

# Chapter 8

## Soil Enzymes and Their Role in Nutrient Cycling



Neemisha and Sandeep Sharma

**Abstract** Soil is a dynamic living non-renewable resource that acts as an interface between agriculture and environment. Soil enzymes play critical role in soil processes ranging from biochemical reactions in plants, animals, and microbes to decomposition of organic matter, soil structure stabilization and nutrients cycling. Different microbes and enzymes are involved in cycling of carbon (C), nitrogen (N), phosphorus (P), and sulfur (S) in an ecosystem. The nutrient cycles function as a result of biological and physico-chemical reactions in soil. The nutrients cycling in an ecosystem are primarily governed by soil microbes and enzyme activities. The biochemical reactions of these cycles are accomplished by enzymes such as amidases, arylsulphatase, cellulases, dehydrogenase, glucosidases, laccase, phosphatases, and urease. These enzymes serve as biological indicators that help to identify variations in soil physical, chemical, and biological properties. Enzymes respond faster to soil management practices long before other soil quality indicator changes are detectable and their method of detection is simple, easy, and quick. In this chapter, we will discuss about the major enzymes involved in the cycling of C, N, P, S, and their mechanisms of action, role in maintaining soil health and factors that affect their activities in soil.

**Keywords** Soil enzymes · biochemical reactions · organic matter · nutrients cycle

### 8.1 Introduction

Soil being an essential resource to biosphere and human beings, is also among the most complex and least understood systems. In soil several biological indicators (soil respiration, microbial biomass, soil fauna, soil organisms (abundance, diversity, structure, community, and food web) etc. are used to assess the quality of soil (Alkorta et al. 2003). Soil organisms respond quickly to agricultural management

---

Neemisha (✉) · S. Sharma  
Department of Soil Science, Punjab Agricultural University, Ludhiana, Punjab, India  
e-mail: [neemisha14@pau.edu](mailto:neemisha14@pau.edu)

practices and climate change. Several useful soil and ecosystem functions are associated with soil organisms such as nutrients cycling, decomposition of organic matter, detoxification of toxicants, suppression of harmful and pathogenic organisms. Microbes and plants secrete certain enzymes into the soil that catalyze specific reactions to release nutrients and the plant growth depends positively on these nutrients (Kandeler et al. 2011). Majority of microbes exist in soil as communities, where they interact in close association with the soil environment. The extracellular enzyme secreted by these organisms play crucial role in transformations of C, N, P, and S at elementary level of nutrient cycling. As compared to physico-chemical parameters of soil, the enzyme activities have been considered as sensitive indicators for soil quality and health (Gelsomino et al. 2006). Plants provide suitable environmental conditions, microhabitats and food reserve for these microorganisms to grow in the rhizosphere (Prober et al. 2015) which in-turn forms various associations with plants like symbiosis, ecto-mycorrhizza and endo-mycorrhizza and improve the nutrient supply to the plants and ultimately increase their yield (Abbott et al. 2015). The plant–microbe interaction in the rhizosphere control the processes like mineralization, transformation of nutrients, organic matter decomposition and agrochemical degradation and improve the soil fertility (Glick 2010; Rajkumar et al. 2013; Song et al. 2019).

Biological indicators are the most informative agents that tell us about the processes and functions mediated by soil organisms. Bacteria, fungi, and plant roots, are primarily responsible for secreting enzyme and hence make possible the flux of C, N, and other essential elements in biogeochemical cycles. The first stage to characterize soil metabolic potential, its quality, fertility, and resilience (natural and anthropogenic factors) is through measuring enzymatic catalysis, understanding the factors that regulate enzyme expression and to determine rates of substrate turnover. A better understanding of the role of soil enzymes activity in the ecosystem will potentially provide a unique opportunity for developing an integrated biological assessment of soils. Moreover, soil biological activities are easy to measure, and produce a rapid response to changes in soil management practices (Caldwell 2005). A good ecological indicator must meet certain conditions (Dale and Beyeler 2001) as: easy measurements, low variability in response, sensitivity to managements and environmental changes, ability to produce consistent, reproducible, and predictable responses to the changes. Moreover, the procedures for enzyme assays are simple and quick. The extracellular enzymes produced by microbes brings about processing and recovery of key nutrients from detrital inputs and accumulated soil organic matter into assimilable subunits (sugars, amino acids,  $\text{NH}_4^+$ ,  $\text{PO}_4^{-3}$ ) (Caldwell 2005). Bacteria are the most abundant microorganisms present in the soil with population of  $10^7$ – $10^9$  per gram soil and fungi has the highest biomass. They secrete various enzymes and are able to either mineralize or partially transform the toxic metabolite into non-toxic form in the soil, where it gets immobilized as humic acid (Murphy 2016). Soil contains free enzymes, immobilized extracellular enzymes and enzymes within microbial cells (intracellular enzymes). Soil enzymes increase decomposition rate of plant residues and help in the release plant-available nutrients.

They serve as catalysts that increases the rate of chemical reactions (Tabatabai 1994) however, they are influenced by several cultural management practices.

## 8.2 Enzymes Involved in Carbon Cycle

Carbon is critical element that contributes for proper functioning and productivity of an ecosystem. Carbon adds quality and life to soil ultimately prevailing healthy conditions in soil system. The availability or release of carbon is mainly dependent on soil management. In C-cycling soil microbes work in soil to decompose the organic matter (Plant/animal) and convert it into simpler forms. This conversion is brought about by several enzymes which work on different constituents of plants and animals. Some of these enzymes that specifically act on specific components of plant and animals are cellulase, hemicellulase, laccase, chitinases, and invertase.

