RESEARCH ARTICLE

EFFECT OF CLIMATE CHANGE SOIL GLOBAL MICROORGANISMS

[1,]*Hamid Kheyrodin and [2]Khosro Ghazvinian

[1] Faculty of Desert Science-Semnan University, Iran
[2]Faculty of Veterinary and Animal Science-Semnan University, Iran

ARTICLE INFO

ABSTRACT

Given that it is generally accepted that the climate is changing, it is essential to investigate the effects that changes in climate parameters (temperature, precipitation) will have on the soil C cycle (through changes in inputs and outputs) and on the activity of soil microorganisms. Soils provide essential ecosystem services such as primary production, regulation of biogeochemical cycles and their consequences on climate, water filtration, resistance to diseases and pests, and regulation of above-ground biodiversity. Concomitantly, soils are exposed to many anthropogenic threats. Scientific and technological knowledge on soil biodiversity and functioning in relation to ecosystem services is required for reaching such a goal. Soils host a huge diversity of microbes (archaea, bacteria, fungi) and fauna (protozoa, microarthropods, nematodes, oligochaeta) for which our SS knowledge of function and diversity remains very limited. This is related to the small size of the soilborne organisms, their diversity, the difficulty of accessing them, and to the great heterogeneity of their habitats at different scale levels. However, recent progresses in the molecular characterization of the biodiversity raises stimulating prospects to explore its complexity and better understand its functioning. Soil microorganisms, the central drivers of terrestrial Antarctic ecosystems, are being confronted with increasing temperatures as parts of the continent experience considerable warming. Here we determined short-term temperature dependencies of Antarctic soil bacterial community growth rates, using the leucine incorporation technique, in order to predict future changes in temperature sensitivity of resident soil bacterial communities. In this study soil samples were collected along a climate gradient consisting of locations on the central Iran in Semnan city. The result demonstrated that the bacterial communities were adapted to the mean annual temperature of their environment, as shown by a significant correlation between the mean annual soil temperature and the minimum temperature for bacterial growth (Tmin). Every 1 1C rise in soil temperature was estimated to increase Tmin by 0.24–0.38 1C. The optimum temperature for bacterial growth varied less and did not have as clear a relationship with soil temperature. Temperature sensitivity, indicated by Q10 values, increased with mean annual soil temperature, suggesting that bacterial communities from colder regions were less temperature sensitive than those from the warmer regions.

INTRODUCTION

Groups of microorganisms for which the soil is the natural habitat. They play an important role in the cycle of matter in nature, soil formation, and soil fertility. Soil microorganisms can develop directly in soil, as well as in decomposing plant residues. Some pathogenic microbes and aquatic microorganisms may accidentally enter soil during the decomposition of dead bodies, from the gastrointestinal tract of animals and man, with irrigation water, or by other routes, but they generally die quickly. However, some of them persist a long time (for example, anthrax bacilli and the causative agents of tetanus) and can become a source of infection for man, animals, and plants. Soil microorganisms constitute a large part of the microorganisms on our planet: 1 g of chernozem contains about 10 billion or more living microorganisms (10 tons per hectare). Soil microorganisms

consist of both prokaryotes (bacteria, actino-mycetes, blue-green algae) and eukaryotes (fungi, microscopic algae, protozoans). Owing to the use of such modern methods as electron and capillary microscopy, many new species of soil microorganisms are discovered every year. Varying greatly in their properties and functions, soil microorganisms include heterotrophs and autotrophs, aerobes and anaerobes (Fig. 1). They differ sharply in optimum pH, relation to temperature, osmotic pressure, and source of organic and inorganic matter used. Despite different and sometimes directly opposite requirements, many of them develop in the same soil, which consists of a large number of very different microenvironments. The number of soil microorganisms varies with the season: there are more in spring and fall and fewer in winter and summer. The top layers of soil have more microorganisms than do the lower layers. Microorganisms are particularly abundant in the root zone of plants, or rhizosphere. Cultivation, the addition of fertilizers, and the creation of a favorable water regime help soil microorganisms become more numerous and active.

