

# Chapter 18

## Impact of Climate Change on the Importance of Plant Growth-Promoting Microbes in the Rhizosphere



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**Abstract** The global climatic changes have made significant alterations in the environment that can be harmful for microbes, plants, animals, and humans. Climatic parameters include the temperature, atmospheric gases, carbon dioxide levels, etc. that have been known to pose harmful effects to microbes when present in excess limits. Among microbes, plant growth-promoting rhizobacteria and arbuscular mycorrhizal fungi have been shown to be affected by climate fluctuations. Hence, this book chapter will discuss the impact of various climate parameters on microbial communities and the microbial processes going on in the soil with special emphasis on plant growth-promoting rhizospheric bacteria and arbuscular mycorrhizal fungi.

**Keywords** Climate change · Plant growth-promoting bacteria · Arbuscular mycorrhizal fungi · Temperature · Carbon dioxide

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## 18.1 Introduction

Till the following century, it is said that the global climate will immensely change and will affect various parameters about this alteration in environment (Houghton et al. 2001), leading to continuous increase of atmospheric CO<sub>2</sub> concentration (IPCC Climate Change 2007), along with the prediction of global surface increase in changing temperature between 1.8 and 3.6 °C until 2100, caused due to human and natural activities to increase atmospheric CO<sub>2</sub> level (IPCC Climate Change 2007). It is expected for the soil water content to decrease due to the increase in temperature (Le Houerou 1996), causing drought to various parts of the world. Terrestrial microorganism will be affected by these climate changes. Along with affecting ecosystem and organisms as well as per the recent studies, it has been seen that there is well-established symbiosis of plant taxa with various types of microorganisms (Nicolson 1967). As characterized by De Bary definition, they can be neutral or pathogenic toward their host (de Bary 1879); additionally they increase plant tolerance toward abiotic and biotic stress and support plant (Bent 2006). The rhizosphere is colonized by plant growth-promoting microorganisms. The part of the soil that is attached to the surface of root and affected by exudes of root and via microorganism (Bent 2006). Microorganisms benefit their host by endophytic life-style by entering in the root system (Stone et al. 2000). This can be referred to as the plant growth-promoting rhizobacteria (PGPR, Kloepper and Schroth 1978) or plant growth-promoting bacteria (PGPB, Bashan and Holguin 1998). Changes in the plant physiology and root exudation are to be likely seen due to the climate change. The increase in carbon (C) allocation in the root zone with the increase level of CO<sub>2</sub> will lead to root exudates composition change. These changes can be chemoattractant or signaling compound along with various carbon and nitrogen ratio or nutritional presence (Kandeler 2007). The plant-associated microbial activities and diversity will be affected by the climate change (Drigo et al. 2008). Beneficial effect of microorganisms on growth of plant or its overall health might be weakened in reference to possessing their required property and their ability to colonize under specific situations. A definite comprehension on how precisely microbes associated with plants are impacted is either influenced directly by modified plant anatomy, or the way it influences plant performance and the environment functioning is yet missing. Mycorrhizal growth and endophytes just as PGPB are used as biocontrol specialists, biofertilizers, and additionally phytostimulators used in agricultural sector (Vessey 2003; Welbaum et al. 2004) or on the other hand used in phytoremediation applications as degrading microorganisms (Denton 2007). The immense effect of global climate change may alter the performance; thus appropriate study is required to choose the strains that will perform well in changed conditions. Here we will discuss about major environment parameter that are likely to be altered via climate change and impact the microbes and related processes.

## 18.2 Influence of Climate Change on Soil Microbial Process

Relative abundance and capacity of soil communities is adjusted by growing change in climate since soil network individuals contrast in their physiology, temperature, affectability, and development rates (Castro et al. 2010, 2012; Gray et al. 2011; Lennon et al. 2012; Briones et al. 2014; Delgado-Baquerizo et al. 2014; Whitaker et al. 2014). There has been wide range of exploring due to sudden impacts of climatic changes occurring on composition of microbial composition and its functioning (Blankinship et al. 2011; Manzoni et al. 2012; Chen et al. 2014). In a temperate forest, warming by 5 °C, for instance, expanded the bacterial to fungal proportion of the soil community (DeAngelis et al. 2015).

Changes in ecosystem functioning can be led by the shifts in microbial community when soil microbes contrast in their functional characteristics or controlling a rate-limiting or fate-controlling step (Schimel and Schaeffer 2012). Nitrification and nitrogen fixation are regulated via specific microbial groups (Isobe et al. 2011), denitrification, and methanogenesis (Bodelier et al. 2000). Alterations in the number of organisms who regulate specific process can directly affect the pace of that process. Nonetheless, a few processes are firmly connected to abiotic factors such as nitrogen mineralization, temp, and dampness than microbial community composition in the soil that a variety of life forms are in charge of these processes (Isobe et al. 2011).

The temperature-sensitive process such as microbial soil respiration rates are likely to be altered by warming effect of climate change. The extensive recent consideration is on the function of raised temperature in microbial metabolism (Compant et al. 2010). The natural temperature affectability of microbial action is characterized as the factor by which microbial activity increments with a 10 °C expansion in temperature (known as Q10). Q10 is a frequently used model in climate change to represent sensitive nature of microbes to temperature; in any case, through this a large number of associations that affect the temperature dependence of microbes in processes such as decomposition can be assessed.

Along these lines, temperature sensitivity represented via Q10 in models may give a bad representation of forecasts. During the field study, the process like decaying of soil organic matter, soil respiration, and development of microbial biomass with increase of temperature in reaction to warming are usually short-lived. The short-lived impacts have been speculated to happen as carbon substrates present in soil are expanded by microbial action and due to exchange as the soil microbial diversities display shift in composition or compel their biomass to react to adjusted conditions and substrate availability (Allison and Martiny 2008, Bradford 2013). Alteration of the composition of microbial communities can be initiated by experimental warming and can also affect the population of both gram-positive and gram-negative bacteria; soil dampness is linked to temperature change which may give clarification to contrasting outcomes from tests evaluating microbial community reactions to climate alterations. For instance, microbial activity at hotter temperatures can be restricted by dissemination and its contact with substrate (Zak et al.