### 8.2.1 Cellulase

Cellulose is a linear polymer made up of  $\beta$ -1,4 linked glucose molecules and cellulases are the enzymes that catalyze the degradation of cellulose. Cellulose is the most abundant biopolymer in plant cell walls (Lynd et al. 2005). Naturally, a combination of three enzymes causes hydrolysis of cellulose and these are endoglucanases, exoglucanases, and  $\beta$ -glucosidase (Yang et al. 2013). Among these, the most common, important, and widely used soil quality indicator is  $\beta$ -glucosidase (Bandick and Dick 1999) which is found in microbes (bacteria, yeasts, and fungi), plants, and animals. The last stage of cellulose degradation process is carried out by  $\beta$ -glucosidase where cellobiose residue is hydrolyzed (Gil-Sotres et al. 2005). The hydrolysis and biodegradation of various  $\beta$ -glucosides in plant debris is carried out by  $\beta$ -glucosidase and the final product is glucose which serves as the energy source for growth and activity of several microbes in the soil (Esen 1993; Merino et al. 2016).  $\beta$ -glucosidase, is considered as one of the most important enzyme for soil quality testing. Cellulase activity in soil is determined on the basis of degradation of substrates such as the cellulose polymer of cellophane, carboxy methyl cellulose, cellulose powder and filter paper assay, and its activity is measured by the DNS method (Pancholy and Rice 1973).

### 8.2.2 Amylase

Starch is a polymer of glucose linked by  $\alpha$  (1  $\rightarrow$  4) glycosidic bonds and it is hydrolyzed by enzyme amylase. Amylases are classified into two types:  $\alpha$ - and  $\beta$ -amylase.  $\alpha$ -Amylase is produced by microorganisms, plants, and, animals,

whereas,  $\beta$ -amylase is produced only by plants.  $\alpha$ -Amylase breaks  $\alpha$  (1  $\rightarrow$  4) glycosidic bond of starch molecule, resulting in the formation of dextrans (Thoma et al. 1971).  $\beta$ -Amylase breaks down starch resulting in the formation of maltose which is further hydrolyzed to glucose units by maltase. The activities of amylase are dependent on several factors including cultural practices, type of vegetation, environment, and soil types. Amylase activities of soil may be directly influenced by the enzyme and exudates secreted by the plants or indirectly by synthetic activities of microorganisms (Ross 1975; Makoi and Ndakidemi 2008).

### 8.2.3 Chitinase

Chitinases are glycosyl hydrolases that catalyze the hydrolytic cleavage of the  $\beta$ -1,4-glycoside bond present in biopolymers of *N*-acetylglucosamine (Collinge et al. 1993). Chitin is the second most abundant polymer in nature after cellulose (Singh et al. 2016). Chitinases are widespread in nature however, their functions are confined to type of organism. They are present in as bacteria and fungi (nutritional needs), plants, and animals (defence against pathogen attacks) and viruses (Singh et al. 2016). Chitinases are classified as endochitinases and exochinases. Endochitinases cleave chitin randomly at internal sites, forming soluble low molecular massmultimers of  $\beta$ -1,4-linked *N*-acetylglucosamine and exochitinases are further classified into two types as chitobiosidases (release of diacetylchitobiose) and 1,4- $\beta$ -*N*-acetylglucosaminidases (cleave endochitinases and chitobiosidases to  $\beta$ -1,4-linked *N*-acetylglucosamine (Sahai and Manocha 1993; Botha et al. 1998; Singh et al. 2016). Plant chitinases play major role by providing defence mechanism against pathogens either by secreting antifungal substances or by eliciting plant defensive responses (Suarez et al. 2001; Gomez et al. 2002). Chitinases helps to convert chitin-containing biomass into depolymerized products, control of insect and fungal pathogens of plants and serves as an indicator of the actively growing fungi in the soil.

### 8.2.4 Laccase

Laccase is a multi-copper oxidase that catalyzes the oxidation of one electron of wide range of phenolic compounds. They need molecular oxygen as co-substrate and release water, so they are considered as eco-friendly enzymes. Laccases are widely distributed in higher plants, bacteria, fungi, lichens, and insects. It is very important enzyme of carbon cycle as it is involved in the degradation of several xenaromatics. Lignin like complex polymers are mainly degraded by laccase producing white-rot fungi along with other extracellular oxidases (Baldrian 2006; Thurston 1994; Claus and Filip 1988). Laccase has significant role in both

lignification and delignification due to its ability to bring about polymerization and depolymerization of compounds (Hatakka 1994; Strong and Claus 2011).

### **8.2.5 Hemicellulase**

Hemicellulase (endo-1,4- $\beta$ -xylanase) enzymes are mainly responsible for decomposition of the polysaccharides of xylose. These are involved in decomposition of the hemicelluloses into short chain glycosides.

### **8.2.6 Invertase**

Invertase catalyzes the hydrolysis of sucrose to glucose and fructose under either acidic or alkaline conditions. This enzyme is present in several organisms as bacteria, actinomycetes, fungi, plants, and animals.

## **8.3 Enzymes Involved in Nitrogen Cycle**

Nitrogen is the most limiting nutrient that is required by the crops in huge quantity. The nitrogen cycle consists of four main steps: nitrogen fixation, ammonification, nitrification, and denitrification. Different microbes and their enzymes complete this cyclic process (Fig. 8.1)

### **8.3.1 Amidohydrolases**

Amidohydrolases are involved in hydrolysis of C–N bond of amides native and added organic N to soils. These include amidase, L-glutaminase, L-asparaginase, and urease that release ammonium into the environment. L-asparaginase catalyzes the hydrolysis of L-asparagine to produce L-aspartic acid and  $\text{NH}_3$  and it is mostly found in microorganisms. Whereas, L-glutaminase, catalyzes the hydrolysis of L-glutamine to L-glutamic acid and  $\text{NH}_3$  and it is widely distributed in plants, animals, and microorganisms (Bacteria, fungi, and yeasts). Estimation of L-asparaginase in soil is used to understand the impact of soil management on N cycling in agricultural ecosystems (Kandeler et al. 2011).

