



A REVIEW

Morphological study of potassium solubilizers

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Abstract : Potassium is an elementary macronutrient besides Nitrogen and Phosphorus for growth of plants. It performs crucial metabolic activities like photosynthesis, activation of enzyme and protein synthesis. Rhizospheric microorganisms are the key segments of viable agricultural ecosystems. These perform a vital character by resolving the unobtainable Potassium form and make accessible for roots of plants. Potassium concentrations which are soluble in the soil are generally less though above 90% Potassium present in the soil subsist in the form of insoluble rock. Solubilization of a certain configuration of K is done by effective K-solubilizers treatment to an obtainable structure of K in the soil. The obtained form of K should be simply absorbed by the plant. Some selected bacterial and fungal isolates based on their morphological characteristics were assessed for their potentiality to resolve K from insoluble K trace. The selected isolates were in the form of rods, Gram positive and motile. This review assays on the morphological study of K-solubilizers for their remarkable supremacy in forming zone of solubilization.

Key Words : Potassium solubilizers, Isolates, Macronutrients, Gram positive, Morphological

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INTRODUCTION

Potassium has a key impact on the growth of plants as it is an essential macronutrient. Other than stimulating plant resistance to pests, diseases and abiotic stresses, Potassium assays a chief performance in translocating the carbohydrates, helps in photosynthesis process and maintains stability among monovalent cations and divalent cations (Brar *et al.*, 2004). The chief components of potassium are present as the insoluble rock form in the

soil like feldspar, illite and micas (Goldstein, 1994). Soil microorganisms avail minerals from soil and perform a great impact on soil fertility and also in cycling of ion (Lian *et al.*, 2010). Capability of soil microorganisms to solubilize unobtainable potassium by excreting organic acids which dissolve rock of potassium to fetch the K back to the solution (Bennett *et al.*, 1998 and Maurya *et al.*, 2014). K solubilizing microorganisms like *Bacillus edaphicus*, *Bacillus circulans*, *Pseudomonas*, *Bacillus mucilaginosus* liberate potassium in get-at-able from

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soil (Sheng *et al.*, 2008; Basakand Biswas, 2012, Rajawat *et al.*, 2012 and Singh *et al.*, 2010). Microbes solubilize Potassium by increasing crop by treatment of potassium rock which could be convenient agriculturally (Rajan *et al.*, 1996). Generally, soils in India with higher K content intensify availability of Potassium for crop production. However, availability of potassium in some soils is inadequate to meet greater potassium requiring crop like tomato, soybean, potato. So that implementation of KSB to boost the crop production. Studies defining potentiality of potassium solubilizers are obtainable in India (Dutta, 2010 and Bahadur *et al.*, 2016).

During green revolution, introducing high-yield hybrid crop varieties of crop was a specific reason that soils lack macronutrients which include potassium. Furthermore, level of Potassium in soil depleted due to soil erosion, leaching resulting potassium inadequacy becoming chief hindrance for crop production (Sheng and Huang, 2002). Thus, crop responding to potassium availability in the soil. However, lack of potassium has also become a major cause in many crop plants (Meena *et al.*, 2014 and Xiao *et al.*, 2017).

To subdue this predicament and to assure greater crop production, growers are largely incorporating chemical fertilizers in soil (Glick, 2012). Although the chemical fertilizers performed great role to attain higher plant growth and yield but they have been maleficent to health of soil (Adesemoye and Kloepper, 2009). Application of such chemical fertilizers at higher limit for increasing production, create severe soil as well as environmental pollutions (Brady, 1990 and Akande *et al.*, 2008).

Potassium rock can be beneficial in greater extent by direct implementation and applying soluble potassium instead of chemical fertilizers to secure environment. Anyhow treatment of complex minerals needs presence or integration of their solubilizers (Rajan *et al.*, 1996).

To reduce the pollution caused by chemical fertilizers, organic agriculture system has come out as a relevant drive internationally for extended health of soil. Thus, Nitrogen-fixing bacteria along with PGPR (plant growth promoting rhizobacteria), Potassium-solubilizing and Phosphate solubilizing bacteria are treated as biofertilizers to decrease the threats of toxicity to environment (Vessey, 2003 and Bahadur *et al.*, 2014).

