 10^{-9} M, whereas at higher pH values (~8.5) this decreases to $\sim 10^{-24}$ M, well below the levels required for

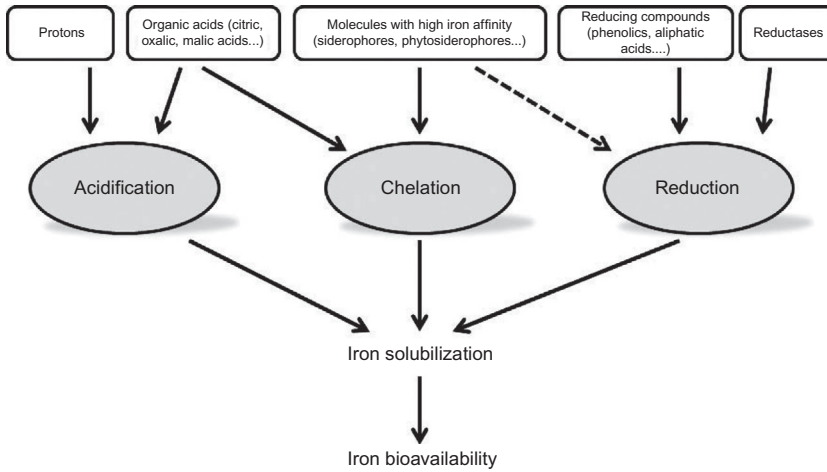

FIG. 16.3 Mechanisms affecting iron availability in soil and rhizosphere. Robin et al. (2008).

bacterial growth (Robin et al., 2008). Soil bacteria and fungi have developed several different mechanisms to increase Fe solubilization in the soil and to enhance the levels of bioavailable Fe, based on acidification, chelation, and reduction (Fig. 16.3, from Robin et al., 2008).

In the rhizosphere, microbial strategies are often assisted by corresponding mechanisms in plants, and there is some evidence for synergism in Fe solubilization, rather than competition for a limiting resource (Robin et al., 2008). Indeed, for aerobic soils, most studies of Fe and metal metabolism by bacteria have focused on the influence of soil bacteria in promoting uptake of Fe by plants for plant health and nutrition (Robin et al., 2008) or in facilitating the sequestration of heavy metals by plants in phytoremediation applications (Glick, 2010).

The most common mechanism used by prokaryotes to enhance Fe solubility is the production of low-molecular-weight Fe-chelating compounds known as siderophores, which are released into the soil solution. These are diverse molecules with affinities for ferric iron of up to 10^{52} (Albrecht-Gary and Crumbliss, 1998) that are able to sequester ferric iron from the soil environment, even when the metal is tightly sorbed onto solid surfaces or metal oxides (Kraemer, 2004). The nature of the chelating group varies between different siderophores and is classified according to the nature of functional ligands as catecholates, hydroxamates, hydroxypyridonates, and hydroxyl- or amino-carboxylates (Robin et al., 2008). A great deal is known about the siderophores of *Pseudomonas* species, called pyoverdins, much of which can be generalized to other aerobic Gr^- bacteria (Cornelis, 2010). These are polyketide and peptide-based molecules, whose synthesis is strongly upregulated by low iron availability and repressed under conditions in which soluble iron is plentiful. Iron is chelated to the siderophore via hydroxy and keto groups, and the Fe-siderophore complex then binds to a siderophore-specific outer membrane receptor on the bacterium. In

pseudomonads and other Gr^- bacteria, this leads to transport of the complex across the outer cell membrane. The metal is released from the siderophore by reduction to Fe^{2+} and then enters the cell via a specific inner-membrane transporter. The Fe-free siderophore molecule (or *apo-siderophore*) is released back into the extracellular environment using a specific exporter (Cornelis, 2010). In Gr^+ bacteria soil bacteria such as *Streptomyces*, the mechanism of Fe uptake is similar, except that the Fe-siderophore complex is reduced directly on binding to the cell. Plant roots also release Fe-binding molecules, called phytosiderophores, which are structurally distinct from bacterial siderophores (Robin et al., 2008) and have a lower affinity for Fe. Many bacteria are also able to obtain iron from phytosiderophores or by "siderophore piracy" from siderophore complexes released by other bacteria (Barona-Gomez et al., 2006; Cornelis, 2010; Traxler et al., 2012). This enhances competition for Fe in the soil environment, and because bacterial siderophores have considerably higher affinity for Fe than do the siderophores released by many fungi, siderophore-synthesizing bacteria are often selected as biocontrol agents against fungal pathogens in agricultural applications (Laslo et al., 2012).

Acidification of the soil increases the solubility of ferric salts and their bioavailability. Plant roots have developed an active strategy of releasing protons to increase the mobilization of Fe. The resulting pH changes have a considerable effect on Fe solubilization in the rhizosphere (Hinsinger et al., 2003). Both microbes and roots release organic acids in considerable quantities, which contribute to decreases in soil pH and also directly help in the mobilization of Fe because carboxylic acids (e.g., citrate and malate) act as lower-affinity ligands for Fe. A large proportion of organic acids in soil solution are complexed to metals, including Fe.

Oxidation and reduction reactions of iron also play a key part in cycling of Fe in soils and sediments. Fe^{2+} is rapidly chemically oxidized to Fe^{3+} at neutral or alkaline pH. For many years, abiotic reactions were thought to dominate Fe redox reactions. More recently, it has become clear that in many environments, microbial metabolism is extremely important in Fe redox transformations (Fig. 16.4; Weber et al., 2006). At acidic pH values, Fe^{2+} is oxidized to Fe^{3+} by chemoautrophic bacteria such as *Acidithiobacillus ferrooxidans*. At neutral pH values, Fe(II) functions as an electron donor for a wide range of lithotrophic Fe-oxidizing organisms, which belong to a diverse range of bacterial and archaeal phyla, the best-characterized of which are *Gallionella* and *Leptothrix* species (Emerson et al., 2010; Hedrich et al., 2011). Fe-oxidizing bacteria typically inhabit environments at redox boundaries, including wetlands, stream sediments, and waterlogged roots, where their activity is revealed by the presence of red Fe oxide precipitates (Emerson et al., 2010).

Insoluble Fe compounds may also be mobilized by dissimilatory Fe(III) reduction, in which Fe(III) is used as a terminal electron acceptor for microbial growth. Given the high levels of Fe(III) in the environment, this reaction is of great importance both for cell bioenergetics in the soil and for cycling of Fe

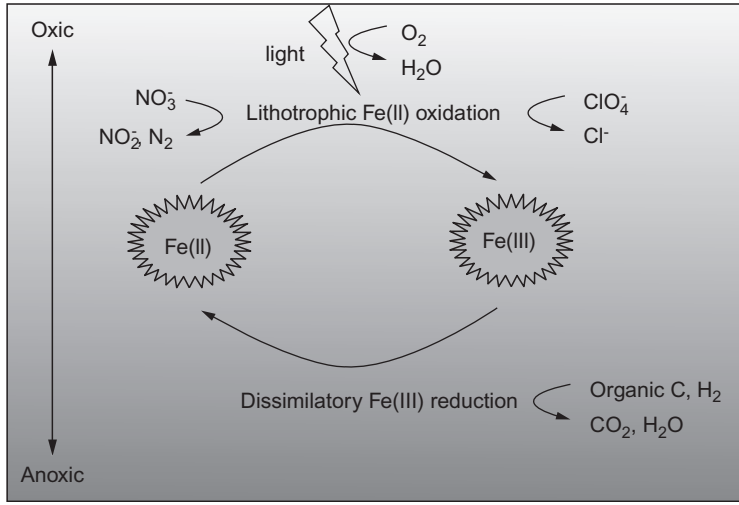

FIG. 16.4 The microbially mediated iron redox cycle. Reduction of Fe(III) to Fe(II) occurs only under anoxic conditions. Lithotrophic Fe oxidation is carried out by a range of organisms under different environmental conditions. Weber et al. (2006).

between different redox states. The best studied organisms that carry out this process belong to the *Geobacter* and *Shewanella* genera, although Fe reducers are also known from many other bacterial and archaeal phyla (Weber et al., 2006). These microbes face a difficult problem in transferring reducing equivalents to an insoluble ferric mineral or salt. *Shewanella* species have solved this problem by using soluble quinones or flavins as electron shuttles between the cell and the substrate, which may be some distance from the cell itself (Roden, 2012). By contrast, *Geobacter* species require direct contact between the cell and the substratum, allowing electrons to be passed from a bacterial biofilm directly to the metal. However, this contact may also be mediated by pili or microbial nanowires, which give metallic-like conductivity and facilitate electron transfer to the solid mineral phase (Lovley, 2012).