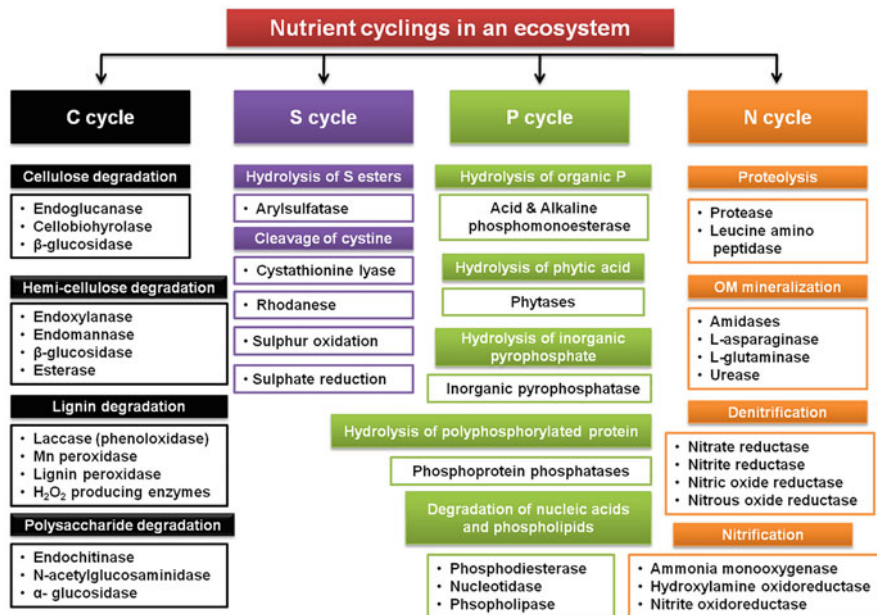


Fig. 8.1 Nutrient cycling of different elements and enzymes associated with these elements

### 8.3.2 Amidase

Amidase (acylamidase, amidohydrolase, acylase, and deaminase) catalyzes the hydrolysis of carbon–nitrogen bond of aliphatic amides producing ammonia and their corresponding carboxylic acids. The enzyme works well at optimum pH of 8.5 and inactivates at temperatures above 60 °C. The substrate of this enzyme is aliphatic amides (formamide, acetamide) from synthetic nitrogenous fertilizers and aromatic amides. This is an inducible enzyme in presence of substrate increases in concentration. Amidases are classified on the basis of catalytic activity (seven types: D-aminopeptidase, aliphatic amidases, aromatic amidases, enantioselective amidases,  $\alpha$ -amino amidase, arylalkylacyl amidases, wide spectrum amidases), amino acid sequence (two types: signature amidases and aliphatic amidases) and phylogenetic relationship (two types: aspartic proteinases and sulfhydryl enzymes) (Chebrou et al. 1996; Pace and Brenner 2001; Fournand and Arnaud 2001; Pertsovich et al. 2005). Amidases play very important role in soil because their distribution in soil profiles helps in reducing the rates of hydrolysis of N fertilizers applied. Ammonia and carboxylic acids are by products of amidase action that serves important role in soil nutrient cycling, metabolic processes and N mineralization. This enzyme controls N in ecosystems contributes toward assessing soil degradation or quality.

### 8.3.3 *Urease*

Urease enzyme hydrolyzes urea to ammonia and carbon dioxide. This reaction increases the pH of soil and causes instant loss of N through volatilization of  $\text{NH}_3$  (Zhang et al. 2014). Urease is produced intra as well as extracellularly in soil microbes and plants. Several factors affect urease stability in soil such as their association with soil organo-mineral or humus-urease complexes is more stable and highly resistant to higher temperatures and proteolytic attack. In agricultural soils urease activity detect N mineralization when soil is amended with organic material. The urease activity is affected by soil properties that include soil nutrient supply, soil pH,  $S_{\text{MBN}}$ ,  $S_{\text{N}}$ , N fertilizers, tillage, agrochemicals use and cropping systems (Moghimian et al. 2017). The hydrolysis of urea occurs in the presence of soil enzyme urease and thus  $\text{NH}_4^+$  is made available to the plants (Wang et al. 2008).

### 8.3.4 *Proteases*

Protease is widely distributed in nature and produced by plants, animals, and microbes through their metabolic activities. This enzyme brings about initial hydrolysis of protein bound to organic nitrogen to polypeptides and oligo-peptides to simple amino acids. The hydrolysis of proteins is an important step in the nitrogen cycle where proteases hydrolyze both native and added proteins in soil (Dedeken and Voets 1965; Raju et al. 2017). The addition of organic inputs increases protease activity in soil whereas, the treatment of crops with agrochemicals often reduces protease activity in soil (Raju et al. 2017).

### 8.3.5 *Nitrification Enzymes*

Nitrification is biological process of oxidation of ammonia to nitrite and nitrate in a two-step procedure which is mediated by microbes and their enzymes. In first step ammonia is oxidized to hydroxylamine by ammonia-oxidizing bacteria through monooxygenase enzyme followed by its conversion to nitrite by hydroxylamine oxidoreductase. The nitrite is further oxidized to nitrate by the nitrite-oxidizing bacteria through ammonia monooxygenase enzyme and this is the rate-limiting step in nitrification process (Kandeler et al. 2011).

