



INFLUENCE OF K- SOLUBILIZING BACTERIA ON RELEASE OF POTASSIUM FROM WASTE MICA

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Received: 25.07.2013

Revised accepted: 18.09.2013

ABSTRACT

Keywords:

Waste mica,
Morphological,
Potassium,
Solubilization

Potassium is vital component of plant nutrition package limiting crop yield and quality that performs a multitude of important biological functions to maintain plant growth, Isolation of potassium solubilizers was carried out using feldspar (insoluble potassium) from the soil samples of Shahanshapur, on Aleksandrov's agar medium. From the 20 bacterial isolated, 7 bacterial isolate were selected which exhibiting highest potassium solubilization on broth culture and characterized on the basis of cultural, morphological characteristics. Solubilization of potassium from the waste mica minerals by the selected bacterial strains resulted to the action of different organic acids.

INTRODUCTION

Potassium is one of the three essential plant nutrients that perform a multitude of important biological functions to maintain plant growth, grain quality, protein synthesis, enzyme activation and photosynthesis (Read *et al.*, 2006). During last decades main focus was aimed with potent environmental impact on use of nitrogen and phosphorus and the issue of sustainable management of potassium in soil has partly been ignored. About 90 to 98% of K in soil exists in form of insoluble K-minerals such as feldspar and mica (McAfee, 2008) which constitute a biggest reservoir of soil K and very little of its potassium becomes available to plants. Soil microorganisms play a central role in nutrient cycling and influence the availability of soil mineral nutrients (Bin Lian *et al.*, 2010; Meena *et al.*, 2013). Potassium solubilizing bacteria (KSB) have capacity to dissolve potassium from insoluble K minerals (Alexander, 1985). India ranks fourth in consumption of K-fertilizers in world (FAI, 2007). However, totally depends on its import due to lack of good quality of reserve K- minerals required for commercial K-fertilizers production and leads to expense of huge amount of foreign currency. India is fortunate enough to have the world's largest deposit of mica mines distributed in Munger district of Bihar and Koderma as well as Giridih districts of Jharkhand.

During the dressing of raw micas, about 75% waste mica is generated which contains 8-10% total K and dumped near the mica mines (Biswas, 2011). These waste mica materials can effectively be used as a source of potassium in agriculture, if K their in is solubilized biologicaly through potassium solubilizing bacteria. This study was under taken with aim to assess the impact of efficient K solubilizing

bacterial isolates on release of K from waste mica and to see their morphological characteristics *in vitro* condition.

MATERIALS AND METHODS

Waste mica was obtained from Koderma mica mines of district Jharkhand, India. Flaks of mica were ground in a Wiley mill and passed through 2 mm sieve for its further use. Seven efficient K solubilizing bacterial isolates emerged out from screening test were used in this study. One mL of three days old KSB isolates suspension grown in culture tubes were plated on Aleksandrov agar medium (5g glucose, 0.005 g MgSO₄.7H₂O, 0.1 g Fe Cl₃, 2.0g Ca CO₃, 3.0 Mica (2.0 g in original media), 2.0 g calcium phosphate and 15 g agar/L) inverted plates were incubated at 28±2 °C in BOD incubator. After 7 days of incubation, potassium solubilizing bacterial colonies showing plaques was taken for their morphological characteristics i.e. color, margin, elevation, zone of solubilization optical density, slime production, shape and Gram reaction. Release of K by the isolates was studied in Aleksandrov broth containing waste mica. Two mL of overnight grown broth culture of each isolate was transferred in to conical flask containing 50 mL of Aleksandrov broth in nine replicates and inoculated in BOD at 28±2°C. Three conical flasks under each isolates were used to estimate the amount of potassium released by the isolates at 7, 14 and 21 days of incubation and compared with uninoculated mica. Inoculated broth was centrifuged at 10,000 rpm for 10 minutes to separate the supernatant from the cell growth and insoluble waste mica and filtered through Whatman No.1 filter paper. There after content of K in filtrate was determined flame photometrically (Jackson, 1973). The same

culture filtrate was used to determine the change in pH of broth culture by pH meter. Data related to effect of different isolates on change in pH of broth and releases of K from waste mica were analyzed employing CRD with factorial concept (Chandel, 1978).

RESULTS AND DISCUSSION

Morphological characteristics of KSB

All the isolates produced slime though magnitude of its production varied from low to high according to their capability (Table 1). This observation is in conformity to the findings of Sugumaran and Janarthanam (2007) who have also reported the slime production by KSB isolates. Isolates showed entire smooth margin except the I₁₄ which had

undulated margin. I₁₂ and I₁₅ showed slightly elevated colonies while highly raised colonies by remaining isolates were observed. On Aleksandrov agar medium, colony formed by most of the isolates appeared to be translucent except colony of I₁₄ which appeared as opaque. Among the 7 isolates, 4 isolates were gram +ve and remaining 3 isolates were gram -ve rods. Colonies of most of the isolates were white to creamy in appearance except the colonies of I₁₂ which were grayish in color. The diameter of zone of solubilization caused by the isolates at seven days of growth ranged from 0.8 to 1.4 cm. Data on zone of solubilization revealed that I₁₅ which procured from rhizosphere of *Cajanus cajan* formed lowest zone of solubilization compared to isolates procured from cereals.