Corresponding author: Hamid Kheyrodin
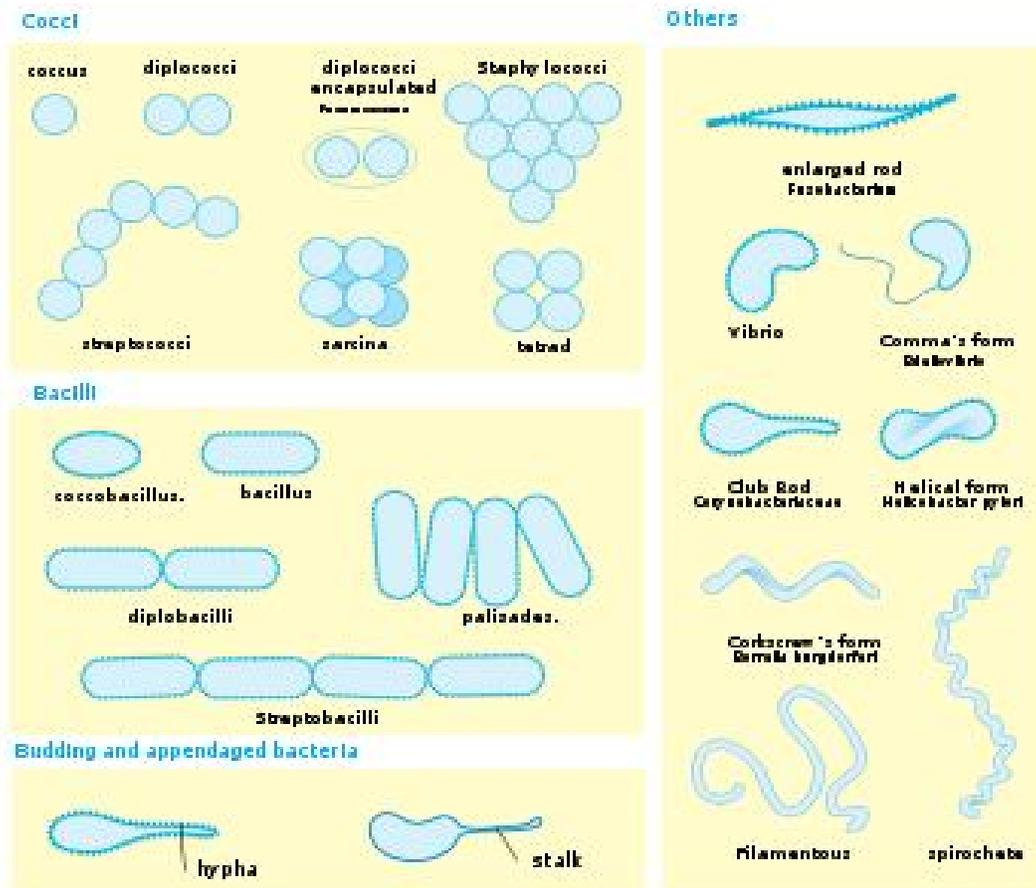Assistant professor Faculty of Desert Science-Semnan University, Iran

Fig. 1. Bacteria display many cell morphologies and arrangements

The most important planetary function of soil microorganisms is to participate in the cycle of matter and in the conversion processes of important biogenic elements—O, C, N, P, S, and Fe. Soil microorganisms are capable of breaking down all natural organic compounds and some organic compounds not found in nature. They perform an important role in freeing the biosphere from pollutants, chiefly by decomposing pesticides and oxidizing carbon monoxide. The properties of different soil groups and the variations in their fertility are largely determined by the nature of the soil microorganisms and by their activity. Some species of soil microorganisms are used in the microbiological synthesis of antibiotics, vitamins, enzymes and other proteins, amino acids, and gibberellins. Most antibiotics, for example, are produced by growing soil actinomycetes in cultures. Microorganisms found in the soil are vital to many of the ecological processes that sustain life such as nutrient cycling, decay of plant matter, consumption and production of trace gases, and transformation of metals (Panikov, 1999).