### 8.3.6 Denitrification Enzymes

Denitrification process is a four step reaction where nitrate is reduced to dinitrogen gas. The first step consists of reduction of nitrate to nitrite by nitrate reductase; second step reduction of nitrite ( $\text{NO}_2^-$ ) to nitric oxide by nitrite reductases; third nitric oxide is converted to nitrous oxide by nitric oxide reductase and finally nitrous oxide is converted to nitrogen by nitrous oxide reductase. Majority of prokaryotes (bacteria) have capability to reduce nitrate. Nitrate reductase activity helps in understanding greenhouse gas fluxes from terrestrial ecosystems.

## 8.4 Enzymes Involved in Phosphorus Cycle

Phosphorus (P) is essential to living organisms and it is found in two forms, organic and inorganic. Organic P contributes to plant mineral nutrition only after its dephosphorylation to release inorganic phosphate, which is of particular importance in phosphate-deficient natural or cultivated ecosystems. The cycling of P in soil dependent on immobilization, mineralization, and redistribution of P. Phosphatases are a broad group of enzymes that catalyze the hydrolysis of esters and anhydrides of phosphoric acid and also serves as good indicators of soil fertility (Acosta-Martínez and Tabataba 2011). Judicious use of P helps in plant growth, development and enhance yield and soil health however, its deficiency causes stunted growth and excess causes environmental implications. The recycling of P forms in soil is mainly mediated by phosphatases (Fig. 8.1) and through addition of manures and fertilizers, into free phosphates ( $\text{PO}_4^{3-}$ ) that can be taken up by plants and soil microorganisms.

### 8.4.1 Phosphatases

Phosphatases catalyze the hydrolysis of both esters and anhydrides of phosphoric acid (Schmidt and Laskowski 1961). On the basis of number of ester bonds, these enzymes are classified into three types as phosphomonoesterases, phosphodiesterases, and phosphotriesterases (Acosta-Martínez and Tabataba 2011). Phosphatases are responsible for at least 50% of soils total P transformations. Phosphates are of different types such as acid and alkaline phosphomonoesterase, phosphoprotein phosphatases, phytases, nucleotidases, phosphodiesterases, phospholipases, and inorganic pyrophosphatase (Nannipieri et al. 2011). According to Burns et al. (2013) these enzymes are associated with active microbial cells, either intracellular or attached to the outer cell surface, extracellular, present in cell debris or dead cells or bacterial spores or entrapped in humic matter (Nannipieri et al. 2011). In soils, phosphatases mainly originate from soil microorganisms, however, they are also



present in the rhizosphere (area around the roots where highest microbial activity is found under the influence of root exudates) and detritosphere (this is the area around decomposed plant material in soil, where microbial community and organic carbon fractions are different than rhizosphere). Higher activities of both acid and alkaline phosphomonoesterase are found near the rhizoplane and it is depended on type of soil, age of plant, and plant species (Tarafdar and Jungk 1987; Tabatabai and Bremner 1969). Phosphatase activity increases with the increase in organic matter content however, it decreases with soil depth (Tabatabai and Dick 1979). The phosphatase activity in soil is affected by soil properties, temperature management practices, tillage, application of manures or sewage sludge and pollutants, addition of fresh litter, and seasonal variation in moisture.

## 8.5 Enzymes Involved in Sulfur Cycle

Sulfur (S) is an essential nutrient required by microbes and plants and its fate in the soil is mainly dependent on microbial activities. S is an important component of S-containing amino acids (methionine and cysteine), sulfated carbohydrates, vitamins (biotin and thiamine), alkaloids (alicyclic), and functional molecules (glutathione) (Tabatabai 1994; Dotaniya et al. 2019). In soil, sulfur is present in two forms organic and inorganic S. Organic sulfur account for 90–98% which exists as organic sulfate S and carbon-bonded S forms. In plant, sulfur is taken up as inorganic sulfate and its availability is dependent on either mineralization or mobilization from organic sulfur (Makoi and Ndakidemi 2008). These transformation reactions are mainly catalyzed by enzymes released into the soil environment by microorganisms, plant roots, and soil fauna (Klose et al. 2011). Some of the enzymes involved in S cycle are mentioned in Fig. 8.1.

### 8.5.1 *Arylsulfatases*

The soil enzymes which bring about the conversion of organic S to inorganic S into soil solution are known as sulfatases. These enzymes are responsible for the S ester hydrolysis in soil. The synthesis of sulfatases in soil is induced by bacterial population in S-limiting conditions (McGill and Colle 1981). The population of aryl sulfatase is affected by S concentration, crop growth stage and bacterial population in soil. The release of sulfate from soluble and insoluble sulfate esters in the soil is affected by different factors such as pH of soil, organic matter, concentration of sulfate esters, and heavy metal pollution.

## 8.6 Enzymes as Indicators of Overall Microbial Diversity

### 8.6.1 *Dehydrogenase*

Soil dehydrogenases belong to the class oxidoreductase enzymes (Gu et al. 2009). Dehydrogenase (DHA) is a sensitive indicator of overall microbial activity in the soil as it brings about oxidation- reduction reactions in living cells (Alef and Nannipieri 1995; Majchrzak et al. 2016). These enzymes transfer hydrogen from organic substrates to inorganic acceptors and causes biological oxidation of SOM (Zhang et al. 2010). DHA do not accumulate extracellular in the soil. The external factors either stimulate or inhibit dehydrogenase activity in the soil. The factors that stimulate dehydrogenase activity are soil temperature, moisture, pH, aeration, organic matter content and season of the year. The factors that inhibit dehydrogenase activity of the soil are soil profile depth, fertilization and pesticide amendments and, heavy metal presence (Wolińska and Stepniewska 2012).