1999). While bacterial community may react quickly to moisture pulse, the fungal counterpart may slow in their reaction (Bell et al. 2008; Kardol et al. 2010). Further, drought intensifies the differential temperature affectability of fungal and bacterial gatherings (Briones et al. 2014). With change in soil moisture availability (<30% decrease in water holding limit), soil fungal communities may shift starting with one dominant member onto the next, while bacterial communities stay consistent. Soil microbial communities acclimated to less water access may inspire to a lesser extent a compositional or functional move to changing water systems. Communications among organisms and background temperature and dampness systems in some random area impact microbial composition and function with evolving climate. Be that as it may, it is yet not satisfactory that (1) how temperature and sogginess and their connection impact unequivocal microbial network, for instance, methanogens inside a community, (2) what impacts microbial network changes that work like rot of new and old soil organic matter, and (3) which system drive the net climate reaction of microbial activities to the ecological change.

### ***18.2.1 Effect of Climate Change on PGPR***

Bacteria are also known to colonize the plants other than fungi (Compant et al. 2010) and possess positive effects on their host which can in turn act as beneficiary for them (Welbaum et al. 2004; Lugtenberg and Kamilova 2009). Microscopic organisms improve plant development and health by different components which incorporate plant growth substances, for example, hormones, mitigation of biotic and abiotic stresses, pathogen antagonism, and induction of systemic responses (Lugtenberg and Kamilova 2009). Even in the adverse conditions like high level of heavy metal in plants, bacterial endophytes may grow (Lodewyckx et al. 2002; Idris et al. 2004). Through specific plant genes under stress conditions, they alleviate abiotic and biotic stress (Hallmann 2001).

### ***18.2.2 Effects of Elevated CO<sub>2</sub> Levels***

Bacteria-associated plants can be affected by elevated CO<sub>2</sub> level. Drigo et al. (2008) reviewed the repercussions of increased CO<sub>2</sub> on various communities of rhizobacteria. Along with rhizospheric bacteria, endophytic populations may also be affected. The “Free-Air CO<sub>2</sub> Enrichment (FACE) experiment” held in Switzerland showed the effect of increased ambient CO<sub>2</sub> (Hebeisen et al. 1997). This experiment involves interaction of legumes with rhizobia that was triggered by elevated CO<sub>2</sub> (Marilley et al. 1999); they are known for their interaction with legumes for nitrogen fixation in addition to their plant growth-promoting activities (Sessitsch et al. 2002). It also depicted that *leguminosarum* strains were favored for the enrichment of CO<sub>2</sub> over the other plants (Montealegre et al. 2000). 17% more

nodulation on the roots were produced by the isolates acclimated to elevated CO<sub>2</sub> than isolates favored by ambient CO<sub>2</sub> levels, telling that under various conditions, plants prefer the selection of specific microorganism (Haase et al. 2007). Nitrogen deficiency symptoms were found under the elevated CO<sub>2</sub> conditions in which N tissue decreased in bean. The reason behind lower N nutritional microbial growth under elevated atmospheric CO<sub>2</sub> concentration is relation between rhizosphere microbial growths is due to increase in competition of plant microbial nitrogen in the rhizosphere (Haase et al. 2007). This experiment provided a lot of information about PGPB such as *Pseudomonas* sp. and even on rhizobia (Marilley et al. 1999), *Actinobacteria*, and *Deltaproteobacteria*. Drigo et al. (2009) gave information on soil-specific effect of elevated CO<sub>2</sub> on *Pseudomonas* and *Burkholderia* sp. and on soil-particular effects on *Pseudomonas* and *Burkholderia* sp. and on the genes producing antibiotics. Elevated CO<sub>2</sub> has different plant responses which led to regulated photosynthate distribution in the rhizosphere. But some strong antibiotic producers such as *Actinomycetes* and *Bacillus* sp. are not influenced. Selective bacterial species responded differently to plants having association with enrichment to atmospheric CO<sub>2</sub> reported by Drigo et al. (2009). The dominance of *Pseudomonas* sp. was increased in presence of elevated CO<sub>2</sub> which includes a variety of plant growth-promoting members, associated with rye and white clover and enhancement of the abundance of *Rhizobium* sp. (Marilley et al. 1999).

Additional data of the relative amount of HCN-delivering *Pseudomonas* strains which are known as likely inhibitors of fungi taken from mass and rhizospheric soil alongside two perpetual perennial grassland root fraction systems (*L. perenne* and *Medicago coerulea*) were diminished in raised CO<sub>2</sub> conditions (Tarnawski and Aragno 2006). There was expanded extent of siderophores and nitrate dissimilating strains. It expresses a model that expanded atmospherically CO<sub>2</sub> levels may affect plant beneficial microbes, which may later have ramifications on their use in agricultural or phytoremediation processes (Fig. 18.1) (Baon et al. 1994; Sanders et al. 1998).

The investigation of practically identical detail on CO<sub>2</sub> restraint of bacterial development is that of King and Nagel. *Pseudomonas aeruginosa* was grown on solitary suboptimum temperature (24 °C) on minimal media and reasoned that inhibition differed directly with CO<sub>2</sub> concentration. A comparative relationship was seen with *P. fluorescens* developing at 30 °C at a restricted range of CO<sub>2</sub> concentration; significant deviation was observed. Low CO<sub>2</sub> fixations created marked hindrance of development in complex medium, yet stimulated growth in least amount of medium, and in complex medium, a most extreme level of restraint was achieved at generally low CO<sub>2</sub> concentration. It is conceivable that a more detailed examination of *P. aeruginosa* would uncover a similar reaction to CO<sub>2</sub>.