Iron is also needed in small amounts by fungi. It is essential as an acceptor and donor of electrons (e.g., in the cytochrome system). Fungi can also acquire Fe by releasing siderophores or by acidifying their environment (Philpott, 2006). The siderophores can be of either the hydroxamate type or the polycarboxylate type. Three families of hydroxamate siderophores have been identified thus far as ferrichromes, fusarinines, and coprogens (Gadd et al., 2012). The fungus can take up the Fe^{3+}-siderophore as a complex, or it can reduce Fe^{3+} to Fe^{2+}, which is then taken up by low- or high-affinity Fe^{2+} transporters (Philpott, 2006). Some of these transporters need Cu to be functional. Some Fe^{2+} transporters can also transport Mn^{2+} and Cu^{2+} in the cell (Bolchi et al., 2011; Philpott, 2006).

D Cycles of Other Elements

Many other elements are subject to microbial transformations in soils and sediments. Microbes have high potassium requirements, whereas assimilation of elements like Ca, Zn, Cr, Se, or Mn is required by microbes in small amounts for specific biochemical functions (Table 16.2). In some cases the uptake of higher concentrations of these elements may be toxic to the cell. Microbes have developed highly regulated mechanisms to ensure that only appropriate amounts of the element are taken up, and that only the correct metal is incorporated into the relevant cofactor or enzyme.

Bacteria and archaea use a range of mechanisms for this purpose, including metal-binding proteins (metallochaperones) that selectively bind the metal ions on entry into the cell. Systems that interact effectively with the target enzyme assemble proteins to insert the metal into its required site (Waldron and Robinson, 2009). The desired metal selectivity is achieved by variation in the number and type of metal ligands on the binding protein, by binding the metal in a cellular compartment that selects for specific redox qualities (the cytoplasm is a much more reducing environment than the periplasm) and by selective protein folding that depends on the type of metal ion bound (Waldron and Robinson, 2009). Comparative genomic studies of soil bacteria and archaea have revealed that these mechanisms are largely conserved (Zhang and Gladyshev, 2009).

In many soils, trace elements (e.g., Cu, Ni, Zn, or Mn) are present at much higher levels than soil microbes can assimilate, either because the soils are derived from weathering of rocks containing these elements or because of industrial contamination with metal-containing effluents or wastes. Most of these elements exist in a number of different oxidation states, and bacteria are involved in conversion between these redox forms. Because the solubility of metal salts often varies considerably between different oxidation states, microbial conversion of a contaminant metal compound to an insoluble form by oxidation or reduction may provide an effective method to immobilize it for bioremediation purposes (Kidd et al., 2009). Conversely, solubilization of insoluble metals by redox transformation may also be used for "biomining" to mobilize valuable metals from low-grade ores (Sorokin, 2003). Examples of organisms involved in the oxidation or reduction of a range of soil elements are given in Table 16.3.

Manganese redox cycling is an extremely dynamic process in soil environments. Manganese-oxidizing bacteria can be isolated from almost any soil or sediment sample, and the Mn oxides they generate are among the most reactive natural oxidizing agents, reacting rapidly with other reduced substrates, including Fe, S, and C compounds. Microbial Mn^{2+} oxidation occurs above pH 5, increases up to pH 8, and is catalyzed by a multicopper oxidase system. It does not appear to provide energy for the bacteria (no Mn^{2+}-dependent chemoautotrophs are known), but it may provide protection against damage by reactive oxygen intermediates, such as hydrogen peroxide or superperoxide (Tebo et al., 2005). In many soils that

TABLE 16.3 Examples of Bacteria Involved in the Oxidation or Reduction of Manganese, Chromium, Arsenic, Mercury, and Selenium

Element	Redox States	Reaction Type	Examples of Bacterial Genera Involved
Manganese (Mn)	Mn^{4+}, Mn^{2+}	Oxidation	*Arthrobacter, Bacillus, Pseudomonas, Leptothrix*
		Reduction	*Bacillus, Geobacter, Pseudomonas*
Chromium (Cr)	Cr^{6+}, Cr^{3+}	Reduction	*Aeromonas, Shewanella, Pseudomonas*
Arsenic (As)	As^{5+}, As^{3+}	Oxidation	*Alcaligenes, Pseudomonas, Thiobacillus*
		Reduction	*Alcaligenes, Pseudomonas, Micrococcus*
Mercury (Hg)	Hg^{2+}, Hg^{0}	Oxidation	*Bacillus, Pseudomonas*
		Reduction	*Pseudomonas, Streptomyces*
Selenium (Se)	Se^{6+}, Se^{4+}, Se^{0}, Se^{2-}	Oxidation	*Bacillus, Thiobacillus*
		Reduction	*Clostridium, Desulfovibrio, Micrococcus*

Data from Sylvia et al. (1999) and Tebo et al. (2005).

are subject to cycles of oxidizing and reducing conditions, such as flooding or a fluctuating water table, the products of Mn^{2+} oxidation can be seen in the form of manganese dioxide (MnO_2) nodules. Microbial dissimilatory reduction of Mn^{4+} is also a common reaction. Most microbes that can reduce Fe^{3+} are also able to reduce Mn^{4+} (Lloyd et al., 2003).

Chromium is required by bacteria as a trace element, but its common use in the metal and tanning industries has made it a priority pollutant in many countries because Cr^{6+} salts are readily mobile, toxic, and carcinogenic. A wide range of microbes are able to reduce the toxic Cr^{6+} form to the Cr^{3+}form. Cr^{3+} is 1000 times less toxic than Cr^{6+}, and its salts are insoluble at neutral pH. The reduction process normally requires anaerobic conditions and has been studied in both facultative anaerobes (e.g., *Shewanella* and *Aeromonas*) and in sulfate reducing bacteria, yet has also been observed in aerobic species of *Pseudomonas*. The bacteria use the Cr as a terminal electron acceptor. The rate of reduction depends on a variety of environmental conditions, including the availability of appropriate electron donors, pH, temperature, and the presence of other metals (Lloyd et al., 2003).

Arsenic is widely distributed in soils and groundwater and is commonly associated with pyrite and other minerals containing sulfides. It is released in high concentrations into geothermal waters and groundwater, where it is a major health risk in several parts of the world, with over 40 million people considered to be in danger from drinking arsenic-containing water. For many years, arsenic-containing compounds were used as pesticides, and although their use has now been banned, they have left a legacy of contamination in agricultural soils. Arsenic exists as two major forms in soils: arsenate (As^{5+}) and arsenite (As^{3+}), with As^{3+} being more toxic and prevalent under reduced conditions. Arsenic is not associated with any essential intracellular microbial process, although its biochemistry resembles that of phosphate and can interfere with many aspects of phosphate metabolism. Bacterial oxidation and reduction of As occurs as a detoxification mechanism and also provides electron donors and acceptors. Oxidation of As^{3+} as an energy source has been seen for bacteria in the *Agrobacterium*/*Rhizobium* family, in which the *aox* genes encoding the arsenite oxidase enzyme are quite widespread. By contrast, As^{5+} reduction, as a terminal electron acceptor, is more common and is carried out by a range of different bacteria. The isolation of arsenate-dissimilating bacteria belonging to widely different phylogenetic groups suggests that they are spread throughout the whole bacterial domain. The electron donors coupled with arsenate reduction vary between strains and environments. Arsenic tolerance in bacteria is mediated primarily by active export of arsenic. The *ars* genes encoding this are widespread in soil environments. However, bacteria also carry out oxidative methylation of arsenic to produce methylarsenite, dimethylarsenate, dimethylarsenite, and trimethylarsine oxide. The ability to carry out this process has been described in a limited number of genera (*Clostridium*, *Desulfovibrio*, *Methanobacterium*), although the *arsM* gene associated with arsenic methylation has been found in over 120 different bacteria and archaea (Slyemi and Bonnefoy, 2012; Stolz et al., 2006).

Fungi can also strongly affect other elemental cycles in soil. Fungi have high requirements in potassium and take it up by two types of transport systems: high affinity and low affinity (Corratge et al., 2007). Paris et al. (1995) showed that two ectomycorrhizal fungi (*Pisolithus tinctorius* and *Paxillus involutus*) were able to solubilize phlogopite (a mica) and to access potassium trapped in the 2:1 layers. Although *Paxillus involutus* irreversibly transformed a fraction of the mica in hydroxy-aluminous vermiculite, the transformations caused by *Pisolithus tinctorius* remained reversible. More recently, Bonneville et al. (2009) studied the weathering of biotite at the nanoscale level by *Paxillus involutus* grown in symbiosis with *Pinus sylvestris*. They showed that the adherence of the hyphae on the mineral surface causes a mechanical distortion of the lattice structure of the mineral, and that chemical weathering leading to the development of a potassium depletion zone only starts later, leading to the oxidation of Fe^{2+} to Fe^{3+} and to the formation of vermiculite and Fe oxides.