Although climate change studies often focus on life at the macroscopic scale, microbial processes can significantly shape the effects that global climate change has on terrestrial ecosystems. According to the International Panel on Climate Change (IPCC) report (2007), warming of the climate system is occurring at unprecedented rates and an increase in anthropogenic greenhouse gas concentrations is responsible for most of this warming. Soil microorganisms contribute significantly to the production and consumption of greenhouse
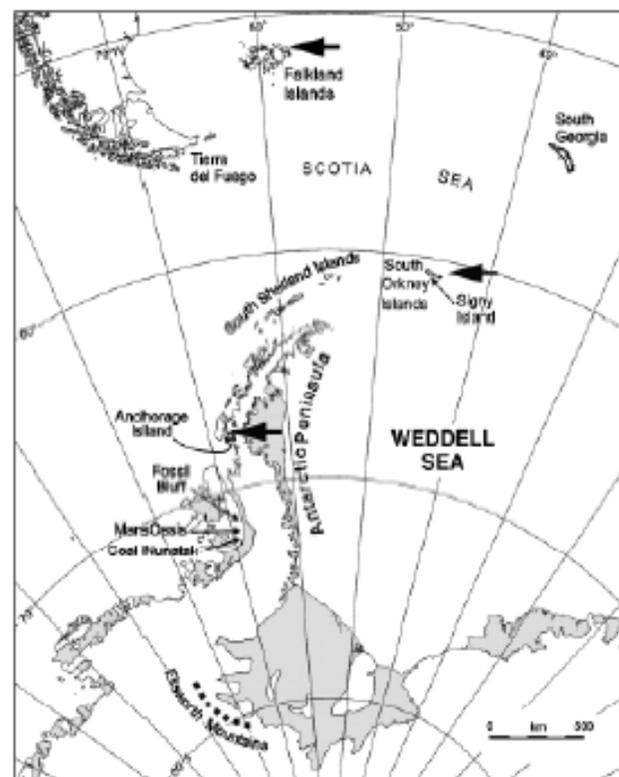

Fig. 2: Map of the Antarctic climate gradient.

gases, including carbon dioxide (CO2), methane (CH4), nitrous oxide (N2O), and nitric oxide (NO), and human activities such as waste disposal and agriculture have stimulated the production of greenhouse gases by microbes. As concentrations of these gases continue to rise, soil microbes may have various feedback responses that accelerate or slow down global warming, but the extent of these effects are unknown. Understanding the role soil microbes have as both contributors to and reactive components of climate change can help us determine whether they can be used to curb emissions or if they will push us even faster towards climatic disaster (Fig. 2). Objective of this paper is to examine two aspects of the relationship between soil microorganisms and global climate change. First, I will explore how soil microorganisms contribute to greenhouse gas emissions and then, conversely, I will examine the possible effects that increased temperature and CO2 concentrations in the atmosphere may have on soil microbial processes.

MATERIAL AND METHOD

One gramof wet soil of each of the 75 soil samples was placed in a 50mL centrifuge tube, and 20mL distilled water was added. After 3 min at full speed on a multivortex shaker and 10min low-speed centrifugation (1000 g), 1.5mL aliquots of the bacterial suspension were distributed to eight 2mL micro centrifugation vials. These were then placed in a water bath for 30 min at 1, 4, 8, 12, 17.5, 24.5, 29 and 34 1C to achieve the correct temperature before 2 mL L-[4,5-3H] leucine (171 CimmoL_1, 1.0 mCimL_1, Amersham) and nonradioactive L-leucine were added, resulting in a final concentration of 270 nM leucine. Incubation times were 48 h at 1 1C, 24 h at 4 1C, 10 h at 8 1C, 6 h at 12 1C, 4 h at 17.5 1C and 2 h at the higher temperatures. These incubation times were chosen in order to achieve similar absolute leucine incorporation in all samples, while not inducing any changes in growth rates during the incubation period. The bacterial incorporation of leucine was terminated by adding trichloroacetic acid. Washing and measurement of the incorporated 3H-leucine was then performed according to Ba°a°th *et al*. (2001). The amount of leucine incorporated into the extracted bacterial suspension per hour per gram soil was used as a measure of bacterial growth.

Data analysis

The data were modelled using one function below optimum and another above optimum temperature for bacterial growth. Between the T min and To pt we used a square root relationship, which has earlier been shown to adequately model bacterial growth in pure culture, including psychrotrophic and thermofilic bacteria (Ratkowsky *et al*., 1982, 2005). The same relationship has also been found to model adequately the temperature relationship of bacterial community growth in both water (Li & Dickie, 1987) and soil Dı´az-Ravin˜ a *et al*., 1994).

$$Leu^{1/2} = a(T - T_{min}) \qquad (1)$$

Leu is the leucine incorporation measured at the temperature T 1C, Tmin is the apparent minimum temperature for leucine incorporation, and a is a slope parameter (without any direct biological meaning). A plot of the square root of the leucine incorporation against temperature will result in a linear relationship, with T min indicated by the intercept with the x-axis. For many environmental samples, T min will fall well below the freezing point of water. For this reason, T min is commonly denoted apparent minimum temperature. However, T min is a useful parameter when comparing different communities, in that lower values generally indicate a better capacity to grow at low temperatures, designating a community adapted to low temperature conditions. This is also the case in pure culture isolates. Ratkowsky *et al*. (2005) reported Tmin for thermophilic bacteria to be around 20 1C, for mesophiles down to _8 1C, and for psychrophiles even lower, down to _15 1C. Effects of the warming treatment, location and vegetation type on the various indexes of temperature adaptation were tested using Linear mixed models of SPSS 14.0. The model included these three factors and all interactions. Bacterial growth rate and the ratio of growth rate at 30 1C/4 1C were log-transformed before analysis.