It has been expected that this perception is relevant to microorganisms; however, plainly with *P. fluorescens*, there is an immediate improvement of CO<sub>2</sub> inhibition with diminishing temperature. The nonlinearity of the connection among temperature and the level of inhibition with minimal medium most likely emerge due to the competition caused due to inhibitory and stimulant impact of CO<sub>2</sub> during development in this medium. Since the inhibitory effect of CO<sub>2</sub> can fluctuate with

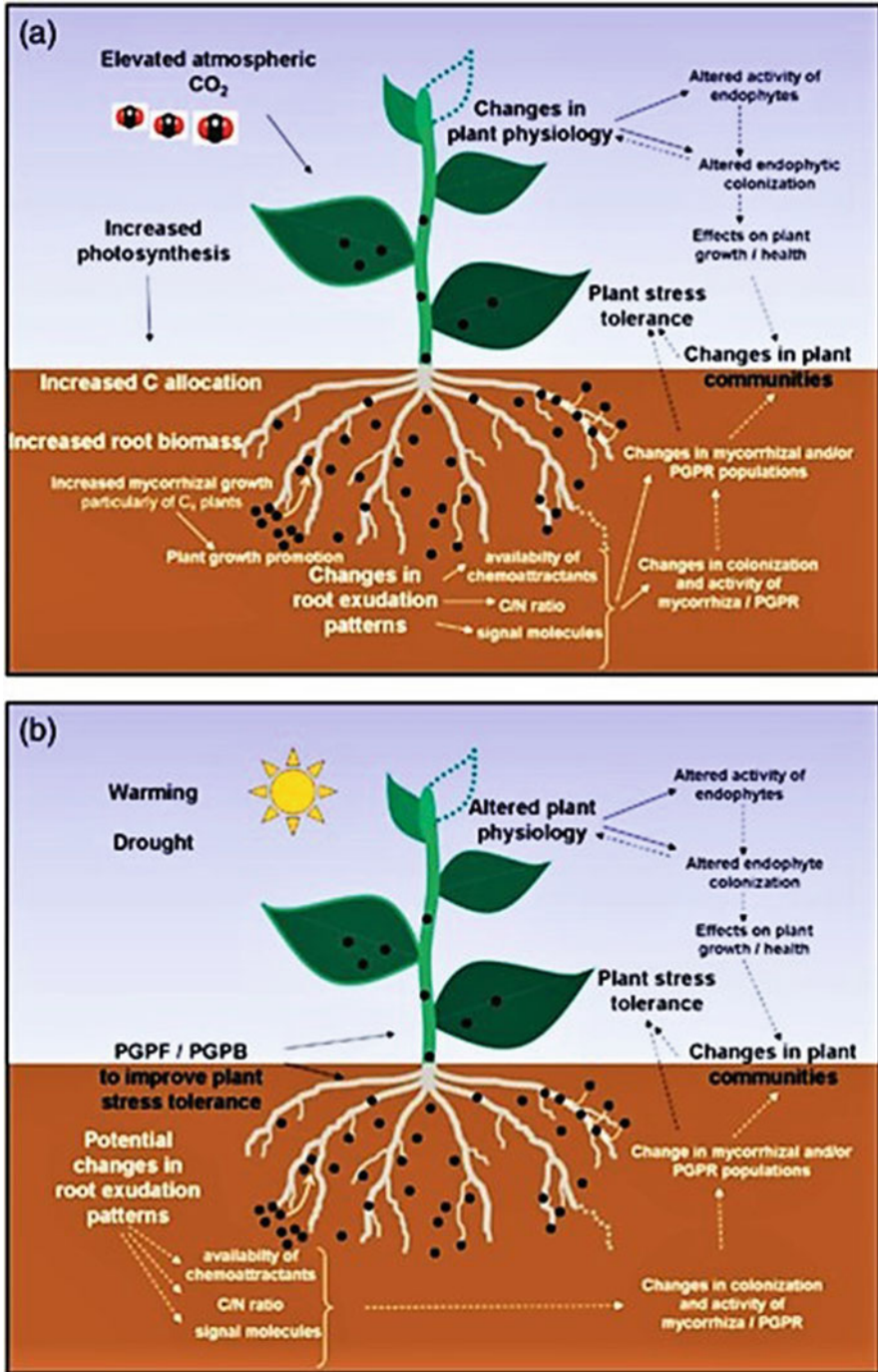
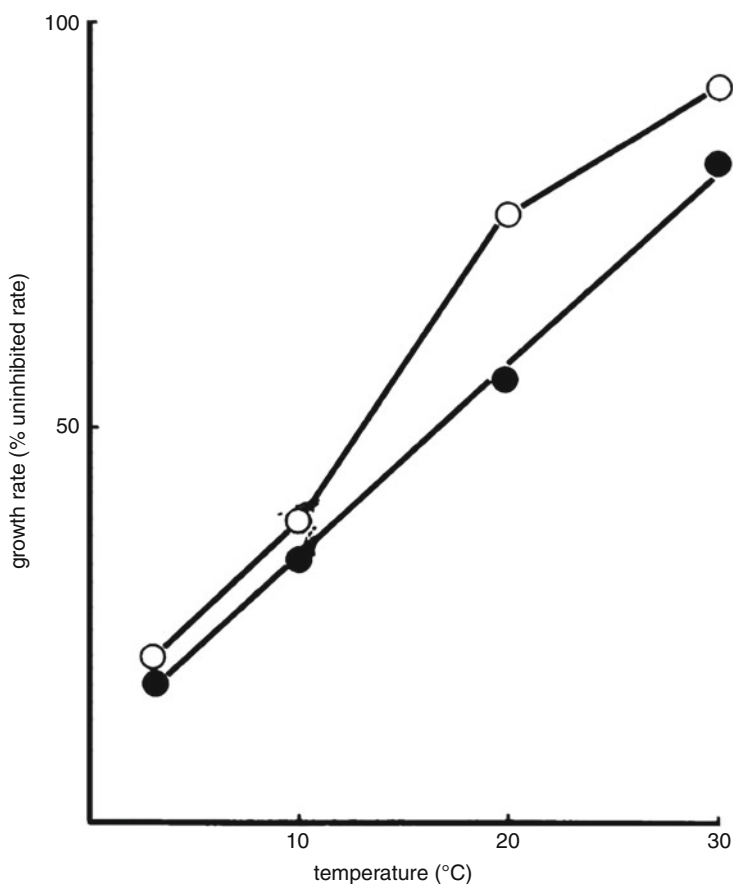