### 8.6.2 *Fluorescein Diacetate Hydrolysis*

Hydrolysis of the fluorescein diacetate (3',6'-diacetylfluorescein) is used to estimate microbial activity in soil. FDA hydrolysis measures microbial decomposer activity and microbial decomposition and contribute to 90% of the energy flow in soil system therefore; it provides a good estimate of total microbial activity. This enzymatic reaction produces fluorescein, that can be quantified by spectrophotometry. FDA has been used to determine amounts of active bacteria, fungi, and acetylsterases in living cells (Schnürer and Rosswall 1982). FDA is hydrolyzed by three enzymes i.e., esterases, proteases, and lipases, which are involved in the microbial decomposition of organic matter in soil. FDA hydrolysis can be used efficiently to estimate microbial activity in soil having different types of cultural practices, organic and inorganic inputs and microbial inoculants (Sánchez-Monedero et al. 2008). Moreover, this enzyme helps in determining potential of the soil to support biochemical processes, which are essential for maintaining soil fertility as well as soil health (Patle et al. 2018).

### 8.6.3 *Catalase*

Catalase brings about conversion of hydrogen peroxide to water and oxygen. This redox enzyme system is very important for synthesis of soil humus and for preventing toxicity of hydrogen peroxide to soil enzymes.

## 8.7 Soil Enzymes in Ecosystem Functioning

### 8.7.1 *Soil Enzymes as Bioindicators*

A bioindicator is defined a microorganism, its part, its product (enzyme), collection of organisms, or biological process used to obtain information on the quality of all or part of the environment (Killham 2002). Soil enzyme activities provides an early indication of changes in soil quality and health due to any factors. In addition, soil enzyme activities can be used as a measure of soil productivity and fertility, an indicative of biological equilibrium in an ecosystem, to better understand changes in ecological processes within the soil ecosystem and an active indicator of soil pollution (Antonious 2003; Gunjal et al. 2019). These soil enzyme activities are influenced by both enviromental changes as well as soil fertility level. Soil organic carbon (SOC) is the key constituent of soil organic matter (SOM) and is considered as a good indicator of soil health. Soils with high SOM content are known to enhance water availability, aggregate formation, adequate aeration, improve porosity, water infiltration, deliver adequate quantity of nutrients to plants, improve soil fertility and enhance food production (Shah et al. 2020).

### 8.7.2 *Soil Enzymes in Functioning of Nutrient Cycling*

Soil enzymes regulate ecosystem functioning and in particular play a key role in nutrient cycling (Makoi and Ndakidemi 2008). Soil enzymes play a pivotal role in nutrients cycling as they perform several biochemical reactions that are necessary for the life processes of soil microbes and also maintain the structure of soil, formation and decomposition of organic matter and nutrient transformations (Joshi et al. 2018). Majority of soil process such as decomposition and transformations of soil organic matter, release of inorganic nutrients for plant growth; N<sub>2</sub> fixation; nitrification; denitrification; and detoxification of xenobiotics, bioremediation are performed by soil enzymes (Sherene 2017). Soil enzymes play major role in cycling of carbon ( $\beta$ -glucosidase, endoglucanase, cellobiohyrolase,  $\beta$ -galactosidase, endoxylasnase, endomannase,  $\beta$ -glycosidase, esterase, laccase, Mn peroxidise, lignin peroxidise, endochitinase, N-acetylglucosaminidase,  $\alpha$ -glucosidase), N (protease, peptidase, urease, N-acetylglucosaminidase), P (phosphomonoesterase, phosphodiesterase), and S (arylsulphatase) cycle (Kandeler et al. 2011; Baldrian 2009). In a study made by Ullah et al. (2019) an enhancement in enzyme activities involved in the C, N, and P cycling of was observed by the addition of N addition, showing that soil microbes produce more enzymes under high N conditions.

### 8.7.3 *Factors Affecting Soil Enzyme Activities*

Activity of enzymes in soil is dependent on several factors including soil properties (soil pH, OM, total nitrogen, phosphorus, sulfur, heavy metals), addition of inputs, climatic conditions, vegetation, cropping systems. Liu et al. (2021) studied soil enzyme activities in peatlands in permafrost regions and found that soil substrate, ammonical nitrogen, soil moisture content and nitrate nitrogen were the main factors affecting soil enzymes activities significantly. The addition of nitrogen in soil enhances activities of glycosidases ( $\beta$ -cellobiosidase,  $\beta$ -glucosidase,  $\beta$ -xylosidase, and  $\alpha$ -glucosidase) and which help in the breakdown of cellulose, chitin, carbohydrates, and in N mineralization (Jian et al. 2016; Saiya-Cork et al. 2002). The addition of N resulted in increase in soil acidity and SOC which significantly affected C-cycling enzymes. Song et al. (2019) showed that total organic carbon, total nitrogen and dissolved organic carbon in soil are correlated with  $\beta$ -glucosidase, acid phosphatase, invertase, and urease activities. Roberto et al. (2009) determined the geometric mean enzyme activity to study the effect of organic and conventionally managed plots on enzyme activities and nematode population. Higher nematode population and enzyme activities were observed in organically managed plots. Urease activity was higher in soils under vegetation than vegetation free soils (Reddy et al. 1987; Gupta and Bhardwaj 1990). Similarly soil enzymes like invertase, soil phosphatase serves as an important indicator of stress in the soil (Liu et al. 2021). Using organic fertilizers like compost, straw mulch and sewage sludge increases and soil tillage decreases the activity of urease. In addition, application of wintery wastewater in four different vineyard soils increased soil urease activity more than municipal water. Elbl et al. (2019) applied organic amendments (compost and vermicompost) in the soil which resulted in higher microbial biomass, DHA, FDA, and phosphatase activities as compared to inorganic fertilizers. Vegetation degradation is a change in the structure of the vegetation community, plays an essential role in changes in soil nutrient and enzyme activities. Greatest variation in soil nutrients and enzyme activities were observed on surface and deeper layers of soil. Vegetation degradation also resulted in reduction in soil carbon storage and nutrient cycling capacity. Wang et al. (2020) studied the variations of soil organic carbon components and enzyme activities (catalase, sucrase, urease, and amylase) in four vegetation types and reported that sucrase and urease activities were significantly correlated with soil organic and particulate organic carbon content and microbial biomass carbon significantly affected catalase activity. Mangalassery et al. (2015) determined the potential of zero tillage in microbial community functioning as reflected by reduced respiration rates and greater enzyme activities. The soil under zero tillage management accumulated greater amounts of total and aromatic carbon. Baoyi et al. (2014) showed that straw returning and deep tillage increased soil microbial and enzyme activities (catalase, phosphatase, saccharase, urease). In this way, soil enzymes activities respond differently to different inputs, management practices and environmental conditions.

