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ADAPTABILITY OF SOIL pH THROUGH INNOVATIVE MICROBIAL APPROACH

Aqarab Husnain Gondal[1+]
Qammar Farooq[2]
Sidra Sohail[3]
Shamal Shasang Kumar[4]
Muhammad Danish Toor[5]
Asma Zafar[6]
Bushra Rehman[7]

[1,2,3,6,7]*Institute of Soil and Environmental Sciences, University of Agriculture, Faisalabad, Pakistan.*
[1]*Email: aqarabhusnain944@gmail.com Tel: +923415831944*
[2]*Email: qammarfarooq742@gmail.com Tel: +923160688683*
[3]*Email: sidfatima65@gmail.com Tel: +923347503681*
[6]*Email: asmaazafar931@gmail.com Tel: +923338656888*
[7]*Email: boshrakhan229@gmail.com Tel: +923346826205*
[4]*Division of Soil Science and Agricultural Chemistry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Jammu and Kashmir, India.*
[4]*Email: shamalkumar1997@gmail.com Tel: +919541185424*
[5]*European University of Lefke, Institute of Graduate Studies and Research, Department of Environmental Sciences Northern Cyprus TR-10 Mersin Turkey.*
[5]*Email: mdtoor336@gmail.com Tel: +923084519649*

(+ Corresponding author)

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ABSTRACT

Soil pH is a critical characteristic that regulates the abundance of essential nutrients in the soil system. Low soil pH reduces the supply of secondary macronutrients, whereas higher pH restricts soil micronutrient availability. In addition, soil nutrient sources such as organic and inorganic fertilizers by plants require an adequate pH for optimum plant growth and productivity. The soil pH is regarded as the “master of soil indices", which has a role to play in controlling biogeochemical cycles that influence plant growth. It also has an enormous influence on microbial biodiversity in the soil. Various approaches have been used to alter the soil pH, demonstrating that it is not easy to adjust soil pH. Therefore, a suitable but practical approach is required to control or change the pH of the rhizosphere. Microbial breeding technique such as genome replication may be an appropriate option to alter the pH of the rhizosphere. Genetically engineered microbes may have the exceptional ability to release sufficient acidic or basic compounds that could increase or decrease the pH levels in the rhizosphere. In recent years, this view has helped answer some common evolutionary concerns regarding how bacteria and their host species have evolved from their early ancestors. Greater exploitation of microbes in this respect would be necessary for sustainable crop production and helping to resolve issues related to soil-plant interactions for nutrients. To breed the microbes selectively for optimal nutritional interaction with plants, the genetic components of different traits must first be explored.

Contribution/Originality: The present review describes the importance of microbes towards pH and their role in altering pH in the rhizosphere. This study is one of the very few studies that have investigated microbial genetics and if the genetic approaches become beneficial, it will lead towards the next revolution.

1. INTRODUCTION

Soil is a three-phase system made up of liquid, gas and solid phase and is the world's most complex biomaterial. It is an essential component of life support systems because it offers a wide range of assistance that benefits human well-being in several multitudinal areas. Different trends affect soil properties at various scales, and it is important to consider them to ensure that they are accurate. In another way, soil is a volatile and heterogeneous ecosystem with complicated trophic interactions that contain diverse species of microbes [1]. In general, due to the ambiguous nature of the soil, any investigation of its interior architecture has been limited. Furthermore, due to recent temperature and anthropogenic changes; soil properties, including pH, have been drastically altered [2]. The pH of the soil is used to determine its acidity and alkalinity, which is linked to numerous soil characteristics, including hydrolysis and ion equilibrium [3], microbial communities [4] and organic matter configuration [5]. It is known as the "principal indicator" of soil chemistry because it influences soil materials' solubility and decides accessibility to plants and biological motion, directly or indirectly. As a result, pH regulation is critical for both environmental and agronomic management [6].

1.1. What is pH

The pH is a crucial component of nutrient availability since it is a “negative logarithm” of hydrogen ion activity/concentration in the soil-water cycle [7]. The prevalent phenomenon in the soil is hydrogen ion activity. At high pH values, the hydrogen ion concentration is low and is vice-versa [8]. A logarithmic pH scale is used when hydrogen ion concentration ranges over a broad range; with a pH decrease of 1, the acidity increases by 10. The pH scale varies between 0-14 [9] and differentiates the soil types with different pH ranges worldwide. For instance, the pH value of ordinary soil ranges from 3.5 to 9 and in precipitated areas ranges from 5 to 7 and in dry regions varies between 6.5 to 9 [10]. Soil pH, also known as soil reaction, is a measurement of the acidity or alkalinity of the soil. The pH scale ranges from 0 to 14, with pH 7 designated as neutral. The soil pH reduces as the volume of hydrogen ions in the soil increases, making it more acidic.