The role of Ca for fungal growth has been debated for a long time, but it is now recognized as a very important secondary messenger within the fungal cell that can transmit a primary stimulus at the outer membrane into intracellular events. The concentration of Ca must be kept at a very low level within the cytoplasm (100 nM), whereas Ca is stored at high concentrations in the vacuole (1 mM). Oxalate-Ca precipitates are often seen on fungal hyphae. Beside the probable role of oxalate in mineral weathering and in softening cell walls, oxalate also regulates the concentration of free Ca and the pH in the cytoplasm (Jennings, 1995). The degradation of oxalate-Ca produced by fungi and plants in the presence of bacteria by the so-called oxalate-carbonate pathway can result in the precipitation of calcite (Martin et al., 2012). The oxalate-carbonate pathway has been shown to produce significant amounts of calcite under trees such as iroko (*Milicia excelsa*) in tropical soils (Cailleau et al., 2011).

Essential heavy metals (Zn^{2+}, Cu^{2+}, Mn^{2+}) are taken up in fungi by transporters. These metals are used in enzymatic reactions, but their concentrations are tightly regulated to avoid toxicity. Bolchi et al. (2011) recently identified the genes involved in metal homeostasis in *Tuber melanosporum*. Mycorrhizal fungi have been shown to alleviate stress to plants growing on soils containing large amounts of heavy metals (Colpaert et al., 2011; Hildebrandt et al., 2007). The mechanisms involved in metal homeostasis are extracellular precipitation (e.g., in the presence of oxalate), sorption on the cell wall and within the cell, binding to low-molecular-weight compounds (glutathione, phytochelatins, metallothioneins, nicotianamine), and transport into the vacuole or extrusion back to the soil solution (Bolchi et al., 2011; Colpaert et al., 2011; Jennings, 1995). Systems allowing the detoxification of reactive-oxygen species resulting from the presence of excessive metal concentrations also contribute to the tolerance of fungi exposed to high metal concentrations (Hildebrandt et al., 2007).

IV EXAMPLES OF INTERCONNECTIONS BETWEEN MICROBIAL COMMUNITY/ACTIVITY AND ELEMENT CYCLES

A Element Cycles During Early Soil Development

The role of living organisms on weathering and soil development has been extensively discussed (e.g., see Finlay et al., 2009; Peltzer et al., 2010). However, there is less information on how nutrient cycles develop during the early stages of soil formation (10s to 100s of years). In this section, we show how the interactions of soil biota and plants affect cycling of nutrients along a soil chronosequence developed in the forefield of the Damma Glacier in the Swiss Alps (Bernasconi et al., 2011). It includes unvegetated soils that are as young as 10 years to vegetated soils deglaciated 150 years ago. The forefield is located at 2000 m altitude on a granitic parent material and subjected to an alpine climate with very strong microclimatic heterogeneity caused by the

different exposure of the slopes (south slope vs. north slope) and topographic heterogeneity (presence of two frontal moraines, 1928 and 1992, and presence of dry and wet sites).

The vegetation of the youngest sites located behind the 1992 moraine (soils between 6 and 14 years) is scarce and patchy. It is dominated by *Agrostis gigantea* Roth, *Rumex scutatus* L., *Cerastium uniflorum* Thom. ex Reichb., and *Oxyria digyna* (L.) Hill. The vegetation of the intermediate sites located between the 1992 and 1928 moraines (soils between 57 and 79 years) cover the ground from partially to fully. The dominant species are *Agrostis gigantea*, *Salix* spp., *Deschampsia cespitosa* (L.) Roem. and Schult., and *Athyrium alpestre* (Hoppe) Milde. The vegetation of the sites located in front of the 1928 moraine (soils between 108 and 140 years) fully covers the soil and is characterized by the presence of woody plants (e.g., *Rhododendron ferrugineum* L. and *Salix* spp.) and grasses (e.g., *Agrostis gigantea* and *Festuca rubra* L). Nitrogen-fixing plants (e.g., *Alnus viridis* [Chaix] and *Lotus alpinus*) are also present on the chronosequence (Bernasconi et al., 2011; Brankatschk et al., 2011).

Plant biomass production, together with soil total organic C, N, and total P content increase with soil age. Total clay increases with soil age as soil pH decreases. Soil microbial biomass, as estimated by total DNA, phospholipid fatty acids, and microbial C content, also increases with soil age (Bernasconi et al., 2011). Similarly, the abundance and richness of testate amoebae also increase with soil age.

Zumsteg et al. (2011) conducted genetic profiling and clone library sequencing to characterize the microbial communities of soils along the Damma Glacier forefield. The major bacterial lineages were Proteobacteria, Actinobacteria, Acidobacteria, Firmicutes, and Cyanobacteria. The bacterial diversity was high, but no trend in the Shannon diversity index could be detected across the chronosequence. Euryarchaeota were found to predominantly colonize younger soils, whereas Crenarchaeota colonized mainly older soils. Ascomycota dominated the fungal community in younger soils, whereas Basidiomycota were more general in older soils. Welc et al. (2012) characterized soil bacterial and fungal communities along the forefield using fatty acid profiling. These authors found that the ratio of arbuscular mycorrhizal fungi to bacteria, and of AMF to other fungi, decreased with soil age, whereas the ratio of other fungi to bacteria remained constant with soil age. This suggests that AMF are more important in younger soils and become less important in older soils as they become more acidic and enriched in organic matter.

Goeransson et al. (2011) studied bacterial growth limitation in soils from the Damma using leucine incorporation under laboratory conditions. Bacterial growth increases with soil age (with total soil C), but as soil gets older, soil C availability to bacteria decreases. In the younger soils, bacterial growth is limited by low C and N availability, whereas in the older soils, bacterial growth is only limited by C. Bacterial growth is never limited by P. Esperschuetz et al. (2011) studied the degradation of ^{13}C enriched litter of *Leucanthemopsis alpina*

L, which is to be found along the entire chronosequence. Their results suggest that the contribution of bacteria (actinomycetes) to litter turnover becomes higher with soil age, whereas archaea, fungi, and protozoa play a more important role in recently deglaciated soil.

Duc et al. (2009) and Brankatschk et al. (2011) studied the role of the soil microbial biomass on the N cycle in the soils from the Damma forefield. Duc et al. (2009) observed a higher rate of biological N fixation by free-living microorganisms in the rhizosphere soil of pioneer plants (*Leucanthemopsis alpina*, *Poa alpina*, *Agrostis* sp.) compared to the bulk soil in a younger soil (8 years) and an older soil (70 years). These authors observed a very high diversity of NifH (the iron protein of the nitrogenase complex) in these soils. Altogether, these results suggest that diazotrophs play a significant role for N input in the Damma forefield. The high diversity of NifH protein was related to the very low soil inorganic N content (Duc et al., 2009). Brankatschk et al. (2011) analyzed the abundance of different genes related to the N cycle and the potential enzyme activities related to these genes in the soils of the Damma forefield. Their results show that in the youngest soils (10 years), N mineralization (chitinase and protease) was the main driver of the soil N turnover. In the intermediate soils (50-70 years), genes related to biological N fixation by free-living organisms were highest. Finally, in the older soils (120-2000 years), genes coding for nitrification (AOA) and denitrification (*nosZ*) enzymes and their potential activities were highest. From their results, Brankatschk et al. (2011) suggest that N and C are provided in sufficient amounts for initial microbial development at the youngest sites, whether through the deposition of allochthonous organic material on the forefield (e.g., animal dung), atmospheric deposition on the surface of the glacier, or the presence of microbes able to feed on ancient C.

Schmalenberger and Noll (2010) analyzed the diversity of the sulfonate-desulfurizing bacteria in soils of the Damma forefield based on the sequences of the oxidoreductase *asfA* gene. They studied bulk soils and rhizosphere soils from *Agrostis rupestris* and *Leucanthemopsis alpina*. Their study shows a wide diversity of this gene, which is affected by both soil age and the plant species. They related this high diversity to both the low level of soil available sulfate and to the inoculation of bacteria by atmospheric deposition. *asfA* associated with *Polaromonas*, a genus associated with very low sulfate availability environments, was found in the younger soils, whereas unidentified species were found in the older soils.