## 8.8 Conclusion

Soil enzymatic activity serves as a critical indicator of soil fertility, quality, and health because it plays key biochemical functions in the overall process of organic matter decomposition, nutrients cycling, mineralization, detoxification of toxicants, and suppression of noxious and pathogenic organisms. Measuring enzymatic catalysis and understanding the factors that regulate enzyme expression significantly contributes to assess the effect of agricultural management practices in improving soil quality and fertility. Thus, determining a suite of enzyme activities in soil amalgamate both the intra and extracellular enzymatic transformations in the soil biological system and also serves as the main feature for soil quality assessment.

## References

- Abbott KC, Karst J, Biederman LA, Borrett SR, Hastings A, Walsh V, Bever JD (2015) Spatial heterogeneity in soil microbes alters outcomes of plant competition. *PLoS One* 10(5):10–25
- Acosta-Martínez V, Tabataba MA (2011) Phosphorus cycle enzymes. In: Dick RP (ed) *Methods of soil enzymology*. SSSA Book Soil Science Society of America, Madison, pp 161–183
- Alef K, Nannipieri P (1995) *Methods in applied soil microbiology and biochemistry*. Academic, London
- Alkorta I, Aizpuru A, Riga P, Albizu I, Amezaga I, Garbisu C (2003) Soil enzyme activities as biological indicators of soil health. *Rev Environ Health* 18(1):65–73
- Antonious GF (2003) Impact of soil management and two botanical insecticides on urease and invertase activity. *J Environ Sci Health Part B* 38(4):479–488
- Baldrian P (2006) Fungallaccases: occurrence and properties. *FEMS Microbiol Rev* 30:215–242
- Baldrian P (2009) Microbial enzyme-catalyzed processes in soils and their analysis. *Plant Soil Environ* 55(9):370–378
- Bandick AK, Dick RP (1999) Field management effects on soil enzyme activities. *Soil Biol Biochem* 31:1471–1479
- Baoyi J, Hao H, Zhao Y, Xinyuan M, Liu K, Li C (2014) Effects of deep tillage and straw returning on soil microorganism and enzyme activities. *Sci World J* 2014:451493
- Botha AM, Nagel MAC, Westhuizen V, Botha FC (1998) Chitinase isoenzymes in near-isogenic wheat lines challenged with Russian wheat aphid, exogenous ethylene, and mechanical wounding. *Bot Bull Acad Sin* 39:99–106
- Burns RG, DeForest JL, Marxsen J, Sinsabaugh RL, Stromberger ME, Wallenstein MD, Weintraub MN, Zoppini A (2013) Soil enzymes in a changing environment: Current knowledge and future directions. *Soil Biol Biochem* 58:216–234
- Caldwell BA (2005) Enzyme activities as a component of soil biodiversity: a review. *Pedobiologia* 49:637–644
- Chebrou H, Bigey F, Arnaud A, Galzy P (1996) Study of the amidase signature group. *Biochim Biophys Acta* 1298:285–293
- Claus H, Filip Z (1988) Behavior of phenoloxidases in the presence of clays and other soil-related adsorbents. *Appl Microbiol Biotechnol* 28:506–511
- Collinge DB, Kragh KM, Mikkelsen JD, Nielsen KK, Rasmussen U, Vad K (1993) Plant chitinases. *Plant J* 3(1):31–40. <https://doi.org/10.1046/j.1365-3113x.1993.t01-1-00999.x>
- Dale VH, Beyeler SC (2001) Challenges in the development and use of ecological indicators. *Ecol Indic* 1:3–10