1.2. How pH is Changed

Acidity and alkalinity develop depending on the source of addition, with all cations representing the soils base pH level and all anions representing the soils acid pH level. The soil characteristics, including pH, are significantly altered by vigorous agriculture and climatic changes [2, 11]. For instance, rainwater leaches out essential ions (magnesium, calcium, sodium and potassium) from soils, and carbon dioxide (CO_2) from decomposing organic matter (OM) and root respiration dissolved in soil water to create a thin organic acid. In addition, strong organic and inorganic acids such as sulfuric acid, carbonic acid and nitric acid are formed when organic matter decays, and ammonium and sulphur fertilizers are oxidized. The activities of these heavy inorganic and organic acids typically results in highly acidic soils and vice versa. Since some primary and secondary macronutrients are essential and acidic, the unequal distribution of these nutrients in the soil directly impacts soil pH. Furthermore, the cation exchange capability (CEC) is directly influencing soil pH [12]. Negative charges on soil colloids help develop the CEC of the soil, and changes in negative charge cause the CEC of the soil to fluctuate dramatically [13]. An increase in negative charges on soil particles (allophones, organic colloids, sesquioxides, and 1:1 forms of silicates) is due to an increase in pH, which also helps raise soil CEC vice-versa [14].

1.3. Effect of pH on Plant Nutrient Sources

Each pesticide, organic and inorganic fertilizer source has its own set of requirements for proper use. Therefore, urea, phosphate, and potassium fertilizers, among all micronutrients, are mainly added to the soil, which is also pH-dependent. Other processes, such as nitrogen (N) cycling, N fertilizer application, significantly reduce soil acidity [15-17]. Simultaneously, soil pH, when approaching an unacceptable level, can become inactive or fail to degrade as predicted [18], posing problems for the crop growth cycle. The pH of the soil solution, both high and low, directly impacts nutrient absorption and on some critical micronutrients, especially zinc [19-21]. The external acidic compounds in the soil may induce acidification or give a rise in soil acidity due to a reduction in the capacity of the soil to neutralize the acid. As a result, certain ecosystem features are affected, often negatively [16, 17]. Acidification of soil occurs in many environments due to acid deposition in the atmosphere. It is caused mainly by anthropogenic emissions of acidic gases such as nitrogen oxides and sulphur dioxide [15]. Soil pH changes may affect biogeochemical processes as well as the role and structure of terrestrial ecosystems [22]. It also affects enzymatic activity and organic matter [23]. In terms of minimum nutrient uptake, however, changes in pH directly impact crop growth, efficiency, and yield.

1.4. Possible Solution: a Way Out

All the organic and inorganic additional sources almost fail to maintain the neutral soil pH level or near to the neutral due to various reasons, e.g. texture. As a result, microbial approaches could be a viable alternative for maintaining pH in the rhizosphere based on plant needs and a cost-effective transition toward agricultural sustainability. Genetically engineered microbes and hydrogen oxidizing bacteria that can release hydrogen ion that can take part in pH alteration must first be practised to achieve our goal: food security and sustainability.

1.5. Role of pH in Microbial Growth

Microbes are widespread in natural environments, ranging from hot springs to deep aquifers in the natural habitats and commonly supported by the microbes in ocean floors [24]. They modify many biogeochemical cycles ranging from global carbon cycling and redox reaction to weathering [25]. A wide variety of environmental factors such as temperature, nutrient supply, salinity and pH regulate their metabolism [26]. Among all these factors, pH has a profound influence [27]. The pH is the indication of managing microbial communities, their activities and composition [23]. Microbes are classified into three groups: alkaliphilic grow fastest above pH 9; acidophiles grow best at pH <5, and neutralophiles grow optimally at pH between 5 and 7 [28]. One unit increase or decrease in the pH reduces the microbial growth up to 50 percent [29, 30].

1.6. Microbial Efficiency towards Neutral soil pH

Plants do not live independently, but they still have dynamic relationships with microbes [31]. Plants allow the microbes (fungi, archaea and bacteria) overall their tissue and the subsequent accumulation of microbes is called a phyto-microbiome [32]. Various organic acids such as malic acid, gluconic acid, citric acid, oxalic acid, tartaric acid, lactic acid, and succinic acid are produced by microbial biota in which both anions and cations serve as chelating agents. Anions trap positively charged ions (Ca^{+2}, Al^{+3}, and Fe^{+3}) present in the soil [33]. Plant roots, organic matter decomposition and bacteria may be the cause of acidity in the soil. Previous studies confirm that microbes are the primary cause of soil organic acid production, and therefore the problems associated with the formation of organic acids are becoming important [34]. From a broader spectrum of an ecosystem, the organic acid concentration varies between 0 to 50 µM for tri or dicarboxylic acids such as tartaric acid, citric acid, oxalic acid, malic, and succinic acid.