Fig. 18.1 Effects of (a) elevated CO<sub>2</sub> concentrations and (b) warming and drought on beneficial plant-microbe interactions (Adapted with permission from Compant et al. 2010)

temperature and medium composition, it is clearly not legitimate to look at results from changed sources except if these variables are considered. King and Nagel could not recognize any change in enzyme levels between cells of *P. aeruginosa* cultured in air and inside seeing CO<sub>2</sub> and stated that adaptive enzyme synthesis in the presence of CO<sub>2</sub> does not happen.

### 18.2.3 Effect of Temperature

Alleviation of temperature stress is depicted by rhizosphere bacteria and endophytes on some plants, and they may cause induction in growth of various crops under altering temperature, climates, and soil conditions (Fig. 18.2) (Bilal et al. 1993;



**Fig. 18.2** Effect of temperature on the growth rate of *P. fluorescens* growing in simple (O) or complex medium with pCO<sub>2</sub> in solution at 150 mm of Hg (Adapted with permission from Gill and Tan 1979)



Bashan and Holguin 1998). Egamberdiyeva and Höflich (2003) postulated that performance of plant beneficial bacteria may be affected by temperature and soil type. *Mycobacterium* sp. 44, *Pseudomonas fluorescens*, and *Pantoea agglomerans* strain which were isolated from a semi-continental climate showed elevation growth properties of root and shoot at 16 °C in comparison to growth at 26 °C in loamy sand. But in any case, *M. phlei* strain MbP18 delivered good results under the two conditions just like *M. bullata* MpB46, showing genotypic-explicit inclinations for some natural conditions. A few strains of PGPB growing at a hot condition than at cold ones can be used in agriculture. In an assessment, it was evidently shown that rhizobia segregated from the desert wood vegetable *Prosopis glandulosa* nodules were grown more noteworthy at 36 °C than at 26 °C. Moreover, microbes colonizing specific destination may react contrastingly to some biological conditions. An assessment with the endophytic microbe *Burkholderia phytofirmans* strain PsJN displayed that increasing the temperature from 10 to 30 °C diminished its establishment in the tomato rhizosphere; however endophytic abundance wasn't impacted (Pillay and Nowak 1997). When successfully colonized in rhizosphere, it may help prevent the temperature or water stress on plants (Aroca and Ruiz-Lozano 2009) by inducing a systemic response (Yang et al. 2009). This shows the expected part of specific strains for their utilization in agriculture; however there is need to consider the impacts of altering conditions on their establishment just as on their plant growth-promoting capacity.

### 18.3 Influence of Climate Change on Arbuscular Mycorrhizal Fungi

Mycorrhizal fungi are known as one of the crucial factors while considering global changes related to variations in the ecosystem, as these fungi are known to play a major role in the plant-soil interface. Global changes encircle natural as well as human-led changes on the Earth's environment. This section will focus mainly on anthropogenic aspects resulting in global changes related to mycorrhiza. While considering climate change, media heeds about global warming mainly, among other factors. But now people perceive that anthropogenic factors not only influence climate change, but they also steer toward many global changes (Vitousek 1994).

Human-induced changes have caused serious repercussions in the Earth's environment other than climate change. Some important examples are increased concentration of the atmospheric gases (ozone, CO<sub>2</sub>, etc.), the ecosystem being exposed to a rising number of nutrients due to atmospheric deposition (Nitrogen), change in climate (altered precipitation as well as temperature regimes), increase in UV radiations, and occurrence of non-native species. These comprise present or upcoming global alterations, displaying good or bad impact on mycorrhizae associations. These aforementioned factors are distinct, based on the fact that mycorrhizal symbiosis involves two classes of organisms. Some factors have direct impact on the



host plant solely (example carbon fixation), and some have an indirect influence on the mycorrhizal fungi (mycobionts), like change in carbon distribution by plant (atmospheric causes can affect the presence of nutrients in the soil) in turn affecting the fungi as well. Warming or an altered precipitation is included in factors that directly affect mycobionts. Understanding this difference is substantial in figuring out the mechanism behind the effects of global change factors and also for experimental proposals. An isolated environment can rarely be subjected to factors responsible for global change. We need to figure out the mechanism behind these factors in order to understand the complexity of the regional combinations of such factors, as experimental exploration of all possible scenarios will be nearly impossible in artificial or mimicked environments. For complete assessment of the impact of global changes on mycorrhiza, we need to consider patterns and processes involved in both temporal scale and spatial scale. Mostly experiments mimic short-term responses, but for a complete study, we need to have an insight about long-term responses as well. A possible solution would include in situ experiments, for example, CO<sub>2</sub> springs. Large-scale events such as variations in distribution of plant species globally (it includes regional extinction of a particular plant) as an adverse effect of climate change may in turn affect the mycorrhizal population dependent on that plant as its host. Therefore, it may alter the mycorrhiza population distribution as well. Some of the mycorrhizal fungi are not dependent on the host (plant) but are still affected by changes in environment. This concept is least understood, but apparently is the most important factor to be considered when analyzing mycorrhiza's response toward global change. Global change is an intricate phenomenon, but here, we focus on how it affects the mycorrhizal fungi. Variations in the symptoms of global changes may be coined important in the future (examples are effect of urbanization, habitat fragmentation, etc.); these eventually may be included in the areas of study concerning mycorrhizal ecology. Factors which are involved in climate change which affect the mycorrhizal fungi are discussed below.