- Dedeken M, Voets JP (1965) Research on the metabolism of amino acids in the soil. I. The metabolism of glycine, alanine, aspartic acid and glutamic acid. *Ann Inst Pasteur (Paris)* 109(3):103–111
- Dotaniya ML, Aparna K, Dotaniya CK, Singh M, Regar KL (2019) Role of soil enzymes in sustainable crop production. *Enzymes Food Biotechnol* 2019:569–589
- Elbl J, Makova J, Javorekova S, Medo J, Kint A, Losak T, Lukas V (2019) Response of microbial activities in soil to various organic and mineral amendments as an indicator of soil quality. *Agronomy* 9:485. <https://doi.org/10.3390/agronomy9090485>
- Esen A (1993)  $\beta$ -Glucosidases. In: Esen A (ed) *B-Glucosidases: biochemical and molecular biology*. American Chemical Society, Washington, DC, pp 1–14
- Fournand D, Arnaud A (2001) Aliphatic and enantio selective amidases: from hydrolysis to acyl transfer activity. *J Appl Microbiol* 91:381–393
- Gelsomino A, Badalucco L, Landi LCG (2006) Soil carbon, nitrogen and phosphorus dynamics as affected by solarization alone or combined with organic amendment. *Plant Soil* 279:307–325
- Gil-Sotres F, Trasar-Cepeda C, Leirós M, Seoane S (2005) Different approaches to evaluating soil quality using biochemical properties. *Soil Biol Biochem* 37:877–887
- Glick BR (2010) Using soil bacteria to facilitate phytoremediation. *Biotechnol Adv* 28(3):367–374
- Gomez L, Allona I, Casado R, Aragoncillo C (2002) Seed chitinases. *Seed Sci Res* 12:217–230
- Gu Y, Wag P, Kong C (2009) Urease, invertase, dehydrogenase and polyphenoloxidase activities in paddy soils influenced by allelopathic rice variety. *Eur J Soil Biol* 45:436–441
- Gunjal AB, Waghmode MS, Patil NN, Nawani NN (2019) Significance of soil enzymes in agriculture. In: *Smart bioremediation technologies*. Springer, New York, pp 159–168
- Gupta RD, Bhardwaj KKR (1990) Phosphatase and urease enzymatic activities in some soil profiles of north west Himalayas. *J Indian Soc Soil Sci* 38(4):756–759
- Hatakka A (1994) Lignin-modifying enzymes from selected white-rot fungi: production and role in lignin degradation. *FEMS Microbiol Rev* 13:125–135
- Jian S, Li J, Chen J, Wang G, Mayes MA, Dzantor KE (2016) Soil extracellular enzyme activities, soil carbon and nitrogen storage under nitrogen fertilization: A meta-analysis. *Soil Biol Biochem* 101:32–43
- Joshi S, Mohapatra B, Mishra JPN (2018) Microbial soil enzymes: implications in the maintenance of rhizosphere ecosystem and soil health. In: Adhya TK et al (eds) *Advances in soil microbiology: recent trends and future prospects, microorganisms for sustainability*. Springer, Singapore
- Kandeler E, Poll C, Frankenberger WT, Tabatabai MA (2011) Nitrogen cycle enzymes. In: Dick RP (ed) *Methods of soil enzymology*. SSSA Book Series, Madison, pp 211–245
- Killham K (2002) Bioindicators and sensors of soil health and the application of geostatistics. In: Burns RG, Dick RP (eds) *Enzymes in the environment: activity, ecology and applications*. Marcel Dekker, Inc., New York
- Klose S, Bilen S, Tabatabai MA, Dick WA (2011) Sulfur cycle enzymes methods of soil enzymology. SSSA Book Series, New York
- Liu C, Song Y, Dong X, Wang X, Ma X, Zhao G, Zang S (2021) Soil enzyme activities and their relationships with soil C, N, and P in peatlands from different types of permafrost regions northeast china. *Front Environ Sci* 9:670769. <https://doi.org/10.3389/fenvs.2021.670769>
- Lynd LR, van Zyl WH, McBride JE, Laser M (2005) Consolidated bioprocessing of cellulosic biomass: an update. *Curr Opin Biotechnol* 16:577–583
- Majchrzak L, Sawinska Z, Natywa M, Skrzypczak G, Wołoszyn R (2016) Impact of different tillage systems on soil dehydrogenase activity and spring wheat infection. *J Agric Sci Technol* 18: 1871–1881
- Makoi JHJR, Ndadikemi PA (2008) Selected soil enzymes: examples of their potential roles in the ecosystem. *Afr J Biotechnol* 7(3):181–191
- Mangalassery S, Mooney SJ, Sparkes DL, Fraser WT, Sjögersten S (2015) Impacts of zero tillage on soil enzyme activities, microbial characteristics and organic matter functional chemistry in temperate soils. *Eur J Soil Biol* 68:9–17