In contrast, these concentrations vary greatly, ranging from 0 to 1 mM in monocarboxylic acids, including formic, valeric, lactic acid, acetic acid, propionic acid, and butyric acids [35]. However, it should be focused that these concentrations are highly variable based on the soil composition, organic matter degradation, root exudates and microbes. Microorganisms, including bacteria, fungi, and lichen species, contain large organic acids [36-38].

1.7. Microbial involvement in Soil Acidification

Soil acidity is an essential characteristic of soil chemical properties that can affect ecological functions and processes such as nutrient supply by regulating soil desorption and sorption reactions and soil microbial population properties [39, 40]. It deals with a higher concentration of heavy metals and other cations in soil, although soil acidity is a complex of elevated proton concentrations and interactions with different mineral ions. Various microbes can suppress acidic soil conditions, take the pH level closer to neutral, and species growing in extreme soil environments have attracted a lot of attention due to their unique ecology and morphology. Acidophile is a term used to describe organisms living in highly acidic environments with shallow pH levels. Acidophiles (AP) are those species that can survive and often flourish in an acidic environment with a pH of 1 to 5 and are eukaryotes, bacteria, and archaea that can be present in a multitude of acidic conditions such as geysers and sulfuric lakes, acid mine drainage fields, and even in our stomachs [40]. Higher acid levels typically kill the microbial cells and the AP; on the other hand, they have evolved many specialized pathways to sustain a constant internal cellular pH. Microbes respond by two mechanisms; "passive" control, which does not require the cell to expend energy, and "aggressive" regulation, which does require the cell to pay energy [41]. The AP microorganisms are resistant to potentially toxic compounds because of elevated levels of heavy metals in normal acidic conditions. *A. ferrooxidans* is a chemolithoautotrophic strain capable of growing at very low pH (pH 1-2) and high metal concentrations. The primary goal of passive pH regulation defence is to protect the cell membrane from the unfavourable environment. Some microorganisms produce a biofilm to delay the diffusion of molecules through the cell, while others may alter their cell membrane to insert protective substances like fatty acids [42]. Microbes also secrete buffer molecules that increase pH, which is another essential way they passively control their pH. Some have already developed active pH modulation, which enables them to continuously pump hydrogen ions out of their cells. They are able to maintain an internal pH of 6.5 to 7.0 by doing so.

1.8. Microbial Involvement in Soil Alkalization

Microorganisms that can survive under alkaline conditions are divided into groups, i.e. Alkaliphiles, also known as “alkalophiles”, and others known as alkali tolerant. Microorganisms that require an alkaline medium for their survival and growth are termed “alkaliphiles”. This term is originated from the Arabic word “alkali”, which means soda ash and “phile” means loving. The last two units of pH (above the neutral pH) are considered optimal pH levels for the growth of alkaliphiles. Alkalitolerants are those organisms that are capable of growing when the pH is > 9.0 or 10, and their optimal growth rates lie near neutral or more negligible pH [43]. Soil alkalinity is less stable and localized highly, so it can be noticed that microbes face challenges for their survival under such conditions. For the optimum growth of alkaliphiles, a pH level above 9.0 is favourable or maybe often 10-12. But when the pH is neutral, they cannot grow, or perhaps they grow slowly. Alkaline soils are shared globally; the pH range that lies under alkaline soils is 10 or maybe above [44]. In alkaline soil conditions, many cyanobacteria species are abundant (*spirulina spp.* and *chromatium spp*). They can provide organic matter for vast groups of some other heterotrophs. Some process encourages alkaliphiles growth because they raise pH level by decomposition of different proteins and urea hydrolysis that release ammonia in higher concentration, resulting in pH increment. In normal soils, number of alkaliphiles is less than the number found in soils with the alkaline condition [45, 46]. Alkaliphilic bacteria have gained much attention because of extracellular enzymes and their biochemical properties, i.e. alkaliphilic and alkali stability [47]. Their survival is possible by maintaining their activities with a proton-transfer system in the cytoplasmic membrane, the ATP pump and sodium-proton antiproton [46]. Species that can inhibit extreme environments, just like higher salinity or alkalinity conditions, are “*Phytoactinopolyspora*” [48, 49]. Plant growth-promoting bacteria can produce some derivative enzymes, organic acids, and secondary metabolites that facilitate soil nutrients mineralization and ensure their availability for sequestration under such stress conditions of alkalinity [50]. Plant growth-promoting bacteria of *Bacillus* genera can lower the pH of soils. Also, their electrical conductivity is significantly reduced in contrast to the control in which PGBR are not inoculated. They increase dehydrogenase, phosphatase and beta-glycosidase activities under alkaline soil condition [51]. It was noticed that a strain “NBRI YN4.4” could survive at a higher pH level of 11. In some earlier studies, bacterial strain can tolerate alkaline condition stress even at 8.0-13 pH levels [52, 53]. The *Aspergillus niger* can produce organic acids. Many studies have shown its dependency on ambient pH at 5.0 to 8.0; oxalic acid production is efficient, and when the pH is below 3.0, it is absent completely [54]. Gluconic acid is produced optimally when the pH level is

