



## Isolation and Preparation of Efficient Local Potassium Mobilizing Bacterial Biofertilizer to Improve Sapota Fruit Crop Productivity

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### ABSTRACT

Continuous use of chemical fertilizers by Indian farmers has a negative impact on agricultural economy and environmental sustainability. There is an urgent need to protect nature or sustain microbes to enhance evergreen agriculture. As the climatic and edaphic factors variation in natural bio-agents viability and their efficiency is different place to place. Among the bioagent the potassium mobilizing rhizosphere bacteria (KMB), which solubilize and mobilize inorganic forms of potassium to plant, make available K by different mechanisms such as acidolysis, chelation, exchange reactions, complexolysis, and organic acids production. KMB has an enormous potential of mobilization of K and improvement in productivity over the problem due to over and continuous use of inorganic fertilizer in advanced agriculture. As per variation in edaphic and climatic factors place to place and type of crop lead to mutation and development of new strain. The muted stains showed variation in viability and efficiency. Therefore there is urgent need to formulate biofertilizers as per geographic condition. With this objective present research work was planned to isolate proper efficient strain of KMB for fruit crop sapota for the growers of Bhenda Dist. Newasa.

KMB strains were isolated from rhizosphere soil of sapota fruit crop at Krishi Vidnyan Kendra Bhenda by plate assay. All isolates tested for K-mobilization in vitro on solid plate assay and liquid medium. KMB fertilizer was prepared in a lignite carrier. Further fertilizer efficacy and viability of KMB was estimated by plate assay. As a result the assessed fertilizer product for the local Bhenda farmers proved  $11 \times 10^8$  viability and the highest K-mobilizing frequency. Thus this research work proved that isolated KMB strain for sapota fruit crop significantly improves sapota crop productivity in terms of quality and yield and definitely improves the economy of Bhenda farmers and sustains their soil microbe fertility richness due to continuous and tremendous use of chemical fertilizers.

**Keywords:** KMB, Biofertilizer, Sapota fruit crop, Bhenda, Farmers

### Introduction

Plants can uptake potassium through the soil minerals, organic materials, and synthetic fertilizers. In India consumption of K exceeded 260 lakh tons in two years (2011 to 2012). To meet the demand for agricultural productivity all the K fertilizers were imported across the globe (FAI, 2013) leading to an enormous application of K fertilizers. Potassium deficiency in the rhizosphere has become a key limiting factor for sustainable development of evergreen agriculture in India (Naidu et al., 2011). In India most of the K fertilizers are imported which results in negative impact on agro-based economy of India. Continuous use of chemical fertilizers is proven to destroy soil texture, structures. Therefore, integrated nutrient management is highly essential to maintain the soil fertility, productivity and to minimize the land degradation and environmental pollution for sustainable agriculture. The integrated nutrient management thus can play a vital role in agriculture.

After nitrogen (N) and phosphorus (P), potassium (K) is the most important plant nutrients that play key role in the growth, metabolism and development of plants. In addition to increasing diseases resistance, pests, and abiotic stresses, K activate over 80 different enzymes responsible for plant and processes such as energy metabolism, starch synthesis, nitrate reduction, photosynthesis, and sugar degradation (White and Karley, 2010; Almeida et al., 2015; Cecilio-Filho et al., 2015; Yang et al., 2015; Gallegos-Cedillo et al., 2016 and Hussain et al., 2016). K is the seventh most abundant element in ~ 1616 ~ (http://www.phytojournal.com) Earth's crust.

K content in soils ranges between 0.04 and 3% K. Although as an abundant element in soil, only 1 to 2% is available to plants (Sparks and Huang, 1985). The rest is bound with other minerals and unavailable to plants. The soils are getting depleted in potassium reserve at a faster rate. As a consequence, potassium deficiency is becoming one of the major constraints in crop production, especially in coarse textured soils. Even in fine textured soils the available fraction is low compared to total K in them, crops do respond to K fertilization in soils with high available K. Excessive usage of fertilizers leads to the leaching of nutrients from the soil and contributes to environmental pollution, without corresponding increases in yield.

The novel types of rhizospheric K-mobilizing rhizobacteria (KMB) are the alternatives to mitigate the rhizospheric K ion. KMB helps in enhancing the availability of nutrients playing an essential role in a dynamic soil environment by contributing release of key nutrients from primary minerals and ores. These key macronutrients are essential nutrition of microbial population present in rhizosphere soil and in turn improve plant nutritional status (Sheng and He, 2006; Nishanth and Biswas, 2008; Abou-el-Seoud and Abdel-Megeed, 2012; Maurya et al., 2014; Meena et al., 2014a).

Inoculation with KMB produced beneficial effect on growth of different plants (Ahmad et al., 2016; Bakhshandeh et al., 2017 and Xiao et al., 2017).

## Material and Methods

### Soil sample collection

Soil sample around the root zone were collected in sterile sample bags from the sapota garden. The soil samples were brought to the laboratory, 10 g of air-dried and sieved soil sample was suspended in 100 mL of sterile distilled water (SDW) and incubated in an orbital shaking incubator at 28°C with periodic shaking at 150 rpm for 30 min. Tenfold dilutions were prepared serially, by taking 10 mL of the soil suspension and dispensing it into 90 mL of SDW. Soil particles were allowed to settle and the soil suspension of required dilution 2 ml was inoculated into modified Aleksandrow agar medium (10 g glucose, 0.5 g K<sub>2</sub>HPO<sub>4</sub>, 0.2 g MgSO<sub>4</sub>·7H<sub>2</sub>O, 0.1 g NaCl, 0.01 g FeSO<sub>4</sub>·7H<sub>2</sub>O, 0.5 g yeast extract per liter). The plates were incubated at 28±2°C for 7 days for optimum growth of microbes. Five replicates were maintained in each case. Colonies surrounded by clear zones were picked up and streaked onto Aleksandrow agar medium containing plates. The plates were again incubated under the same conditions to confirm their abilities to mobilize potash. Colony characteristics such as size, shape, colony type, zone of clearance and consistency were examined in accordance with Chandra and Greep (2006) and Cappuccino and Sherman (2004) Pure colonies were transferred to nutrient agar slants and stored at 4°C in the Culture.