Frey et al. (2010) grew bacteria isolated from unvegetated granitic sand from the Damma forefield in the presence of glucose, ammonium, and granite powder from the forefield. They found that isolates of *Arthrobacter* sp., *Janthinobacterium* sp., *Leifsonia* sp., and *Polaromonas* sp. were able to cause a significant increase of granite dissolution as measured by Fe, Ca, K, Mg, and Mn release. The effects of these bacteria on weathering were related to the fact that: (1) they could attach to the granite surfaces, (2) they secreted high amounts of oxalic acid, (3) they lowered the pH of the solution, and (4) they produced HCN. However,

these bacteria had a weak impact on P solubilization. Brunner et al. (2011) isolated fungi from unvegetated granitic sediments in the Damma forefield and conducted granite dissolution studies. They showed that *Mucor hiemalis*, *Umbelopsis isabellina*, and *Mortierella alpina* were able to exude large amounts of citrate, malate, and oxalate and could thereby release significant amounts of Ca, Cu, Fe, Mg, Mn, and P from granite powder. This work shows that fungi are also able to weather minerals in the absence of plants. By analyzing the isotopic composition of oxygen associated with phosphate in different soil and plant pools along the Damma chronosequence, Tamburini et al. (2012) found that at each sampled site the soil available P had been fully processed by soil microbial biomass. These results suggest that at the youngest site, P was taken up by soil microorganisms as soon as it was released from the apatite. The turnover time of microbial P at this site might be as short as a few weeks.

The insights from the publications above are summarized in Fig. 16.5.

The parent material is colonized by microorganisms using C from exogenous inputs to fix N and which release P, Ca, Mg, Fe, and so on from the parent material. These elements arrive in the soil solution in forms that can be taken up by the plant. This allows the development of different plant species and an increase in plant biomass. In the younger stages, AMF mediate an efficient capture of nutrients by plants. The development of N-fixing plants (*Alnus, Lotus*) allows a higher rate of N inputs into the ecosystem. At a later stage, the litter produced by the plants and the SOM can be degraded by soil microorganisms and microfauna, and the elements are then recycled in the microbial-plant loop. This organic matter or direct C inputs from the plant through rhizodeposition can be used by microorganisms to accelerate the weathering of minerals. This leads to deeper, more developed soils. Finally, as the ecosystem becomes richer in available nutrients, the tendency to lose a fraction of these to water and the atmosphere will increase.

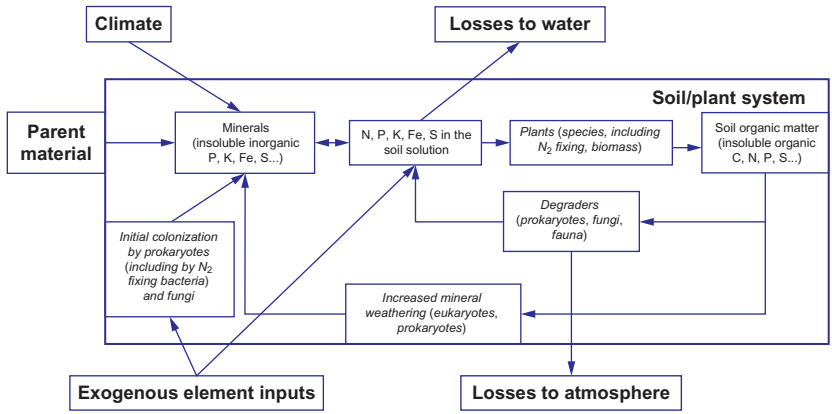

FIG. 16.5 Element cycling during the early stages of soil development as affected by the interactions between soil prokaryotes, fungi, fauna, and plants.

B Coupling of Iron and Sulfur Transformations in Acid Mine Drainage

The leachate that comes from the waste rocks and mineral tailings left behind after mining operations is known as *acid mine drainage* (AMD), and its release has economic and environmental impacts worldwide. As the name implies, AMD is extremely acidic, with pH values as low as 2-3, and it also contains high concentrations of metal ions, both Fe and more toxic heavy metals. In ores, metals such as Cu, Pb, and Zn are commonly found as sulfide minerals together with pyrite (FeS_2), and coal deposits can contain up to 20% of S by weight, largely as sulfides. The combination of microbial Fe and S transformations leads to the production of sulfuric acid that characterizes AMD and is environmentally damaging.

The overall reaction of pyrite minerals involves reaction of FeS_2 with oxygen to generate sulfuric acid (Reaction 16.1). However, this process requires higher concentrations of oxygen than are commonly present in the subsurface environment. This reaction is much more efficient when the oxidant is not molecular oxygen, but Fe^{3+} (Reaction 16.2), which is thereby converted to the ferrous form (Fe^{2+}). Under the acidic conditions of AMD, the Fe^{2+} is then reoxidized to Fe^{3+} by Fe-oxidizing bacteria, the best-characterized of which is *Acidithiobacillus ferrooxidans* (Reaction 16.3).

$$FeS_2 + 3.5O_2 + H_2O \rightarrow Fe^{2+} + 2H^+ + 2SO_4^{2-} \quad (16.1)$$

$$FeS_2 + 14Fe^{3+} + 8H_2O \rightarrow 15Fe^{2+} + 16H^+ + 2SO_4^{2-} \quad (16.2)$$

$$14Fe^{2+} + 3.5O_2 + 14H^+ \rightarrow 14Fe^{3+} + 7H_2O \quad (16.3)$$

Due to the nature of AMD, the key organisms catalyzing this process are necessarily acidophiles. These include not just autotrophs like *A. ferrooxidans* and *Leptospirillum ferrooxidans*, but also a range of acidophilic heterotrophs belonging to the *Acidophilum* genus, and heterotrophic archaea such as *Ferroplasma acidiphilum*. Because the inputs of organic C and N to these systems are minimal, the population of heterotrophs is low. Most of the known acid-tolerant archaea are thermophilic, whereas the temperatures in AMD environments tend to be constantly low. The dominant organisms in these environments therefore tend to be psychrophiles (Hallberg, 2010; though there are a few mines where the energy released from rapid pyrite oxidation is sufficient to cause increased temperatures).

In principle, the presence of high levels of sulfate in AMD might select for heterotrophic sulfate-reducing bacteria in AMD sediments. Although evidence for sulfate reduction has occasionally been observed (presence of blackened sulfide deposits in AMD sediment), no acidophilic sulfate-reducing bacteria or archaea are known. Many members of the AMD microbial community can carry out S oxidation, using either sulfide, S, or reduced S compounds, such as trithionate or tetrathionate, as an electron donor. The common acidophile

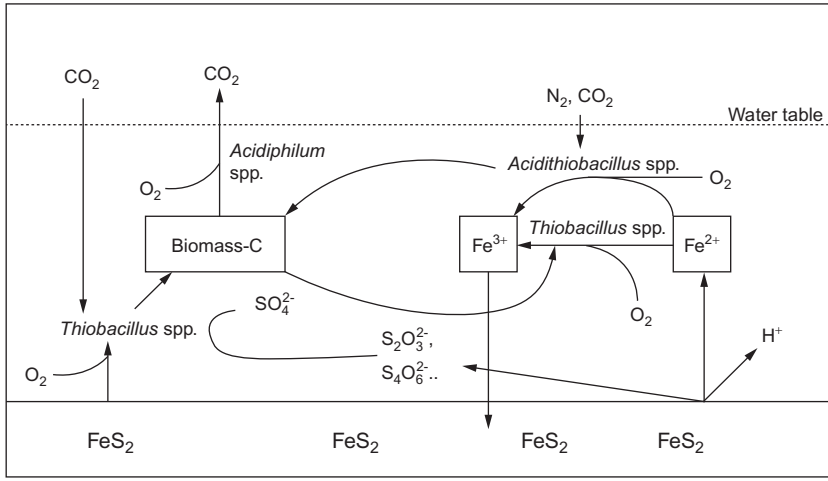

FIG. 16.6 Simplified overview of the processes in acid mine drainage and the key groups of organisms that control these transformations at temperatures <30 °C. *Redrawn from Baker and Banfield (2003).*

A. ferrooxidans can grow autrophically with elemental S as the electron donor and Fe^{3+} as the electron acceptor, and thiobacilli, like *Thiobacillus acidophilus,* use tetrathionate or trithionate for autotrophic growth. Overall, AMD ecosystems contain a fairly limited range of bacterial and archaeal taxa, but there is significant variation within these taxa due to the availability of a range of specialist niches that provide slightly different conditions of pH, temperature, and metal content (Fig. 16.6).