Estimation of K solubilization potential of isolates:

Aleksandrov agar medium is used to examine the

isolated potassium solubilizers qualitatively taking the zone of solubilization Ratio or index of solubilization (Hu *et al.*, 2006). Solubilization efficiency and index are assessed by taking the Ratio of total diameter *i.e.* (halo zone-colony diameter)/2. (Edi-premono *et al.*, 1996).

$$\text{Solubilization efficiency (SE)} = (D-d)/2 (> 5\text{mm})$$

$$\text{Solubilization zone ratio/Solubilization index (SI)} = D/d$$

where, D = Diameter of zone of clearance (colony/ Mycelium + Clear zone), d = Diameter of colony/ Mycelium + growth zone

Morphological characterization:

Morphological characterization is done by analyzing the morphology of colony, Gram staining, shape of cell of chosen isolates (Bartholomew and Mittewer, 1950). Colony attributes of isolates like margin, elevation, colour, pigmentation, shape and size are studied. (Fig. 1) displays Potassium solubilization by different isolates Aleksandrov agar medium.

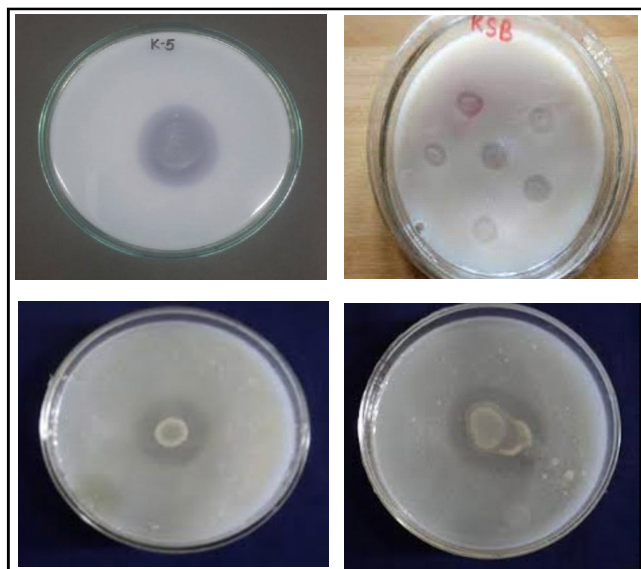


Fig. 1: Potassium solubilization by different isolates on Aleksandrov agar medium

Bacillus:

Species of Bacillus bacteria belongs to prokaryotic group of microorganisms *i.e.* they are devoid of any distinct nucleus, nuclear membrane and any membrane bound organelles. From the life form point of view, Bacillus are aerobic bacteria and strictly requires oxygen for their growth. The cell size usually ranges from 1-2 micro meter in length and 3-4 micro meter in width. They

are rod like structure, forms endospore and are basically gram positive. However, some species of *Bacillus* shows the characterization of gram negative with the increase in lifetime. Each cell consists of one dormant, tough and non-reproductive spore *i.e.* endospore. These endospores are resistant to abiotic conditions like rise in temperature, excess cold and biotic factors like disinfectants.

The external side of the cell consists of the cell wall that acts as the hurdle between the species and the surroundings. This wall is composed of teichoic acids. Cytoskeleton is a structure which is responsible for the generation of shape in bacteria. The bacteria mostly prefer to live in solitary condition. But in some can form the groups like two members in one group called as diplobacilli, bacteria clustered in chains known as streptobacilli and many more. The unique feature involves the presence of distinct junction among the group member's membrane. Regarding the shape they may be straight like a line or may be semi-curved. The cells are devoid of any capsule. The bacteria move with the help of peritrichous flagella which is found all over the body surface.

Acidithiobacillus:

It is the rod-shaped bacteria with the circular ends. From lifeform point of view, it can exist in several forms starting from acidophile to obligate aerobe and sometimes even chemolithotroph. It is medium sized bacteria with the size ranging from 0.5-1 micrometer in diameter and less than or equal to 1 micrometer in length. They exist in the environment either in solitary form or colonial form consisting of 2-3 members. The flagella present on the polar end of the body helps in their movement. The bacteria do not retain the crystal violet color so belongs to gram negative group. The unique feature is the presence of mucous micro capsule surrounding the cell.

Paenibacillus:

These bacteria can be isolated from any ecological system like terrestrial or aquatic ecosystem. They can grow in presence or absence of the oxygen. They form the non-reproductive spores known as endospores. The spores are oval and to some extent bigger than the vegetative spores. They are generally rod shaped and medium sized bacteria with the diameter around 0.5 micrometer and length ranging from 1-2 micrometer. The cells are devoid of capsule. Motility occurs by peritrichous flagella. They are staining variable *i.e.* they

can be gram positive or gram negative depending on the species.

Gram staining of potassium solubilizing bacteria :

Gram staining is regarded as the most common method to outline the difference between two groups of bacteria based on their cell wall components. The principle includes that the bacterial group that gives violet color on staining belong to gram positive and those give red color on staining are included under gram negative. The procedure of gram staining is carried out in three steps: Staining with crystal violet, decolorization and counterstain with safranin.