Table 1 Source and morphological colony characteristics of K-solubilizer isolates on Aleksandrov agar medium

Isolates	Crop	Color	Margin	Colony elevation		Zone of solubilization (cm)	Optical Density		Slime production	Shape
				Slightly raised	Highly raised		Translucent	Opaque		
I ₁	Maize	White	Entire	-	+	1.2	+	-	High	Rod
I ₈	Maize	White	Smooth	-	+	1.2	+	-	Low	Rod
I ₁₀	Wheat	Creamy	Smooth	-	+	1.4	+	-	High	Rod
I ₁₂	Maize	White	Smooth	+	-	1.0	+	-	Low	Rod
I ₁₄	Wheat	Creamy	Rough	-	+	1.2	-	+	Low	Rod
I ₁₅	Arhar	Creamy	Smooth	+	-	0.8	+	-	Medium	Rod
I ₁₇	Maize	Creamy	Smooth	-	+	1.4	+	-	High	Rod

Periodical change in pH

pH of the uninoculated mica broth did not vary much from its initial value during the incubation. All the isolates caused significant decrease in pH of broth when compared with uninoculated mica broth and obvious differences were recorded among them (Table 2). Significant decrease in pH due to inoculation of different isolates was accordance to the incubation periods. Isolates produce organic acids in broth which resulted in lower down the pH during the incubation period. This finding is in agreement with the result of Girgis *et al.* (2008): Binbin and Bin (2011): Stillings *et al.* (1996) who have reported that the bacterial isolates produce several organic acids i.e., oxalic, fumaric, citric and tartaric which cause decrease in pH of the growing medium. Lowest mean value of pH such as 6.35, 5.16 & 4.52 were recorded with I₁₄, I₁₇ & I₁ at 7, 14 & 21 days of incubation, respectively. Zone of solubilization was negatively and significantly correlated with pH of the broth ($r = 0.63^*$). I₁₇ exhibited lowest value of broth pH and maximum zone of solubilization. However, the I₁₀ which had equal zone of solubilization as I₁₇ showed comparatively greater pH of broth. Isolates contribute more in decrease of broth pH in comparison to days of incubation. However, Isolates and period of inoculation were significantly interacted in lowering the pH of broth.

Periodical K-solubilizing capacity

Very less increase in K content in mica broth was observed that might be due the structural disturbance in mica caused by hydrolysis which resulted to release of K in broth (Walther, 1997). K solubilization of K was significantly influenced by inoculants, incubation periods and their interactions. Capacity of K solubilization tended to increase with incubation period (Table 2). All the isolates had significantly greater K solubilization capacity in comparison to uninoculated mica broth though they varied in their capacity to release the K from mica. This release of K may be due to the production of different kind of organic acids by the isolates. This fact is in agreement with the findings of Prajapati and Modi (2012) who have also reported that K solubilizers produce organic acids and cause reduction in pH. No any definite relation in pH of broth and release of K by the isolates was observed. However, positive correlation exists in between pH of the broth and release of K from waste mica. Release of K was greatly influenced by the incubation period in comparison to isolates. Maximum K solubilization capacity ($17.33 \mu\text{g K mL}^{-1}$) was recorded with I₁₅ at 21 days of incubation which interacted significantly with other isolates. An average, minimum $7.28 \mu\text{g K mL}^{-1}$ was recorded with isolate no I₁₀.

A perusal of the data reveals that pH of the broth medium was lower with I₁₀ than I₁₅. However, just reverse trend was recorded in release of K from mica. Probably, I₁₀ is aluminophilic which released aluminium from aluminium sheet of mica and hydrolysis of this released aluminium gave H⁺ to broth. Thus pH of the broth is decreased greatly but accordingly K⁺ was not released in solution as it is bound with silica sheet of mica. On the other hand, I₁₅ is silicophilic

which chelated silica of the silica sheet of mica which caused comparatively greater amount of K⁺ in broth without much influencing the pH of medium. This fact is in conformity to the finding of Liu *et al.* (2006) and Friedrich *et al.* (1991) who have also reported dissolution of K from Potassic mineral by silicate solubilizing bacteria at acidic to alkaline pH in liquid medium.