RESULT AND DISCUSSION

Microbial contributions to greenhouse gas emissions

Soil microorganisms are a major component of biogeochemical nutrient cycling and global fluxes of CO2, CH4, and N. Global soils are estimated to contain twice as much carbon as the atmosphere, making them one of the largest sinks for atmospheric CO2 and organic carbon (Jenkinson and Wild, 1991; Willey *et al*., 2009). Much of this carbon is stored in wetlands, peat lands, and permafrost, where microbial decomposition of carbon is limited. The amount of carbon stored in the soil is dependent on the balance between carbon inputs from leaf litter and root detritus and carbon outputs from microbial respiration underground (Davidson and Janssens, 2006). Soil respiration refers to the overall process by which bacteria and fungi in the soil decompose carbon fixed by plants and other photosynthetic organisms and release it into the atmosphere in the form of CO2. This process accounts for 25% of naturally emitted CO2, which is the most abundant greenhouse gas in the atmosphere and the target of many climate change mitigation efforts. Small changes in decomposition rates could not only affect CO2 emissions in the atmosphere, but may also result in greater changes to the amount of carbon stored in the soil over decades (Davidson and Janssens, 2006).

Methane is another important greenhouse gas and is 25 times more effective than CO2 at trapping heat radiated from the Earth (Schlesinger and Andrews, 2000). Microbial methanogenesis is responsible for both natural and human-induced CH4 emissions since methanogenic archaea reduce carbon into methane in anaerobic, carbon-rich environments such as ruminant livestock, rice paddies, landfills, and wetlands. Not all of the methane produced ends up in the atmosphere however, due to methanotrophic bacteria, which oxidize methane into CO2 in the presence of oxygen. When methanogens in the soil produce methane faster than can be used by methanotrophs in higher up oxic soil layers, methane escapes into the atmosphere (Willey *et al*., 2009). Methanotrophs are therefore important regulators of methane fluxes in the atmosphere, but their slow growth rate and firm attachment to soil particles makes them difficult to isolate. Further exploration of these methanotrophs' nature could potentially help reduce methane emissions if they can be

added to the topsoil of landfills, for example, and capture some of the methane that would normally be released into the atmosphere. Not unlike their role in the carbon cycle, soil microorganisms mediate the nitrogen cycle, making nitrogen available for living organisms before returning it back to the atmosphere. In the process of nitrification (during which ammonia is oxidized to nitrate), microbes release NO and N_2O, two critical greenhouse gases, into the atmosphere as intermediates. Evidence suggests that humans are stimulating the production of these greenhouse gases from the application of nitrogen-containing fertilizers (Willey *et al*., 2009). For example, *Nitrosomonas eutropha* is a nitrifying proteobacteria found in strongly eutrophic environments due to its high tolerance for elevated ammonia concentrations. N-fertilizers increase ammonia concentrations, causing *N. eutropha* to release more NO and N_2O in the process of oxidizing ammonium ions. Since NO is necessary for this reaction to occur, its increased emissions cause the cycle to repeat, thereby further contributing to NO and N_2O concentrations in the atmosphere (Willey *et al*., 2009).

Microbial responses to global climate change

Microbial processes are often dependent on environmental factors such as temperature, moisture, enzyme activity, and nutrient availability, all of which are likely to be affected by climate change (IPCC, 2007). These changes may have greater implications for crucial ecological processes such as nutrient cycling, which rely on microbial activity. For example, soil respiration is dependent on soil temperature and moisture and may increase or decrease as a result of changes in precipitation and increased atmospheric temperatures. Due to its importance in the global carbon cycle, changes in soil respiration may have significant feedback effects on climate change and severely alter aboveground communities. Therefore, understanding the response of soil respiration to climate change is of great importance and will be discussed in detail in this report.