### ***18.3.1 Effect of Warming***

Soil is termed as “chief organizing center for ecosystem function”, and the part played by soil biota in modifying plant responses as well as ecosystem as a whole, toward global change, is being highly acknowledged. Microbes found in rhizosphere of a plant have a major role in sending strong feedbacks based on the plant's growth (Bever 1994), so any difference in the behavior or functions of these microorganisms as a result of global changes is of significance, as it may alter plant response to certain stimuli. When noting the key role of mycorrhiza at root-soil interface as well as its effect on plant physiology, community, and ecosystem, it is evident after a detailed consideration that mycorrhiza play a major role in studies related to global changes. AMF entirely depend upon their host (plant) for carbon requirement. Therefore, if the plant is affected by global changes, it will indirectly affect the AMF as well. Mycorrhiza will be considered important, if there is a notable

interrelation between global changes and presence of symbiosis, i.e., if the symbiont's presence or its function affects the response of plant communities or the ecosystem. Mycorrhizae are present abundantly in natural ecosystems, but in the case of habitat fragmentation or change in land utilization patterns, it may lead to decrease in the population of the fungi. This can contribute as another factor for global change (Vitousek 1994). It is important to know the inter-relation between factors leading to global changes and the presence of mycorrhiza. It will give a better insight on the underlying mechanism of variations occurring in the ecosystem as a response to global changes. The global change factors can affect the ecosystem in complex ways resulting in numerous variations even at regional levels, so it is important to prioritize understanding the mechanism behind it than to document the variations or changes. Based on these circumstances, mycorrhizal fungi (or fungi in general) has crucial role in maintaining and forming the soil structure, thereby benefiting the ecosystem (Tisdall and Oades 1982). If the extraradical mycelium of the mycorrhizal fungi is affected by any factors influenced by global change, it may indirectly impact the soil structure as well (Young et al. 1998; Rillig et al. 1999). Any change in soil structure will affect water infiltration ability, soilborne aspects involved in biogeochemical cycle operations, maintenance of sustainable agriculture, and resistance to soil erosion (Oades 1984; Elliott and Coleman 1988; Hartge and Stewart 1995; Jastrow and Miller 1997). Group of hyphae contribute to forming the mycelia of mycorrhizal fungi, which have a potential effect on the ecosystem. Based on these facts, mycorrhiza should be included in the research conducted to further understand the effects of global change.

### ***18.3.2 Effect of Elevated CO<sub>2</sub> Levels***

Approximately 90% of the plants form firm associations with mycorrhiza (Smith and Read 1997). Immediate need has been felt of understanding the impact of carbon (C) due to its increase in atmosphere in the form of carbon dioxide, on symbioses of mycorrhiza. Carbon is the main component for functioning of AM associations which are fixed in photosynthesis and has been seen to move from plant to fungus. Environment rich in carbon dioxide is the main factor for existence of fungus, be it inside the root or in the soil. Thus, if elevated atmospheric carbon increases the fixation of carbon and supply of it to roots then it might help in the promotion of fungal growth. This could lead to the consequence of increased fungal performance, which is best characterized for uptake of phosphate, increased uptake of nutrients rich in minerals, and resistance toward many phytopathogens and drought as well (Newsham et al. 1995). Elevated carbon dioxide can also have a positive aspect in which elevation can lead to enhanced fixation of carbon followed by increased carbon transfer to root system. Improved uptake of phosphate with increased mycorrhizal fungal growth due to elevated fixation of carbon might lead to efficient functioning of plants leading to relief in potential deficiency which could have otherwise caused restriction in ability of responding to increased level of

atmospheric carbon (suggestion by photosynthetic downregulation alleviation) (Staddon et al. 1999a). Important and essential processes in the plant metabolism of phosphorus are determined by carbon availability; these metabolisms include uptake, transportation, and utilization. In consideration, photosynthetic rate is determined by status of phosphorus of leaf. The positive feedback which could be possible eventually has constrained by few deficiencies, for example, nitrogen level and water content; thus it can act as a controlling or limiting factor. The main reason for understanding the responses of AM fungi is because they have potential of regulating response of plant toward climatic change; this is quite evident from the interface representation of AM fungi in soil-plant system. Link in transfer chain by which movement of carbon is seen from plant to soil is performed by mycorrhizal fungi, which makes it another major feature of interest (Staddon et al. 1999b). Thus, AM fungi can potentially influence cycling rates of carbon and can even use this structural carbon in construction of extraradical mycelium which is transferred to symbiotic AM fungus. The fine network formed by these mycelia has a faster turnover rate as compared to either root or shoot material, and fungal carbon is therefore a medium for a rapid movement of carbon back to atmosphere, whereas some of the fungal compounds might adapt and become resistant to attack of microbial community, thus entering the slow pool of recalcitrant soil carbon which leads to retardation in carbon cycle (Treseder and Allen 2000). This behavior of some fungal compound can lead to the consequence of either increased accumulated or reduced carbon content in plant soils. The consequence of elevated carbon dioxide in atmosphere for functioning of mycorrhiza could be changes in way of promotion in plant growth or changes in cycling rates of carbon. These effects are considered to be indirect, which is regulated by responses created by plants due to presence of carbon dioxide. Promotion in fungal ability to provide phosphorus to plants and enhance plant growth can be done by elevated carbon fixation and increased availability of carbon to fungus.