The bacteria belonging to potassium solubilizer may be gram negative or gram positive based on the ability to stain the crystal violet. Species of *Bacillus* and *Acidithiobacillus* are gram negative. On the other hand, species of *Paenibacillus* can be variable ranging from gram positive to gram negative. The former bacteria are not able to keep the crystal violet stain after decolorisation. The peptidoglycan layer in such bacteria much narrow and present between the inner and outer plasma membrane. The later one can retain the crystal violet colour. They have the characteristics of presence of wide peptidoglycan layer.

Gram reaction and microscopic examination :

By following Hucker's modified method, Gram staining of the inoculants are executed (Rangaswami,

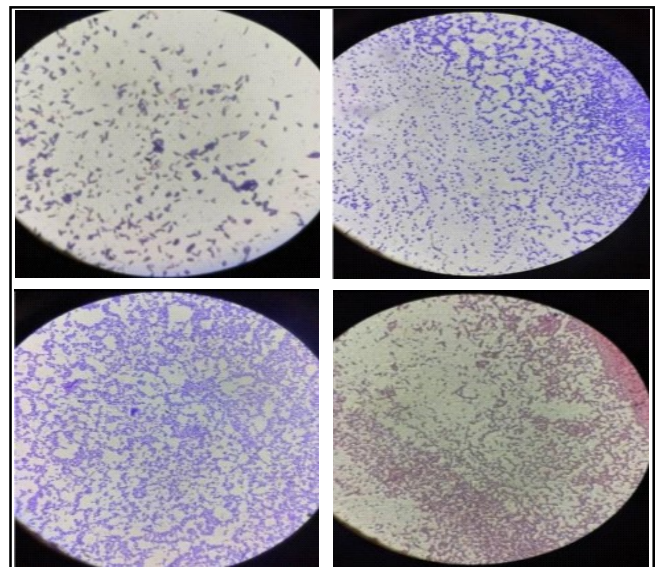


Fig. 2: Microscopic characteristics and gram staining of different K solubilizing isolates

1975). Sterilized distilled water drop to be held on middle of each slide followed by shifting of bacterial suspension to sterilized distilled water drop. On each slide, a very thin film is set up. For setting of the film, it is required to move it over the gentle flame for 2-3 times. Crystal violet solution is needed for the immersion of the slides and let to stay for 30 seconds and clean ed properly. Then the slides are dipped in iodine solution for 60 seconds and properly rinsed with 95% alcohol for 10 seconds. Gentle tap water stream is used to ringe alcohol thoroughly. Covering of the slides with safraninis done for one minute. Then the slides are washed properly with distilled water and let the dried air pass over it. The cellular morphology and the Gram reaction of inoculants are inspected under microscope (Fig. 2) shows microscopic characteristics and gram staining of different K solubilizing isolates.

Test for sporulation :

Some metabolically passive bacteria have the capacity to change into quiescent structures. These quiescent structures are called endospores as they are formed within the cells. These structures are exceptionally resistant to heat, radiation, chemicals and other agents those are generally noxious to the organisms. A single bacterium generates a single spore by sporulation process. Sporulation takes place during unfavourable conditions or by dwindling of essential nutrients.

5 ml quantities of test tubes is required to allocate the medium of spore induction. The medium is decontaminated and also inoculation is done with 0.1 ml each of 48 hour test culture. Incubation period of the medium is for 7 days at room temperature and 0.1 ml of each culture broth is drawn. The smear is formulated in each slide. The film is moved over flame to dry. The slide is held over a beaker and 5% malachite green is added drop wise. By addition of more malachite green, boiling of the malachite green can be avoided. After with drawing the slide from the stream, it is cleaned very mildly with tap water. Preparation staining is done with solution of safranin for one minute and cleaned properly with moderate stream of tap water. The preparation is put under immersion lens with immersion oil.

Conclusion :

The isolated potassium solubilizers were characterized in the terms of the morphological parameters like cell shape, gram reaction, arrangement, pigmentation, shape, margin, size, texture, elevation,

consistency and opacity. Application of the potassium fertilizers in agriculture at present needs a greater vision that makes them exorbitant for the farmers of developing nations. Since chemical fertilizers are not economical and most soils lack in plant-available potassium, so that scientists have to find out ways for humankind as well as environment. As a cost-effective replacement for exorbitant chemical fertilizers, it is very much essential to produce natural potassium resources accessible to crops. It will be a pivotal consequence to figure out the other effective rhizosphere competent bacteria (RCB) or soil microorganisms responsible for higher potassium-solubilizing capacity by assuming the above points. Now, It is very much clear that applying KSB is an emerging procedure to solubilize the potassium resources from soil and by making it accessible to plants which enhances growth of plant and reduces the application of fertilizers.

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