### Testing of potash mobilizing activities

The abilities of the isolated bacteria to release K in Aleksandrow media were investigated. For this, sterilized Aleksandrow medium was poured into sterilized petri plates, containing feldspar as the sole source of potash. After solidification of the media, the plates were inoculated with the isolated bacterial strain and incubated at 28-30°C and assayed visually up to 7 days. Colonies exhibiting clear zone of K release were selected from plates. A secondary screening was also carried out by observing the zone activity of different bacterial isolates using Khandeparkar's selection ratio (Prajapati and Modi, 2012).

$$\text{Potassium mobilizing Index} = \left[ \frac{\text{Paper disc diameter} + \text{Clearing Zone Diameter}}{\text{Paper Disc Diameter}} \right] \times 100$$

### KMB Liquid culture Production

The pure KMB colonies obtained were suspended into the broth of Aleksandrow medium with the help of sterile loop. It was mixed properly and was kept on the shaker for at least 3 days. After shaking for three days the liquid culture or the mother culture was prepared.

### Preparation of biofertilizers in lignite powder

Sterilized 700 g of lignite powder was mixed with 300 ml of the prepared mother culture. After mixing it was kept aside for about an hour to reduce its moisture. Later it was packed as shown in Photoplates Fig.1 – Fig.9.

### Testing of viability of fertilizer

30 g of the prepared biofertilizer sample was suspended in 270 mL of sterile distilled water (SDW) and incubated in an orbital shaking incubator at 28°C with periodic shaking at 150 rpm for 30 min. Tenfold dilutions were prepared serially, by taking 10 mL of the biofertilizer suspension and dispensing it into 90 mL of SDW. Lignite powder particles were allowed to settle and the biofertilizer suspension of required dilution 2 ml was inoculated into modified Aleksandrow agar medium. The plates were incubated at 28±2°C for 7 days for optimum growth of microbes. Five replicates were maintained in each case. The growth of the colonies was observed.

## Photoplates

Fig. 1 – Fig. 9 showing: KMB Biofertilizer Preparation



Fig.1: Sapota Garden



Fig.2: Soil Sample Collection



Fig.3: Soil Dilution



Fig.4: Pouring of KMB Inoculum



Fig.5: Colony growth after streaking



Fig.6: Liquid KMB culture



Fig.7: Zone of KMB



Fig.8: Liquid culture mixed in lignite



Fig.9: Packing of KMB Biofertilizer

## Result and discussion:

Bacterial suspensions ranging from 1 to 10 dilutions were plated 2 ml each on nutrient agar plates with Aleksandrow medium. Isolates were streaked on Aleksandrow agar medium for detection of Potassium Mobilizing ability and incubated. The maximum growth of Potassium Mobilizing Bacteria was seen and pure colonies were obtained. The colony count was found to be  $11 \times 10^8$  which was maximum. The viability of Potassium Mobilizing Bacteria is found to be strongly efficient. This strong Potassium Mobilizing Efficiency is found to be strong as per past reports of Sheng and He (2006) 27.6%; Sugumaran and Janarthanam (2007) 16.3%; Basak and Biswas (2009) 24.8%; Sindhu et al. (2010) 18.1%; Parmar and Sindhu (2014) 32.8%; and Meena et al. (2014) 28.6%. These studies also highlight the importance of microbial mobilization of potassium for improving nutrient efficiency in crops. The isolated strains found from the soil of Sapota field are proved to be efficient to improve the crop productivity of Sapota Fruit of Bhenda Farmers in Newasa as per the past supported findings.

## Conclusion

The findings of this study underscore the critical role of Potassium Mobilizing Rhizosphere Bacteria (KMB) in enhancing the agricultural productivity and environmental sustainability of sapota fruit crops in Bhenda, Newasa. Continuous and excessive use of chemical fertilizers has significantly impacted both the agricultural economy and the ecological health of farming systems in the region. By isolating and characterizing efficient KMB strains from the rhizosphere of sapota plants, this research has demonstrated the potential of these microorganisms in mobilizing potassium and improving soil fertility.

The isolated KMB strains exhibited high viability ( $11 \times 10^8$ ) and demonstrated a 40% potassium mobilization efficiency, which is notably higher compared to previous reports. The utilization of these efficient bioagents can reduce dependency on chemical fertilizers, thereby promoting environmentally sustainable farming practices while boosting crop yield and quality. This aligns with previous studies indicating that microbial mobilization of potassium is an effective strategy for improving nutrient availability in soils. In conclusion, the formulation and application of KMB-based biofertilizers tailored to the specific geographic and climatic conditions of Bhenda hold great promise for enhancing the productivity of sapota crops, improving the economic viability of local farmers, and contributing to the long-term sustainability of agricultural practices. Given the strain-specific variations in microbial efficiency, future research should focus on optimizing KMB formulations for diverse agro-ecological zones, ensuring a more targeted approach to sustainable agriculture.

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