With respect to effects of mycorrhiza, elevation in carbon dioxide is considered as the best researched factor of global change. Increase in biomass of root of nitrogen fixing plant by 114%, in non-mycorrhizal individuals, was observed under elevation of carbon dioxide. But reduced or slower rate by 31% of effect was seen when AM species of genus *Glomus* was considered. Mycorrhizal fungi are a means of nutrient accession for plants. Uptake of inorganic phosphorus (Smith and Read 1997) and nitrogen (Ames et al. 1983; Tobar et al. 1994) is enhanced by AM fungi. Access to pool of organic nutrients in soil is usually provided by ecto-mycorrhizal fungi (Smith and Read 1997). Carbon source is required by fungi and uses an important amount of net photosynthate of plant which is usually about 10–20% (Allen 1991). As it is seen that capacity of photosynthesis, efficiency of water use, and growth are enhanced in plants which are exposed to elevation of carbon dioxide (Bazzaz 1990), then increase in investment of mycorrhizal fungi of amount of available carbon can be possible. Along with the rise of carbon demand, demand of nitrogen, phosphorus also rises. Therefore, it is assumed that allocation of resources for plants is done efficiently according to economic theories (Bloom et al. 1985; Read 1991); elevated carbon dioxide might increase mycorrhizal fungi investment. To study the effect of

elevated carbon in atmosphere, numerous workers experimented on plants by growing them at ambient (usually  $350 \mu\text{l}^{-1}$ ) as well as at elevated levels (variously 500, 600, 610, 700, and  $710 \mu\text{l}^{-1}$ ) ( $\text{eCO}_2$ ) of atmospheric carbon dioxide ( $\text{aCO}_2$ ) and measured root colonization degree by AM fungi (Staddon and Fitter 1998). Appropriate evidence has not been found from results acquired from experiment. Nevertheless, rarely seen case was about decreased colonization, and null to increased responses are found equally more frequent. Examination of morphological structures (hyphae, arbuscules, and, where appropriate, vesicles) of fungi was done more efficiently by workers who participated in the experiment. Later, they reported all possible combinations of decrease, increase, and null responses. Argument was made by Staddon and Fitter (1998) on effects which were due to serious flaw made in experiment; according to the researcher as elevated  $\text{CO}_2$  was observed to have a positive effect on growth of plants, he suggested that if plants show the same level of growth in both atmospheric and elevated carbon dioxide concentrations, then there must be some flaw in conducting experiment. As we know that plant characteristic does not include nature and degree of root mycorrhizal colonization, instead are dependent on conditions of plants in which they are grown; thus various changes in colonization were expected (Staddon 1998).

### 18.3.3 Effect of Temperature

Slow or poor spore germination was observed at RT ( $20\text{--}23^\circ\text{C}$ ) when continued experiments were performed specially to obtain axenic growth of various species of AMF. Several temperatures were compared to study its impact on spore germination and incidence of contaminating bacteria and fungi which are in association with surface-sterilized spores. Results were acquired with only those species that gave consistent spore germination rate, although experiments were conducted for evaluating various species of VA mycorrhizal fungi as well. When pot cultures were maintained with VA mycorrhizal fungi, then spores of respective fungi were obtained. As it was possible to maintain few isolates from various samples on pot cultures, it was used to maintain few isolates on pepper and soybean pot cultures. An isolate of *Gigaspora coralloidea* Trappe, Gerdemann and Ho and *G. heterogama* (Nicol. and Gerd.) Gerdemann and Trappe obtained from soybean in Florida were maintained on Bahia grass (*Paspalum notatum* Flugge) pot cultures. An isolate of *Glomus mosseae* (Nicol. and Gerd.) Gerdemann and Trappe from Washington was grown on pepper (*Capsicum annum* L.) pot cultures. Sieving and decanting techniques were used to remove spores from pot culture soil and roots. Surface sterilization was done of spores by using surface sterilizing agent that is 0.5% of sodium hypochlorite for three rounds and was later rinsed two times in a deionized sterile water before transfer to either soil extract agar or Mosse's medium No. 16 (Mosse and Phillips 1971). Preparation of soil extract agar was done by boiling finely sieved sandy loam of amount 200 g in a liter of deionized water. Later filtration was performed under vacuum, and addition of 15 g agar was done to a liter. Small

petri dishes were used for transferring spores and incubating them at desired temperatures; petri plates were incubated after transferring it in polyethylene bag to prevent dehydration and were later covered with aluminum foil for excluding light. Contamination from fungi and bacteria along with germination of spores was noted at regular intervals for 2–3 times. Three replicates were used of each treatment per temperature, and six to nine spores were observed at every replication. Studies were carried out with *Gigaspora coralloidea*, *G. heterogama*, and *Glomus mosseae* and were repeated three times, twice, and once, respectively. Maximum germination was obtained at temperature of 34 °C of two isolates taken from Florida (*Gigaspora coralloidea* and *G. heterogama*), of Washington isolate (*Glomus mosseae*) was 20 °C, and no germination was observed at the same temperature or below for *Gigaspora heteroganza* spores. Most surface sterilized spores were found to be contaminated by fungi and bacteria highly at a temperature of 25 °C. *Gigaspora heterogama* spores were highly contaminated with fungi, while no fungal association with spores of *Glomus mosseae* was found. Light was found to be a negative factor for growth of spores, and it was evident that maximum more growth of both *Gigaspora coralloidea* (at temperature of 20 and 25 °C) and *G. heterogama* (at temperature of 34 °C) was observed. Occurrence of contamination through bacteria was not found to differ on the basis of presence of light, whereas fungal contamination was observed to occur on surface-sterilized spores which were exposed to light ( $\times 14.8\%$ ). Study conducted by Schenck and Schroder (1974) showed the results of maximum sporulation and vesicle formation of Florida isolate of *Gigaspora calospora* (Nicol. and Gerd.) Gerdemann and Trappe on soybean pot culture [*Glycine max* (L.) Merrill] at temperature of 35 °C. Results of spore germination gave endorse evidence that adaptation at high temperature occurs in Florida isolates *Gigaspora* spp. (Schenck and Schroder 1974). It was also assumed that northern isolates of endomycorrhizal fungi, such as the Washington isolate of *Glomus mosseae*, may be more adapted to lower temperatures.