- Merino C, Godoy R, Matus F (2016) Soil enzymes and biological activity at different levels of organic matter stability. *J Soil Sci Plant Nutr* 16:14–30
- McGill WB, Colle CV (1981) Comparative aspects of cycling of organic C, N, S and P through soil organic matter. *Geoderma* 26:267–286
- Moghimian N, Hosseini SM, Kooch Y, Darki BZ (2017) Impacts of changes in land use/cover on soil microbial and enzyme activity. *Catena* 157:407–414
- Murphy CD (2016) Microbial degradation of fluorinated drugs: biochemical pathways, impacts on the environment and potential applications. *Appl Microbiol Biotechnol* 100(6):2617–2627
- Nannipieri P, Giagnoni L, Landi L, Renella G (2011) Role of phosphatase enzymes in soil. In: Beunemann EK et al (eds) *Phosphorus in action*, vol 26. Springer, Berlin. [https://doi.org/10.1007/978-3-642-15271-9\\_9](https://doi.org/10.1007/978-3-642-15271-9_9)
- Pace HC, Brenner C (2001) The nitrilase superfamily: classification, structure and function. *Genome Biol* 2(1):REVIEWS0001
- Pancholy SK, Rice EL (1973) Soil enzymes in relation to old field succession: amylase, cellulase, invertase, dehydrogenase and urease. *Soil Sci Soc Am Proc* 37:47–50
- Patle PN, Navnage NP, Barange PK (2018) Fluorescein diacetate (FDA): measure of total microbial activity and as indicator of soil quality. *J Curr Microbiol Appl Sci* 7(6):2103–2107. <https://doi.org/10.20546/ijcmas.2018.706.249>
- Pertsovich SI, Guranda DT, Podchernyaev DA, Yanenko AS, Svedas VK (2005) Aliphatic amidase from *Rhodococcus rhodochrous* M8 is related to the nitrilase/cyanide hydratase family. *Biochemistry* 70:1280–1287
- Prober SM, Leff JW, Bates ST, Borer ET, Firn J, Harpole WS, Lind EM, Seabloom EW, Adler PB, Bakker JD (2015) Plant diversity predicts beta but not alpha diversity of soil microbes across grasslands worldwide. *Ecol Lett* 18:85–95
- Rajkumar M, Prasad MNV, Swaminathan S, Freitas H (2013) Climate change driven plant–metal–microbe interactions. *Environ Int* 53:74–86
- Raju MN, Golla N, Vengatampalli R (2017) *Soil protease*. Springer, Cham
- Reddy GB, Faza A, Bennett R Jr (1987) Activity of enzymes in rhizosphere and nonrhizosphere soils amended with sludge. *Soil Biol Biochem* 19(2):203–205
- Roberto G-R, Victoria O, Benjamín V, Belen HM, Reyes P-S, Liébanas G, Juan L, José C (2009) Soil enzymes, nematode community and selected physico-chemical properties as soil quality indicators in organic and conventional olive oil farming: influence of seasonality and site features. *Appl Soil Ecol* 41:305–314
- Ross DJ (1975) Studies on a climosequence of soils in tussock grasslands-5: invertase and amylase activities of top soils and their relationships with other properties. *N Z J Sci* 18:511–518
- Sahai AS, Manocha MS (1993) Chitinases of fungi and plants: their involvement in morphogenesis and host-parasite interaction. *FEMS Microbiol Rev* 11:317–338
- Saiya-Cork K, Sinsabaugh R, Zak D (2002) The effects of long term nitrogen deposition on extracellular enzyme activity in an *Acer saccharum* forest soil. *Soil Biol Biochem* 34(9):1309–1315
- Sánchez-Monedero MA, Mondini C, Cayuela ML, Roig A, Contin M, De Nobili M (2008) Fluorescein diacetate hydrolysis, respiration and microbial biomass in freshly amended soils. *Biol Fertil Soils* 44:885–890. <https://doi.org/10.1007/s00374-007-0263-1>
- Schmidt G, Laskowski MSK (1961) *Phosphate cstr cleavage (survey)*, 2nd edn. Academic, New York
- Schnürer J, Rosswall T (1982) Fluorescein diacetate hydrolysis as a measure of total microbial activity in soil and litter. *Appl Environ Microbiol* 43:1256–1261
- Shah T, Lateef S, Noor MA (2020) Carbon and nitrogen cycling in agroecosystems: an overview. In: Datta R et al (eds) *Carbon and nitrogen cycling in soil*. Springer, Singapore
- Sherene T (2017) Role of soil enzymes in nutrient transformation: a review. *Biol Bull* 3(1):109–131
- Singh M, Meena SC, Jain RK (2016) Application of chitinase for sustainable agriculture. *Adv Life Sci* 5(24):1131–1138

- Song Y, Song C, Shi F, Wang M, Ren J, Wang X (2019) Linking plant community composition with the soil C pool, N availability and enzyme activity in boreal Peatlands of Northeast China. *Appl Soil Ecol* 140:144–154. <https://doi.org/10.1016/j.apsoil.2019.04.019>
- Strong PJ, Claus H (2011) Laccase: a review of its past and its future in bioremediation. *Crit Rev Environ Sci Technol* 41(4):373–434. <https://doi.org/10.1080/10643380902945706>
- Suarez V, Staehelin C, Arango R, Holtorf H, Hofsteenge J, Meins F Jr (2001) Substrate specificity and antifungal activity of recombinant tobacco class I chitinases. *Plant Mol Biol* 45:609–618
- Tabatabai MA (1994) Soil enzymes. In: Weaver RW et al (eds) *Methods of soil analysis, part 2. Microbiological and biochemical properties*. Springer, New York, pp 775–833
- Tabatabai MA, Bremner JM (1969) Use of p-nitrophenyl phosphate for assay of soil phosphatase activity. *Soil Biol Biochem* 1:301–307
- Tabatabai MA, Dick WA (1979) Distribution and stability of pyrophosphatase in soils. *Soil Biol Biochem* 11:655–659
- Tarafdar JC, Jungk A (1987) Phosphatase activity in the rhizosphere and its relation to the depletion of soil organic phosphorus. *Biol Fertil Soils* 3:199–204
- Thoma JA, Spradlin JE, Dygerd S (1971) Plant and animal amylases. Academic, New York
- Thurston CF (1994) The structure and function of fungal laccases. *Microbiology* 140:19–26
- Ullah S, Ai C, Huang S, Zhang J, Jia L, Ma J, Zhou W, He P (2019) The responses of extracellular enzyme activities and microbial community composition under nitrogen addition in an upland soil. *PLoS ONE* 14(9):e0223026
- Wang SL, Wang CY, Huang TY (2008) Microbial reclamation of squid pen for the production of a novel extracellular serine protease by *Lactobacillus paracaseisubsp. paracasei* TKU012. *Bioresour Technol* 99:3411–3417
- Wang H, Wu J, Li G, Yan L (2020) Changes in soil carbon fractions and enzyme activities under different vegetation types of the northern Loess Plateau. *Ecol Evol* 10:12211–12223
- Wolińska A, Stepniewska Z (2012) Dehydrogenase activity in the soil environment. In: *Dehydrogenases*. Intech Open, London
- Yang ST, El-Enshasy HA, Thongchul N (2013) Cellulases: characteristics, sources, production, and applications. In: Zhang X-Z, Yi-Heng PZ (eds) *Bioprocessing technologies in biorefinery for sustainable production of fuels, chemicals, and polymers*, 1st edn. Wiley, Hoboken, pp 131–146
- Zhang NH, Xing-Dong G, Yu-Bao LI, Yong-Hong W, Hai-Tao MA, Di ZR, Yang S (2010) Pedogenic carbonate and soil dehydrogenase activity in response to soil organic matter in *Artemisia Ordosica* community. *Pedosphere* 20:229–235
- Zhang T, Wan S, Kang Y, Feng H (2014) Urease activity and its relationships to soil physiochemical properties in a highly saline-sodic soil. *J Soil Sci Plant Nutr* 14(2):304–315