Microbial response to increased temperatures

One of the major uncertainties in climate change predictions is the response of soil respiration to increased atmospheric temperatures (Briones *et al*., 2004; Luo *et al*., 2001). Several studies show that increased temperatures accelerate rates of microbial decomposition, thereby increasing CO_2 emitted by soil respiration and producing a positive feedback to global warming (Allison *et al*., 2010). Under this scenario, global warming would cause large amounts of carbon in terrestrial soils to be lost to the atmosphere, potentially making them a greater carbon source than sink (Melillo *et al*., 2002). However, further studies suggest that this increase in respiration may not persist as temperatures continue to rise. In a 10-year soil warming experiment, Melillo *et al*. (2002) show a 28% increase in CO_2 flux in the first 6 years of warming when compared to the control soils, followed by considerable decreases in CO_2 released in subsequent years, and no significant response to warming in the final year of the experiment. The exact microbial processes that cause this decreased long-term response to heated conditions have not been proven, but several explanations have been proposed. First, it is possible that increased temperatures cause microbes to undergo physiological changes that result in reduced carbon-use efficiency (Allison *et al*., 2010). Soil microbes may

also acclimate to higher soil temperatures by adapting their metabolism and eventually return to normal decomposition rates. Lastly, it can be interpreted as an aboveground effect if changes in growing-season lengths as a result of climate change affect primary productivity, and thus carbon inputs to the soil (Davidson and Janssens, 2006). The effects of increased global temperatures on soils is especially alarming when considering the effects it has already begun to have on one of the most important terrestrial carbon sinks: permafrost. Permafrost is permanently frozen soil that stores significant amounts of carbon and organic matter in its frozen layers. As permafrost thaws, the stored carbon and organic nutrients become available for microbial decomposition, which in turn releases CO_2 into the atmosphere and causes a positive feedback to warming (Davidson and Janssens, 2006). One estimate suggests that 25% of permafrost could thaw by 2100 as a result of global warming, making about 100 Pg of carbon available for microbial decomposition (Davidson and Janssens, 2006; Anisimov *et al*., 1999). This could have significant effects on the global carbon flux and may accelerate the predicted impacts of climate change. Moreover, the flooding of thawed permafrost areas creates anaerobic conditions favorable for decomposition by methanogenesis. Although anaerobic processes are likely to proceed more slowly, the release of CH_4 into the atmosphere may result in an even stronger positive feedback to climate change (Davidson and Janssens, 2006).

Microbial response to increased CO2

Atmospheric CO_2 levels are increasing at a rate of 0.4% per year and are predicted to double by 2100 largely as a result of human activities such as fossil fuel combustion and land-use changes (Lal, 2005; IPCC, 2007). Increased CO_2 concentrations in the atmosphere are thought to be mitigated in part by the ability of terrestrial forests to sequester large amounts of CO_2 (Schlesinger and Lichter, 2000). To test this, an international team of scientists grew a variety of trees for several years under elevated CO_2 concentrations. They found that high CO_2 concentrations accelerated average growth rate of plants, thereby allowing them to sequester more CO_2. However, this growth was coupled with an increase in soil respiration due to the increase in nutrients available for decomposition by releasing more CO_2 into the atmosphere (Willey *et al*., 2009). This suggests that forests may sequester less carbon than predicted in response to increased CO_2 concentrations, however more research is needed to investigate this hypothesis.

Soil-borne pathogens and climate change

According to the IPCC (2007) report, climate change will alter patterns of infectious disease outbreaks in humans and animals. Soil pathogens are no exception: case studies support the claim that climate change is already changing patterns of infectious diseases caused by soil pathogens. For example, over the last 20 years, 67% of the 110 species of harlequin frogs (*Atelopus*) native to tropical regions in Latin America have gone extinct from chytridiomycosisthe, a lethal disease spread by the pathogenic chrytid fungus (*Batrachochytrium dendrobatidis*) (Willey *et al*., 2009). Research suggests that mid- to high-elevations provide ideal temperatures for *B. dendrobatidis*. However, as global warming progresses, *B. dendrobatidis* is able to expand its range due to increasing

moisture and warmer temperatures at higher elevations (Muths *et al*., 2008). This expansion exposes more amphibian communities in previously unaffected or minimally affected areas, specifically at higher elevations, to chytridiomycosisthe. As seen in the case of *Atelopus* harlequin frogs, the spread of soil pathogens due to climatic changes can significantly affect life at the macro scale and ultimately lead to species extinction.