#### **18.3.4 Effect of Gases on Spore Germination of AM Fungi**

Production of aqueous extracts or suspensions was done for testing various effects of beet root HT material on spore germination of AM fungi; the suspension was made by forming a mixture of HTC material with deionized water thereby making adjustments in pH to 6.0, later autoclaved for about 20 min at a temperature of 121 °C. Since pH is found to influence germination of spores, thus it was kept constant (Green et al. 1976). Concentrations used were 0.05, 0.25, 1.25 g HTC 100 mL<sup>-1</sup> and the parent material (also ground and sieved to 500 m) 1.25 g beet root chips 100 mL<sup>-1</sup>. Addition of 12 milliliters of suspensions were performed under axenic conditions, to each petri dish which were filled prior with sterilized silica sand (48 g) for total of 50 units of experiment ( $n = 10$ ). Small discs were cut of nitrous cellulose with a help of paper hole punch after sterilizing them with 70% of ethanol. Preparation of spores was done according to the description made by Antunes et al.

(2008). Paraffin tape was used to seal all petri dish, and later petri dishes were kept for incubation at 25 °C for 4 days at random positions. After the completion of incubation duration, spores were stained using 0.05% (w/v); trypan blue examination was done using dissecting microscope (40×); thus, hyphal growth was evident, which was carefully observed, and distinguished germination hyphae from already present hyphae found attached to spores. Previous trails found showed various germination ranges, and it was concluded that a spore is considered non-viable if germination does not take place within duration of 48 h under the provided conditions (Antunes et al. 2008). This experiment finally concluded that range of spore germination is from 50 to 80% across all treatments, besides that significant difference was found in effect of treatments on germination of spores ( $p < 0.0001$ ). Increased germination of spores than parent material was recorded after application of all three HTC treatments. But this experiment had a limitation that addition of only highest rate showed effective high germination of spores than no-addition control. Parent material was found to have negative effect on germination as decrease of spore germination was observed. Thus, evidence was confirmed by concluding deleterious effect of HTC material on growth of plants, but at least symbionts like AM fungi are found to be less sensitive than plants and more stimulated under higher rates of addition.

## 18.4 Impact of Climate Change in Plant-Microbe Interactions

Elucidation of physical processes occurring in upper soil layers can be done by studying climatic parameters, and atmospheric region at lower level plays a significant role in determining the climate for the local or regional biosphere (Monteith and Unsworth 2007). Migration of plant species to more altitudes and latitudes (Grabherr et al. 1994; Walther et al. 2002; Parmesan and Yohe 2003) has been observed with warming; also these species are leafing out and flowering in the growing season to its earliest (Cleland et al. 2007; Wolkovich et al. 2012) and even seen to be creating alterations in the expression of functional traits (Walker et al. 2006; Hudson et al. 2011; Verheijen et al. 2015). Observations were made when scaling up happened to community level; shrubification of the arctic was seen as a result of warming as woody shrubs have been replaced grasses and forbs in several regions which lead to carbon feedbacks in system and transformation in properties of ecosystem (Sturm et al. 2001; Hinzman et al. 2005; Lawrence and Swenson 2011; Pearson et al. 2013). Plant community transitions may be facilitated or retarded by soil communities, especially the ones which are tightly coupled with plants. For instance, plant survival, phenotype, and expression of functional traits, that are responsible for climatic change, can be strongly influenced by root-associated microbe communities (Van der Heijden et al. 1998; Friesen et al. 2011; Wagner et al. 2014). There is no clear understanding made till date on plant-plant

interactions, their association with microbial community, and ecosystem functioning due to climatic changes (Fischer et al. 2014; Mohan et al. 2014). Induction in major shifts in bulk of microbial community by climatic alterations have shown extended effects on establishments and performance of plants as well as balance of carbon in the soil. Responses of ecosystem will be dependent on association between plants and soil microbes, and if alterations are made in soil microbial communities, due to climatic change and plant growth, establishment of plant species is also determined by these changes. Buffering of plants to drought stress can be done by rapid responses of surrounding soil community which is becoming a suggestion by recent studies (Lau and Lennon 2012). Microbial diversity changes are responsible for alteration of functional traits in plants (Lau and Lennon 2011). Significant difference is there in direct and indirect impact of climate change on plants, bulk soil community, and their associated soil communities (Kardol et al. 2010). For instance, Kardol et al. (2010) found out that alterations of soil community and its functions in TN old field occurred with changes in precipitation, but variations were observed with respect to precipitation on soil microbes, its functioning, and composition on basis of location from where the sample soil was collected. Moreover, response of being relatively muted was seen from soils which were collected from different locations, and these were homogenized to access to climatic change impact. Results suggested that offset responses of soil ecosystem to climatic changes could be expected if shift in composition of plant community due to climate change occurs. Therefore, under-representation of these soils which are collected across plant species has been seen in most of the studies to cause shift in functional and communal plant species (Kardol et al. 2010).