5.5; it is also found at other pH levels from 2 through 8 [55]. Production of citric acid begins at pH level 3, and its optimum level is below [56].

1.9. Mechanism of Innovative Microbial Approach

The fluctuation of pH in the rhizosphere could be a suitable phenomenon that increases or decreases the pH by several folds in the rhizosphere. The microbial approach can be beneficial if managed properly. In this technique, the collected microbes (that release organic acids or essential compounds) may be multiplied through genome transferring until their characteristics become too acidic or indispensable in the rhizosphere to help to alter soil pH where nutrients are readily available to the plants. The microbial community collected from different sites, alkaline and acidic medium can be helpful for this purpose because every bacterium has its characteristics collected from various locations. Furthermore, in recent years, this view has helped answer some common evolutionary concerns regarding how bacteria, along with their host species, have evolved from their early ancestors. It is vital to consider how to plant tolerance has been affected by their encounters with microbes, although much remains unclear.

1.10. Genetically Engineered Microbes

Over the world, before the creation of genetically engineered microorganisms, we need to find the gene of interest from the incredible biodiversity of microorganisms in the soil. Maybe the influence on pH associated with a particular consortium of different organisms but not only a single strain. First, the screening method for desired microbial activity needs to be developed, which allows performing preliminary detection.

1.11. Object of Interest

The fundamental goal of this approach is to identify a new generation of microbes that is better suited to changing the pH of the rhizosphere where excess nutrients are required for plant uptake. Combining SSU rRNA or rDNA sequences with fluorescent oligonucleotide probes offers an effective method for researching soil microorganisms that aren't amenable to current culturing techniques [57].

1.12. Microbial Activity we are Looking towards and Microbial Profiling Method

Microbes that modify the pH from acidic to basic to neutral by releasing appropriate compounds should be chosen. It is conceivable that by altering the genes of microbes by genetic modifications, more resilient strains of microbes can be created with the potential to change the pH up to many folds. Technological progress is at the core of microbial ecology in the research area. The throughput of DNA sequencing has dropped dramatically in the last decade, making it possible for most study groups to map microbial population diversity in environments of interest. Tiny subunit rRNA genes are amplified from soil-extracted nucleic acids and are one of the most valuable approaches. These methods may be used to identify and analyses soil microbes that can't be cultured right now. Microbial rRNA genes can be detected and sequenced directly from soil samples. The genomes of these microorganisms will then be compared to those of other recognized microorganisms. Microbiome community profiling (MiCoP) is also a suitable method for profiling eukaryotes and viruses in metagenomic samples [58].

1.13. Other Microbial Processes

The metabolism of microbial populations changes in a variety of ways. Acidophilic microbes *Thiobacillus acidophilus* (a form of bacteria), *Vorticella* (a type of eukaryote), and *Crenarchaeota* (a classification of archaea) secrete essential compounds in the soil solution to keep the pH near neutral [59]. They are resistant to salinity and other abiotic stresses. Similarly, alkaliphilic microbes (*Thiohalospira alkaliphila*) release acidic compounds to get the pH closer to neutral and are susceptible to stress by various mechanisms [60]. For example, the added sulphur is converted into hydrogen sulphate or sulfuric acid by specific microbes, which aids in lowering the pH of the soil [61].

2. CONCLUSION

Changing the pH of the soil has a direct impact on the essential nutrients and, as a result, the growth and yield of crops. Various methods are used to solve these problems, but it is a complex phenomenon whereby the breeding of microbes may be a suitable option. Microbes release organic acids that take part in much of the physicochemical processes and make the soil ecological system strong. Future crop production may entail more breeding for pH stress resistance and introducing microbial technologies that have improved tolerance to pH stress. Comparing current and previous genes characteristics by these microbes should be checked through experiments. To selectively breed the microbes for optimal nutritional interactions for plants, the genetic components of this trait must first be established.

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