C Integration of Element Cycles in Wetland Systems

Wetland ecosystems provide excellent examples of the interplay between plants and different groups of microbes under a range of environmental conditions. The deeper sediments tend to be anoxic, but this varies through the year, both in natural wetland systems (changes in groundwater flow at different seasons) and in agricultural environments (temporary flooding of rice fields or other irrigation). The upper layers of the soils tend to be high in organic C from plant inputs and are dominated by interactions with the plant roots themselves, providing C inputs in the form of root-derived materials and a partially aerobic environment in the immediate vicinity of the roots due to oxygen release from the root tissues.

In the anoxic environment of flooded wetland soils, ferric compounds are utilized by the resident microbial communities as electron acceptors; reduced Fe compounds therefore accumulate over time. However, when such soils are drained and become aerobic, Fe-oxidizing bacteria become more active, leading to the production of ferric oxyhydroxides (FeOOH), which strongly

bind phosphate. When the soil is flooded once more, the cycle is reversed; once oxygen has been depleted, denitrification at the expense of soil C will dominate microbial respiration until nitrate is exhausted. Dissimilatory Fe reduction then becomes energetically favorable, leading to conversion of the ferric-phosphate complexes mentioned earlier into soluble Fe^{2+}, and releasing inorganic phosphate into the groundwater. Because sulfate reduction and Fe^{3+} reduction are energetically not dissimilar, Fe-reducing activity is quickly followed by an increase in the activity of heterotrophic sulfate reducing bacteria, which convert inorganic sulfate in the groundwater into sulfide using organic matter as electron donors. These sulfide products form highly insoluble iron sulfide precipitates (FeS_x) with reduced iron compounds, which are then stable until the system becomes aerobic once again (Fig. 16.7; Burgin et al., 2011).

There are a number of consequences of this complex combination of Fe, N, S, and P cycles. Once the water table falls in the wetland and the soil once again becomes aerobic, the FeS_x precipitates are oxidized by Fe-oxidizing bacteria, producing a pulse of sulfuric acid as a by-product of the process. The resultant acidification of the wetland soil can cause changes in both plant and microbial communities and can be sufficient to mobilize toxic metals, such as aluminum. Leachate from the wetland can then have damaging environmental effects on ecosystems downstream (Burgin et al., 2011).

In the past century, the application of synthetic fertilizers has had a dramatic effect on both natural and agricultural wetlands. Nitrate is one of the most common groundwater contaminants, and nitrate runoff contributes to surface water eutrophication. However, the impact of nitrate in wetlands goes beyond the N cycle. Although nitrate concentrations are high, ferrous concentrations in the groundwater tend to remain low because nitrate is energetically more favorable as an electron acceptor and therefore provides a redox buffer before ferric compounds are reduced. However, the presence of increased nitrate levels also

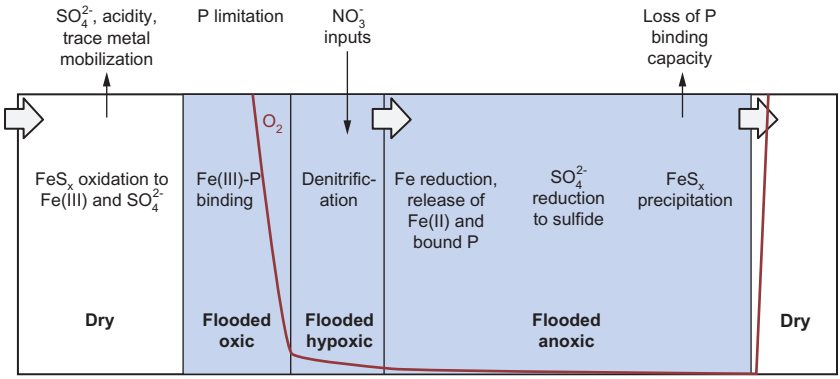

FIG. 16.7 Hypothetical cycle of Fe and S transformations in wetlands and their implications for P and N metabolism. Note that the length of the Fe and sulfate reduction phases under anoxic conditions will vary depending on nitrate inputs to the system. *Adapted from Burgin et al. (2011).*

stimulates what has been described as the "ferrous wheel," where chemolithoautotrophic nitrate reduction is coupled to the oxidation of Fe sulfide deposits, causing an increase in sulfate concentrations in the groundwater. This effect can be considerable; for example, between 1960 and 2000, sulfate concentrations in Dutch groundwater increased nearly threefold, where up to 70% of this sulfate is thought to be derived from pyrite (FeS_x). Sulfate-reducing bacteria convert this sulfate to sulfide, which displaces even more phosphate from Fe-P complexes, causing a large pulse of phosphate to be released. Hence the nitrate runoff and leaching from agricultural soils in this case can also cause phosphate-induced eutrophication within the wetland environment and reduced phosphate binding capacity of the soil in subsequent oxic/anoxic cycles (Smolders et al., 2010).

Nitrate pollution of wetland groundwater also affects the vegetation composition as a result of these coupled element cycles. Wetland plants are largely adapted to prefer ammonium (NH_4^+) as an N source, although this depends on the pH of the surface water. Changes in pH and in the availability of N sources can have a selective effect on the plant community. More importantly, the sulfide produced by sulfate-reducing bacteria and archaea is toxic to plants and also inhibits the growth of many plant species. Precipitation of iron-sulfide as a plaque around the roots interferes with root physiology and P uptake and generates significant problems, yet these problems are to some extent alleviated by diffusion of oxygen from the plant root, which stimulates the activity of S-oxidizing bacteria in the immediate vicinity of the root, converting toxic sulfide to elemental S (Lamers et al., 2012).

REFERENCES

Albrecht-Gary, A.M., Crumbliss, A.L., 1998. Coordination chemistry of siderophores: thermodynamics and kinetics of iron chelation and release. Met. Ions Biol. Syst. 35, 239–327.

Autry, A.R., Fitzgerald, J.W., 1990. Sulfonate S—a major form of forest soil organic sulfur. Biol. Fertil. Soils 10, 50–56.

Baker, B.J., Banfield, J.F., 2003. Microbial communities in acid mine drainage. FEMS Microbiol. Ecol. 44, 139–152.

Barona-Gomez, F., Lautru, S., Francou, F.X., Leblond, P., Pernodet, J.L., Challis, G.L., 2006. Multiple biosynthetic and uptake systems mediate siderophore-dependent iron acquisition in *Streptomyces coelicolor* A3(2) and *Streptomyces ambofaciens* ATCC 23877. Microbiology 152, 3355–3366.

Benning, C., 1998. Biosynthesis and function of the sulfolipid sulfoquinovosyl diacylglycerol. Annu. Rev. Plant Physiol. Plant Mol. Biol. 49, 53–75.

Bernasconi, S.M., Bauder, A., Bourdon, B., et al., 2011. Chemical and biological gradients along the Damma glacier soil chronosequence, Switzerland. Vadose Zone J. 10, 867–883.

Blackwell, M.S.A., Brookes, R.C., de la Fuente-Martinez, N., Gordon, H., Murray, P.J., Snars, K.E., Williams, J.K., Bol, R., Haygarth, P.M., 2010. Phosphorus solubilization and potential transfer to surface waters from the soil microbial biomass following drying-wetting and freezing-thawing. Adv. Agron. 106, 1–35.

Bojinova, D., Velkova, R., Ivanova, R., 2008. Solubilization of Morocco phosphorite by *Aspergillus niger*. Bioresour. Technol. 99, 7348–7353.

Bolchi, A., Ruotolo, R., Marchini, G., Vurro, E., di Toppi, L.S., Kohler, A., Tisserant, E., Martin, F., Ottonello, S., 2011. Genome-wide inventory of metal homeostasis-related gene products including a functional phytochelatin synthase in the hypogeous mycorrhizal fungus *Tuber melanosporum*. Fungal Genet. Biol. 48, 573–584.

Bonneville, S., Smits, M.M., Brown, A., Harrington, J., Leake, J.R., Brydson, R., Benning, L.G., 2009. Plant-driven fungal weathering: early stages of mineral alteration at the nanometer scale. Geology 37, 615–618.

Boye, K., Almkvist, G., Nilsson, S.I., Eriksen, J., Persson, I., 2011. Quantification of chemical sulphur species in bulk soil and organic sulphur fractions by S K-edge XANES spectroscopy. Eur. J. Soil Sci. 62, 874–881.