**Temperature** Alleviation of negative impact of temperature stress on plants could be done by few organisms such as rhizospheric bacteria and endophytes; these organisms also help in expansion of capability of host plants to show growth at different ranges of temperatures. Example of the symbiosis of the organisms for allowance of both organisms to grow at high temperatures of soil is symbiosis between tropical panic grass *Dichanthelium lanuginosum* and the fungus *Curvularia protuberata*, but neither of them can survive independently at such high temperatures (Marquez et al. 2007). Infection of fungus by thermal tolerance virus, that is *Curvularia*, is required to confer ability of *C. protuberata*, for tolerating heat to the host plant (Marquez et al. 2007). Observation on tomato was made that in addition to panic grass, it could also have *C. protuberata*-mediated heat tolerance (Rodriguez et al. 2008); thus it could suggest that broad application of this underlying mechanism to diversity of plants could cope with increased temperatures. There are some organisms which can even help plants in coping up with different multiple stresses. An intriguing example of this is *Burkholderia phytofirmans* strain PsJN, which helps in improvement in tomato for plant tolerance to heat, cold in grapevine, water stress in wheat, and salinity along with freezing in *Arabidopsis* (Issa et al. 2018; Miotto-Vilanova et al. 2016). This bacterium is not only capable of creating heat tolerance but also has capability of acting as antifungal and can act as a plant defense mechanism primely and formation of better mobilization resource in plants



(Miotto-Vilanova et al. 2016; Timmermann et al. 2017). The mechanism remains elucidated by which PsJN confers multistress tolerance, and this elucidation can act as an interesting topic for crop improvement which will be mediated by microbes.

**Drought** Water has an essential part in life on Earth, and variation in the amount of water that is too little (lead to osmotic stress) or too much (leads to flooding) can have great impact in many areas of biology of microbes and plants. Regulation of phytohormone level, ABA, occurs during the deficiency of water in plants. Increase of ABA is responsible for triggering a cascade of signals, which results in reprogramming of transpiration at large scale and changes in physiology, which may include stomatal closure for reducing transpiration (Zhu 2016). Studies have been conducted on *Arabidopsis* which showed that pathogens specifically bacterial, such as *P. syringae* or PAMPs, like flg22 (a 22-amino-acid epitope of *Pseudomonas flagellum*), can be perceived by FLS2, which results in closure of stomata, thus helping in reduction of pathogenic entry (Melotto et al. 2006). Thus, stomatal closure, induced by ABA, might help in reduction of bacterial entry through stomata during drought stress. Suppression of SA signaling pathway in mesophyll cell of leaf can occur on elevation of ABA, and thus it can lead to decrease in post-invasion resistance mediated by SA (Jiang et al. 2010). Plant-microbiome is also affected by drought. Increase in intensity of drought leads to increased effect on microbial community composition (bulk soil, rhizosphere, and root endosphere) which resulted in immense intimate association of community with the root, also greater shift of composition in stressed rice plants which was found out by Santos-Medelln et al. (2017). Xu et al. (2018) found out in a study which examined influence of soil moisture on microbiome of sorghum root that there was significant reduction in rhizospheric diversity in root endosphere, while bacterial community diversity in soil surrounding remains mostly unaltered.

**Nutritional Status** Plant-microbe interaction-driven ultimate force is acquisition of nutrients. There is a significant effect on plant-microbe interactions, and this is possible due to plant nutritional level and availability of nutrients in the environment.

**Phosphate** Regulation of complex symbiotic association between land plants and phosphate accessing AMF is done by phosphate status in soil and its presence in the plants (Müller and Harrison 2019). Therefore, plant AMF interactions are out of the reach of effect of phosphate on plant-microbe interactions. *Arabidopsis thaliana* is considered to be a non-host for AMF that acquires phosphate (Fernandez et al. 2019). Identification was done by Hiruma et al. (2016) of a natural fungus which is endophytic in nature, known as *Colletotrichum tofieldiae* (Ct), isolated from wild *Arabidopsis* found in central Spain. Ct is responsible for transfer of phosphate to *Arabidopsis* and for promotion of fertility and growth of plants. Promotion of growth which is mediated by Ct is also observed when study was conducted on plants grown under conditions having deficiency of phosphate.

**Nitrogen** Symbiotic nodules are formed in root as a result of interactions between legumes and *Rhizobium* spp. Thus, it represents a biological process in which

conversion of inert atmospheric  $N_2$  occurs biologically into a useful  $NH_3$  for facilitating growth and development of plants. Although this reaction is expensive energetically, the formation of nodules by legume host may act as cost-effective approach when nitrogen-rich environment is required for growing plants (Morgan et al. 2005). Susceptible state of rhizobium symbiotic interaction is maintained by communication of shoots with roots under conditions having deficiency of nitrogen.

**Iron** Induced systemic resistance is a form of plant immunity which is triggered by few microbes which live in rhizosphere, forming a mutual relation with prime host, against potential attacks from pathogens. Mutant analysis and microarray were conducted for identifying Arabidopsis transcription factor MYB72 which act as a key regulator of ISR (Van der Ent et al. 2008). Induction of MYB72 factor expression occurs in roots during the deficiency of iron (Buckhout et al. 2009).

## 18.5 Conclusion

Human-induced changes have led to serious repercussions in the Earth's environment other than climate change. Some important examples are increased concentration of the atmospheric gases (ozone,  $CO_2$ , etc.), the ecosystem being exposed to a rising number of nutrients due to atmospheric deposition (nitrogen), change in climate (altered precipitation as well as temperature regimes), increase in UV radiations, and occurrence of non-native species. These components comprise present or upcoming global changes, which can have a positive or negative impact on microbial associations and their existence. Hence, it is necessary to control the climatic alterations and overcome the harmful effects caused to the beneficial microbes such as plant growth-promoting rhizobacteria and arbuscular mycorrhizal fungi.

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