Table 2 Effect of K-solubilizer on periodic change in pH and release of potassium from waste mica (Initial broth pH=7.34)

Treatments	pH				Potassium (µg m/L)			
	7 days	14 days	21 days	Mean	7 days	14 days	21 days	Mean
Mica broth	7.24	7.22	7.22	7.23	2.58	3.08	3.10	2.92
Mica broth + isolate ₁	6.53	5.85	5.42	5.93	6.59	10.52	10.82	9.31
Mica broth + isolate ₈	6.61	5.91	5.18	5.90	8.48	10.52	13.15	10.72
Mica broth + isolate ₁₀	6.92	5.60	4.52	5.68	5.76	6.95	9.14	7.28
Mica broth + isolate ₁₂	6.50	5.49	4.56	5.52	7.80	8.36	9.60	8.59
Mica broth + isolate ₁₄	6.35	5.78	5.05	5.72	8.63	10.49	11.77	10.30
Mica broth + isolate ₁₅	6.63	5.95	5.30	5.96	8.95	13.13	17.33	13.14
Mica broth + isolate ₁₇	6.59	5.16	5.04	5.49	7.48	10.75	12.50	10.24
Mean	7.62	6.70	6.04	-	8.048	10.54	12.49	-
		CD. (P=0.05)				CD. (P=0.05)		
Isolate		0.31				1.21		
Days		0.07				0.74		
Isolate× Days		0.48				1.82		

CONCLUSION

Thus, observations and results of the present study indicate that colonies of K solubilizer are creamy to hyaline, raised and slimy. Microscopically they are rods of varying Gram reactions. They decrease pH of the growing medium. Decrease in pH is not only the mechanism of solubilization of K from Potassic mineral. This also depends on nutritional nature of the K- solubilizers. I₁₅ procured from legume emerged out as the best K solubilizing bacterial isolate which can be used as K biofertilizers.

REFERENCES

- Alexander. M. 1985. Introduction to Soil Microbiology. John Wiley and Sons Inc., New York, USA, 382-385.
- Bin, L., Bin, W., Mu, P., Congqiang, L. and Teng, H.H. 2010. Microbial release of potassium from K bearing minerals by thermophilic fungus *Aspergillus fumigatus*. *Geochimica Cosmochimica Acta*. 72: 87-98.
- Binbin, M.O. and Bin, L. 2011. Interactions between *Bacillus mucilaginosus* and silicate minerals (weathered adamellite and Feldspar): Weathering rate, products, and reaction mechanisms. *Chinese Journal of Geochemistry*. 30: 187-192.
- Biswas, D.R. 2011. Nutrient recycling potential of rock phosphate and waste mica enriched compost on crop productivity and changes in soil fertility under potato-soybean cropping sequence in an Inceptisol of Indo-gangetic plains of India. *Nutrient Cycling Agroecosystem*. 89: 15-30.
- Chandel, S. R. S. 1978. A hand book of agricultural statistics. Published by A.S. Chandel, Achal Prakashan Mandir, Parmat, Kanpur.
- FAI. 2007. Fertilizers Statistics 2006-2007. The Fertilizer Association of India, New Delhi.
- Friedrich, S., Platonovo, N. P., Karavaiko, G. I., Stichel, E. and Glombitza, F. 1991. Chemical and biological solubilization of silicates. *Acta Biotechnology Journal*. 11:187-196.
- Girgis, M. G. Z., Khalil, H.M.A. and Sharaf, M.S. 2008. *In Vitro* evaluation of rock phosphate and potassium solubilizing potential of some *Bacillus* strains. *Australian Journal of Basic and Applied Science*. 2(1): 68-81.
- Jackson, M. L. 1973. Soil chemical analysis. Prentice Hall India Pvt. Ltd., New Delhi.
- Liu, W., Xu, X., Wu S., Yang, Q, Luo ,Y. and Christie, P. 2006. Decomposition of silicate minerals by *B. mucilaginosus* in liquid culture. *Environmental Geochemistry and Health*. 28: 133-140.

- McAfee, J. 2008. Potassium, a Key Nutrient for Plant Growth. Department of Soil and Crop Sciences <http://jimmacafee.tamu.edu/files/potassium>.
- Meena, V.S., Maurya, B.R. and Verma, J.P. 2013. Does a rhizospheric microorganism enhance K⁺ availability in agricultural soils. *Microbiological Research*. Doi.org/10.1016/j.micres.2013.09.003.
- Prajapati, K.B. and Modi, H. A. 2012. Isolation and characterization of potassium solubilizing bacteria from ceramic industry soil. *CIB Tech Journal of Microbiology*. 1: 8-14.
- Read, J.J., Reddy, K.R. and Jenkins, J.N. 2006. Yield and quality of upland cotton as influenced by nitrogen and phosphorus. *European Journal of Agronomy*. 24: 282-290.
- Stillings, L.L., Drever, S.L., Brantley, Y.S. and Oxburgh, R. 1996. Rates of feldspar dissolution at pH 3-7 with 0-8 mM oxalic acid. *Chemical Geology* 132: 79-90.
- Sugumaran, P. and Janarthnam, B. 2007. Solubilization of potassium containing minerals by bacteria and their effect on plant growth. *World journal of Agricultural Sciences*. 3 (3): 350-355.
- Walther, J.V. 1997. Comment and reply: Feldspar dissolution at 25°C and low pH. *American Journal of Science*. 297: 1012-1032.