Brankatschk, R., Toewe, S., Kleineidam, K., Schloter, M., Zeyer, J., 2011. Abundances and potential activities of nitrogen cycling microbial communities along a chronosequence of a glacier forefield. ISME J. 5, 1025–1037.

Brunner, I., Ploetze, M., Rieder, S., Zumsteg, A., Furrer, G., Frey, B., 2011. Pioneering fungi from the Damma glacier forefield in the Swiss Alps can promote granite weathering. Geobiology 9, 266–279.

Bünemann, E.K., Prusziz, B., Ehlers, K., 2011. Characterization of phosphorus forms in soil micro-organisms. In: Bünemann, E.K., Oberson, A., Frossard, E. (Eds.), Phosphorus in Action: Biological Processes in Soil Phosphorus Cycling. Springer, Berlin, pp. 37–57.

Bünemann, E.K., Oberson, A., Liebisch, F., Keller, F., Annaheim, K.E., Huguenin-Elie, O., Frossard, E., 2012. Rapid microbial phosphorus immobilization dominates gross phosphorus fluxes in a grassland soil with low inorganic phosphorus availability. Soil Biol. Biochem. 51, 84–95.

Bünemann, E.K., Keller, B., Hoop, D., Jud, K., Boivin, P., Frossard, E., 2013. Increased availability of phosphorus after drying and rewetting of a grassland soil: processes and plant use. Plant Soil 370, 511–526.

Burgin, A.J., Yang, W.H., Hamilton, S.K., Silver, W.L., 2011. Beyond carbon and nitrogen: how the microbial energy economy couples elemental cycles in diverse ecosystems. Front. Ecol. Environ. 9, 44–52.

Cailleau, G., Braissant, O., Verrecchia, E.P., 2011. Turning sunlight into stone: the oxalate-carbonate pathway in a tropical tree ecosystem. Biogeosciences 8, 1755–1767.

Cartron, M.L., Maddocks, S., Gillingham, P., Craven, C.J., Andrews, S.C., 2006. Feo - Transport of ferrous iron into bacteria. Biometals 19, 143–157.

Chen, C.R., Condron, L.M., Davis, M.R., Sherlock, R.R., 2001. Effects of land-use change from grassland to forest on soil sulfur and arylsulfatase activity in New Zealand. Aust. J. Soil Res. 39, 749–757.

Cleveland, C.C., Liptzin, D., 2007. C:N:P stoichiometry in soil: is there a "Redfield ratio" for the microbial biomass? Biogeochemistry 85, 235–252.

Colpaert, J.V., Wevers, J.H.L., Krznaric, E., Adriaensen, K., 2011. How metal-tolerant ecotypes of ectomycorrhizal fungi protect plants from heavy metal pollution. Ann. Forest Sci. 68, 17–24.

Cornelis, P., 2010. Iron uptake and metabolism in pseudomonads. Appl. Microbiol. Biotechnol. 86, 1637–1645.

Corratge, C., Zimmermann, S., Lambilliotte, R., Plassard, C., Marmeisse, R., Thibaud, J.-B., Lacombe, B., Sentenac, H., 2007. Molecular and functional characterization of a Na^+-K^+ transporter from the Trk family in the ectomycorrhizal fungus *Hebeloma cylindrosporum*. J. Biol. Chem. 282, 26057–26066.

Cregut, M., Piutti, S., Vong, P.-C., Slezack-Deschaumes, S., Crovisier, I., Benizri, E., 2009. Density, structure, and diversity of the cultivable arylsulfatase-producing bacterial community in the rhizosphere of field-grown rape and barley. Soil Biol. Biochem. 41, 704–710.

Cregut, M., Piutti, S., Slezack-Deschaumes, S., Benizri, E., 2012. Compartmentalization and regulation of arylsulfatase activities in *Streptomyces* sp., *Microbacterium* sp. and *Rhodococcus* sp. soil isolates in response to inorganic sulfate limitation. Microbiol. Res. 168, 12–21.

Dedourge, O., Vong, P.C., Lasserre-Joulin, F., Benizri, E., Guckert, A., 2004. Effects of glucose and rhizodeposits (with or without cysteine-S) on immobilized-S-35, microbial biomass-S-35 and arylsulphatase activity in a calcareous and an acid brown soil. Eur. J. Soil Sci. 55, 649–656.

Doolette, A.L., Smernik, R.J., 2011. Soil organic phosphorus speciation using spectroscopic techniques. In: Bünemann, E.K., Oberson, A., Frossard, E. (Eds.), Phosphorus in Action Biological Processes in Soil Phosphorus Cycling. Springer, Berlin, pp. 3–36.

Duc, L., Noll, M., Meier, B.E., Buergmann, H., Zeyer, J., 2009. High diversity of diazotrophs in the forefield of a receding alpine glacier. Microb. Ecol. 57, 179–190.

Ehlers, K., Bakken, L.R., Frostegard, A., Frossard, E., Buenemann, E.K., 2010. Phosphorus limitation in a Ferralsol: impact on microbial activity and cell internal P pools. Soil Biol. Biochem. 42, 558–566.

Ehrlich, H.L., Newman, D.K., 2009. Geomicrobiology, fifth ed. CRC Press/Taylor and Francis, Boca Raton.

Emerson, D., Fleming, E.J., McBeth, J.M., 2010. Iron-oxidizing bacteria: an environmental and genomic perspective. Annu. Rev. Microbiol. 64, 561–583.

Erb, T.J., Kiefer, P., Hattendorf, B., Guenther, D., Vorholt, J.A., 2012. GFAJ-1 is an arsenate-resistant, phosphate-dependent organism. Science 337, 467–470.

Eriksen, J., 1997. Sulphur cycling in Danish agricultural soils: turnover in organic S fractions. Soil Biol. Biochem. 29, 1371–1377.

Eriksen, J., 2009. Soil sulfur cycling in temperate agricultural systems. Adv. Agron. 102, 55–89.

Eriksen, J., Lefroy, R.D.B., Blair, G.J., 1995. Physical protection of soil organic S studied by extraction and fractionation of soil organic matter. Soil Biol. Biochem. 27, 1011–1016.

Esperschuetz, J., Perez-de-Mora, A., Schreiner, K., Welzl, G., Buegger, F., Zeyer, J., Hagedorn, F., Munch, J.C., Schloter, M., 2011. Microbial food web dynamics along a soil chronosequence of a glacier forefield. Biogeosciences 8, 3283–3294.

Fierer, N., Strickland, M.S., Liptzin, D., Bradford, M.A., Cleveland, C.C., 2009. Global patterns in belowground communities. Ecol. Lett. 12, 1238–1249.

Finlay, R., Wallander, H., Smits, M., Holmstrom, S., Van Hees, P., Lian, B., Rosling, A., 2009. The role of fungi in biogenic weathering in boreal forest soils. Fungal Biol. Rev. 23, 101–106.

Freney, J.R., Melville, G.E., Williams, C.H., 1975. Soil organic matter fractions as sources of plant available sulfur. Soil Biol. Biochem. 7, 217–221.

Frey, B., Rieder, S.R., Brunner, I., Ploetze, M., Koetzsch, S., Lapanje, A., Brandl, H., Furrer, G., 2010. Weathering-associated bacteria from the Damma glacier forefield: physiological capabilities and impact on granite dissolution. Appl. Environ. Microbiol. 76, 4788–4796.

Frossard, E., Condron, L.M., Oberson, A., Sinaj, S., Fardeau, J.C., 2000. Processes governing phosphorus availability in temperate soils. J. Environ. Qual. 29, 15–23.

Gadd, G.M., Rhee, Y.J., Stephenson, K., Wei, Z., 2012. Geomycology: metals, actinides and biominerals. Environ. Microbiol. Rep. 4, 270–296.

Ghani, A., McLaren, R.G., Swift, R.S., 1992. Sulphur mineralisation and transformations in soils as influenced by additions of carbon, nitrogen and sulphur. Soil Biol. Biochem. 24, 331–341.

Ghani, A., McLaren, R.G., Swift, R.S., 1993a. Mobilization of recently-formed soil organic sulphur. Soil Biol. Biochem. 25, 1739–1744.

Ghani, A., McLaren, R.G., Swift, R.S., 1993b. The incorporation and transformations of sulfur-35 in soil: effects of soil conditioning and glucose or sulphate additions. Soil Biol. Biochem. 25, 327–335.

Gianfreda, L., Ruggiero, P., 2006. Enzyme activities in soil. In: Nannipieri, P., Smalla, K. (Eds.), Nucleic Acids and Proteins in Soil. Springer, Berlin.

Glick, B.R., 2010. Using soil bacteria to facilitate phytoremediation. Biotechnol. Adv. 28, 367–374.