Conclusion

The complexity of microbial communities living belowground and the various ways they associate with their surroundings make it difficult to pinpoint the various feedback responses that soil microbes may have to global warming. Whether a positive feedback response results, in which microbial processes further contribute to climate change, or whether a negative feedback response slows its effects, it is clear that microbes can have a huge impact on future climate scenarios and ecosystem-level responses to climate change. Soil respiration plays a pivotal role in these effects due to the large amount of CO_2 and CH_4 emissions produced during respiration, the reliance of carbon stocks in soils on rates of respiration, and the initial sensitivity of soil respiration to increased atmospheric temperatures. Further studies in long-term feedback effects of soil respiration on climate change can contribute to our understanding of the overall impacts of climate change, including the ability of terrestrial forests to uptake excess CO_2 from the atmosphere. As we attempt to mitigate greenhouse gas emissions and adapt to predicted climate change effects, turning towards microscopic life that lies below the surface can perhaps help us to become better equipped for future changes at the macroscopic and even global scale.

Acknowledgements

REFERENCES

Agren, G.I. 2010. Climate Change: Microbial mitigation. *Nature Geoscience* 3, 303-304.

Allison, S.D., Wallenstein, M.D., Bradford, M.A. 2010. Soil-carbon response to warming dependent on microbial physiology. *Nature Geoscience* 3, 336-340.

Anisimov, O.A., Nelson, F.E., Pavlov, A.V. 1999. Predictive scenarios of permafrost development under conditions of global climate change in the XXI century. *Earth Cryology* 3, 15-25.

Ba°a°th E, Pettersson M, So¨derberg KH (2001) Adaptation of a rapid and economical microcentrifugation method to measure thymidine and leucine incorporation by soil bacteria. Soil Biology and Biochemistry, 33, 1571–1574.

Briones, M.J.I., Poskitt, J., Ostle, N. 2004. Influence of warming and enchytraeid activities on soil CO_2 and CH_4 fluxes. *Soil Biology and Biochemistry* 36, 1851-1859.

Davidson, E.A. and Janssens, I.A. 2006. Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. *Nature* 440, 165-173. IPCC. 2007. Climate Change 2007: Synthesis Report.

Dı´az-Ravin˜ a M, Frostega°rdA °, Ba°a°th E (1994) Thymidine, leucine and acetate incorporation into bacterial assemblages at different temperatures. FEMS Microbiology Ecology, 14, 221–231.

Jenkinson, D.S., Adams, D.E., Wild, A. 1991. Model estimates of CO_2 emissions from soil in response to global warming. *Nature* 351, 304–306.

Lal, R. 2005. Forest soils and carbon sequestration. *Forest Ecology and Management* 220, 242-258.

Li WKW, Dickie PM (1987) Temperature characteristics of photosynthetic and heterotrophic activities: seasonal variation in temperate microbial plankton. Applied and Environmental Microbiology, 53, 2282–2295.

Luo, Y., Wan, S., Hui, D., Wallace, L.L. 2001. Acclimatization of soil respiration to warming in a tall grass prairie. *Nature* 413, 622-625,

Melillo, J.M., Steudler, P.A., Aber, J.D., Newkirk, K., Lux, H., Bowles, F.P., Catricala, C., Magill, A., Ahrens, T., Morrisseau, S. 2002. Soil Warming and Carbon-Cycle Feedbacks to the Climate System. *Science* 298, 2173-2176.

Muths, E., Pilliod, D.S., Livo, L.J. 2008. Distribution and environmental limitations of an amphibian pathogen in the Rocky Mountains, USA. *Biological Conservation* 141, 1484-1492.

Novogrudskii, D. M. *Pochvennaia mikrobiologiia.* Alma-Ata, 1956.

Panikov, N.S. 1999. Understanding and prediction of soil microbial community dynamics under global change. *Applied Soil Ecology* 11, 161-176.

Ratkowsky DA, Olley J, McMeekin TA, Ball A (1982) Relationship between temperature and growth rate of bacterial cultures. Journal of Bacteriology, 149, 1–5.

Ratkowsky DA, Olley J, Ross T (2005) Unifying temperature effects on the growth rate of bacteria and the stability of globular proteins. Journal of Theoretical Biology, 233, 351–362.

Schlesinger, W. H. and Andrews, J. A. 2000. Soil respiration and the global carbon cycle. *Biogeochemistry* 48, 7–20.

Schlesinger, W.H. and Lichter, J. 2001. Limited carbon storage in soil and litter of experimenta forest plots under increased CO_2. *Nature* 411, 466-469.

Willey, J.M., Sherwood, L.M., Woolverton, C.J. Prescott's Principles of Microbiology. McGraw-Hill, New York, NY: 2009.

****