Goeransson, H., Venterink, H.O., Baath, E., 2011. Soil bacterial growth and nutrient limitation along a chronosequence from a glacier forefield. Soil Biol. Biochem. 43, 1333–1340.

Goux, X., Amiaud, B., Piutti, S., Philippot, L., Benizri, E., 2012. Spatial distribution of the abundance and activity of the sulfate ester-hydrolyzing microbial community in a rape field. J. Soils Sediments 12, 1360–1370.

Griffiths, B.S., Philippot, L., 2013. Insights into the resistance and resilience of the soil microbial community. FEMS Microbiol. Rev. 37, 112–129.

Gyaneshwar, P., Kumar, G.N., Parekh, L.J., Poole, P.S., 2002. Role of soil micro-organisms in improving P nutrition of plants. Plant Soil 245, 83–93.

Hallberg, K.B., 2010. New perspectives in acid mine drainage microbiology. Hydrometallurgy 104, 448–453.

Hedrich, S., Schlomann, M., Johnson, D.B., 2011. The iron-oxidizing proteobacteria. Microbiology 157, 1551–1564.

Hildebrandt, U., Regvar, M., Bothe, H., 2007. Arbuscular mycorrhiza and heavy metal tolerance. Phytochemistry 68, 139–146.

Hinsinger, P., Plassard, C., Tang, C.X., Jaillard, B., 2003. Origins of root-mediated pH changes in the rhizosphere and their responses to environmental constraints: a review. Plant Soil 248, 43–59.

Hsieh, Y.J., Wanner, B.L., 2010. Global regulation by the seven-component Pi signaling system. Curr. Opin. Microbiol. 13, 198–203.

Jennings, D.H., 1995. The Physiology of Fungal Nutrition. Cambridge University Press, Cambridge.

Keller, M., Oberson, A., Annaheim, K.E., Tamburini, F., Mäder, P., Mayer, J., Frossard, E., Bünemann, E.K., 2012. Phosphorus forms and enzymatic hydrolysability of organic phosphorus in soils after 30 years of organic and conventional farming. J. Plant Nutr. Soil Sci. 175, 385–393.

Kelly, D.P., Murrell, J.C., 1999. Microbial metabolism of methanesulfonic acid. Arch. Microbiol. 172, 341–348.

Kertesz, M.A., 1999. Riding the sulfur cycle—metabolism of sulfonates and sulfate esters in Gram-negative bacteria. FEMS Microbiol. Rev. 24, 135–175.

Kertesz, M.A., Fellows, E., Schmalenberger, A., 2007. Rhizobacteria and plant sulfur supply. Adv. Appl. Microbiol. 62, 235–268.

Kidd, P., Barcelo, J., Bernal, M.P., Navari-Izzo, F., Poschenrieder, C., Shilev, S., Clemente, R., Monterroso, C., 2009. Trace element behaviour at the root-soil interface: implications in phytoremediation. Environ. Exp. Bot. 67, 243–259.

King, J.E., Quinn, J.P., 1997. The utilization of organosulphonates by soil and freshwater bacteria. Lett. Appl. Microbiol. 24, 474–478.

Kirchman, D.L., 2012. Processes in Microbial Ecology. Oxford University Press, New York.

Kirkby, C.A., Kirkegaard, J.A., Richardson, A.E., Wade, L.J., Blanchard, C., Batten, G., 2011. Stable soil organic matter: a comparison of C:N:P:S ratios in Australian and other world soils. Geoderma 163, 197–208.

Kizewski, F., Liu, Y.T., Morris, A., Hesterberg, D., 2011. Spectroscopic approaches for phosphorus speciation in soils and other environmental systems. J. Environ. Qual. 40, 751–766.

Kraemer, S.M., 2004. Iron oxide dissolution and solubility in the presence of siderophores. Aquat. Sci. 66, 3–18.

Lamers, L.P.M., van Diggelen, J.M., Op den Camp, H.J., Visser, E.J.W., Lucassen, E.C., Vile, M.A., Jetten, M.S.M., Smolders, A.J., Roelofs, J.G., 2012. Microbial transformations of nitrogen, sulfur, and iron dictate vegetation composition in wetlands: a review. Front. Microbiol. 3, 156.

Laslo, E., Gyorgy, E., Mara, G., Tamas, E., Abraham, B., Lanyi, S., 2012. Screening of plant growth promoting rhizobacteria as potential microbial inoculants. Crop. Prot. 40, 43–48.

Lloyd, J.R., Lovley, D.R., Macaskie, L.E., 2003. Biotechnological application of metal-reducing micro-organisms. Adv. Appl. Microbiol. 53, 85–128.

Loper, J.E., Buyer, J.S., 1991. Siderophores in microbial interactions on plant surfaces. Mol. Plant-Microbe Interact. 4, 5–13.

Lovley, D.R., 2012. Electromicrobiology. Annu. Rev. Microbiol. 66, 391–409.

Martin, G., Guggiari, M., Bravo, D., Zopfi, J., Cailleau, G., Aragno, M., Job, D., Verrecchia, E., Junier, P., 2012. Fungi, bacteria and soil pH: the oxalate-carbonate pathway as a model for metabolic interaction. Environ. Microbiol. 14, 2960–2970.

Marzluf, G.A., 1997. Molecular genetics of sulfur assimilation in filamentous fungi and yeast. Annu. Rev. Microbiol. 51, 73–96.

McGill, W.B., Cole, C.V., 1981. Comparative aspects of cycling of organic C, N, S and P through soil organic matter. Geoderma 26, 267–286.

Metcalf, W.W., Griffin, B.M., Cicchillo, R.M., et al., 2012. Synthesis of methylphosphonic acid by marine microbes: a source for methane in the aerobic ocean. Science 337, 1104–1107.

Meyer, B., Kuever, J., 2007. Molecular analysis of the diversity of sulfate-reducing and sulfur-oxidizing prokaryotes in the environment, using *aprA* as functional marker gene. Appl. Environ. Microbiol. 73, 7664–7679.

Miao, Z., Brusseau, M.L., Carroll, K.C., Carreon-Diazconti, C., Johnson, B., 2012. Sulfate reduction in groundwater: characterization and applications for remediation. Environ. Geochem. Health 34, 539–550.

Muyzer, G., Stams, A.J.M., 2008. The ecology and biotechnology of sulphate-reducing bacteria. Nat. Rev. Microbiol. 6, 441–454.

Olsson, P.A., Hammer, E.C., Wallander, H., Pallon, J., 2008. Phosphorus availability influences elemental uptake in the mycorrhizal fungus *Glomus intraradices*, as revealed by particle-induced X-ray emission analysis. Appl. Environ. Microbiol. 74, 4144–4148.

Paris, F., Bonnaud, P., Ranger, J., Lapeyrie, F., 1995. In vitro weathering of phlogopite by ectomycorrhizal fungi. 1. Effect of K^+ and Mg^{2+} deficiency on phyllosilicate evolution. Plant Soil 177, 191–201.

Passman, F.J., Jones, G.E., 1985. Preparation and analysis of *Pseudomonas putida* cells for elemental composition. Geomicrobiol. J. 4, 191–206.

Peltzer, D.A., Wardle, D.A., Allison, V.J., et al., 2010. Understanding ecosystem retrogression. Ecol. Monogr. 80, 509–529.

Pester, M., Schleper, C., Wagner, M., 2011. The Thaumarchaeota: an emerging view of their phylogeny and ecophysiology. Curr. Opin. Microbiol. 14, 300–306.

Philpott, C.C., 2006. Iron uptake in fungi: a system for every source. Biochim. Biophys. Acta 1763, 636–645.

Plassard, C., Dell, B., 2010. Phosphorus nutrition of mycorrhizal trees. Tree Physiol. 30, 1129–1139.

Plassard, C., Louche, J., Ali, M.A., Duchemin, M., Legname, E., Cloutier-Hurteau, B., 2011. Diversity in phosphorus mobilisation and uptake in ectomycorrhizal fungi. Ann. Forest Sci. 68, 33–43.

Plett, J.M., Martin, F., 2011. Blurred boundaries: lifestyle lessons from ectomycorrhizal fungal genomes. Trends Genet. 27, 14–22.

Prietzel, J., Botzaki, A., Tyufekchieva, N., Brettholle, M., Thieme, J., Klysubun, W., 2011. Sulfur speciation in soil by S K-edge XANES spectroscopy: comparison of spectral deconvolution and linear combination fitting. Environ. Sci. Technol. 45, 2878–2886.

Randriamanantsoa, L., Morel, C., Rabeharisoa, L., Douzet, J.M., Jansa, J., Frossard, E., 2013. Can the isotopic exchange kinetic method be used in soils with a very low water extractable phosphate content and a high sorbing capacity for phosphate ions? Geoderma 200, 120–129.

Reid, R.K., Reid, C.P.P., Szaniszlo, P.J., 1985. Effects of synthetic and microbially produced chelates on the diffusion of iron and phosphorus to a simulated root in soil. Biol. Fertil. Soils 1, 45–52.

Richardson, A.E., Hocking, P.J., Simpson, R.J., George, T.S., 2009. Plant mechanisms to optimise access to soil phosphorus. Crop Pasture Sci. 60, 124–143.

Robin, A., Vansuyt, G., Hinsinger, P., Meyer, J.M., Briat, J.F., Lemanceau, P., 2008. Iron dynamics in the rhizosphere: consequences for plant health and nutrition. Adv. Agron. 99, 183–225.

Roden, E.E., 2012. Microbial iron-redox cycling in subsurface environments. Biochem. Soc. Trans. 40, 1249–1256.

Rodriguez, H., Fraga, R., 1999. Phosphate solubilizing bacteria and their role in plant growth promotion. Biotechnol. Adv. 17, 319–339.

Ross, D.J., 1990. Estimation of soil microbial C by a fumigation extraction method—influence of seasons, soils and calibration with the fumigation incubation procedure. Soil Biol. Biochem. 22, 295–300.

Scherer, P., Lippert, H., Wolff, G., 1983. Composition of the major elements and trace elements of 10 methanogenic bacteria determined by inductively coupled plasma emission spectroscopy. Biol. Trace Elem. Res. 5, 149–163.

Schmalenberger, A., Noll, M., 2010. Shifts in desulfonating bacterial communities along a soil chronosequence in the forefield of a receding glacier. FEMS Microbiol. Ecol. 71, 208–217.

Schmalenberger, A., Hodge, S., Bryant, A., Hawkesford, M.J., Singh, B.K., Kertesz, M.A., 2008. The role of *Variovorax* and other *Comamonadaceae* in sulfur transformations by microbial wheat rhizosphere communities exposed to different sulfur fertilization regimes. Environ. Microbiol. 10, 1486–1500.

Schmalenberger, A., Hodge, S., Hawkesford, M.J., Kertesz, M.A., 2009. Sulfonate desulfurization in *Rhodococcus* from wheat rhizosphere communities. FEMS Microbiol. Ecol. 67, 140–150.

Schmalenberger, A., Telford, A., Kertesz, M.A., 2010. Sulfate treatment affects desulfonating bacterial community structures in *Agrostis* rhizospheres as revealed by functional gene analysis based on *asfA*. Eur. J. Soil Biol. 46, 248–254.

Silva, M.C.P., Semenov, A.V., Schmitt, H., Elsas, J.D.V., Salles, J.F., 2013. Microbe-mediated processes as indicators to establish the normal operating range of soil functioning. Soil Biol. Biochem. 57, 995–1002.

Slyemi, D., Bonnefoy, V., 2012. How prokaryotes deal with arsenic. Environ. Microbiol. Rep. 4, 571–586.

Smith, S.E., Smith, F.A., 2011. Roles of arbuscular mycorrhizas in plant nutrition and growth: new paradigms from cellular to ecosystem scales. Annu. Rev. Plant Biol. 62, 227–250.

Smits, M.M., Bonneville, S., Benning, L.G., Banwart, S.A., Leake, J.R., 2012. Plant-driven weathering of apatite—the role of an ectomycorrhizal fungus. Geobiology 10, 445–456.

Smolders, A.J.P., Lucassen, E., Bobbink, R., Roelofs, J.G.M., Lamers, L.P.M., 2010. How nitrate leaching from agricultural lands provokes phosphate eutrophication in groundwater fed wetlands: the sulphur bridge. Biogeochemistry 98, 1–7.

Sorokin, D.Y., 2003. Oxidation of inorganic sulfur compounds by obligately organotrophic bacteria. Microbiology 72, 641–653.

Stolz, J.E., Basu, P., Santini, J.M., Oremland, R.S., 2006. Arsenic and selenium in microbial metabolism. Annu. Rev. Microbiol. 60, 107–130.

Sylvia, D.M., Fuhrmann, J.F., Hartel, P.G., Zuberer, D.A., 1999. Principles and Applications of Soil Microbiology. Prentice-Hall, New Jersey.

Tamburini, F., Pfahler, V., Buenemann, E.K., Guelland, K., Bernasconi, S.M., Frossard, E., 2012. Oxygen isotopes unravel the role of micro-organisms in phosphate cycling in soils. Environ. Sci. Technol. 46, 5956–5962.

Taylor, J.P., Wilson, B., Mills, M.S., Burns, R.G., 2002. Comparison of microbial numbers and enzymatic activities in surface soils and subsoils using various techniques. Soil Biol. Biochem. 34, 387–401.

Tebo, B.M., Johnson, H.A., McCarthy, J.K., Templeton, A.S., 2005. Geomicrobiology of manganese (II) oxidation. Trends Microbiol. 13, 421–428.

Traxler, M.F., Seyedsayamdost, M.R., Clardy, J., Kolter, R., 2012. Interspecies modulation of bacterial development through iron competition and siderophore piracy. Mol. Microbiol. 86, 628–644.

Uroz, S., Calvaruso, C., Turpault, M.P., Frey-Klett, P., 2009. Mineral weathering by bacteria: ecology, actors and mechanisms. Trends Microbiol. 17, 378–387.

Vong, P.C., Dedourge, O., Lasserre-Joulin, F., Guckert, A., 2003. Immobilized-S, microbial biomass-S and soil arylsulfatase activity in the rhizosphere soil of rape and barley as affected by labile substrate C and N additions. Soil Biol. Biochem. 35, 1651–1661.

Waldron, K.J., Robinson, N.J., 2009. How do bacterial cells ensure that metalloproteins get the correct metal? Nat. Rev. Microbiol. 7, 25–35.

Weber, K.A., Achenbach, L.A., Coates, J.D., 2006. Micro-organisms pumping iron: anaerobic microbial iron oxidation and reduction. Nat. Rev. Microbiol. 4, 752–764.

Welc, M., Buenemann, E.K., Fliessbach, A., Frossard, E., Jansa, J., 2012. Soil bacterial and fungal communities along a soil chronosequence assessed by fatty acid profiling. Soil Biol. Biochem. 49, 184–192.

White, A.K., Metcalf, W.W., 2007. Microbial metabolism of reduced phosphorus compounds. Annu. Rev. Microbiol. 61, 379–400.

Williams, P.H., Haynes, R.J., 1993. Forms of sulfur in sheep excreta and their fate after application on to pasture soil. J. Sci. Food Agric. 62, 323–329.

Wolfe-Simon, F., Blum, J.S., Kulp, T.R., et al., 2011. A bacterium that can grow by using arsenic Instead of phosphorus. Science 332, 1163–1166.

Yang, Z.H., Stoven, K., Haneklaus, S., Singh, B.R., Schnug, E., 2010. Elemental sulfur oxidation by *Thiobacillus* spp. and aerobic heterotrophic sulfur-oxidizing bacteria. Pedosphere 20, 71–79.

Zhang, Y., Gladyshev, V.N., 2009. Comparative genomics of trace elements: emerging dynamic view of trace element utilization and function. Chem. Rev. 109, 4828–4861.

Zhang, Q., van der Donk, W.A., 2012. Answers to the carbon-phosphorus lyase conundrum. Chembiochem 13, 627–629.

Zhao, F.J., Wu, J., McGrath, S.P., 1996. Soil organic sulphur and its turnover. In: Piccolo, A. (Ed.), Humic Substances in Terrestrial Ecosystems. Elsevier, Amsterdam, pp. 467–506.

Zhao, F.J., Lehmann, J., Solomon, D., Fox, M.A., McGrath, S.P., 2006. Sulphur speciation and turnover in soils: evidence from sulphur K-edge XANES spectroscopy and isotope dilution studies. Soil Biol. Biochem. 38, 1000–1007.

Zhou, J.Z., He, Q., Hemme, C.L., et al., 2011. How sulphate-reducing micro-organisms cope with stress: lessons from systems biology. Nat. Rev. Microbiol. 9, 452–466.

Zumsteg, A., Bernasconi, S.M., Zeyer, J., Frey, B., 2011. Microbial community and activity shifts after soil transplantation in a glacier forefield. Appl. Geochem. 26, S326–S329.