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EFFECT OF TEMPERATURE ON PHOSPHATE DISSOLUTION EFFICACY OF MICROORGANISMS ISOLATED FROM THE COLD DESERT HABITAT OF *POPULUS ALBA* (WHITE POPLER) IN TRANS HIMALAYAS

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ABSTRACT

Amongst a wide array of elements required by the plants for their growth, phosphate happens to be the second most important element. However, the degrading availability of soil and the costly chemical fertilizers lead to a shortage of adequate phosphorous supply to the plants. Phosphate Solubilising microorganisms are capable of solubilising inorganic phosphate from the insoluble deposits thus making phosphate readily available for plants. In the present research, Phosphate solubilising microorganisms isolated from the trans Himalayan habitat were tested for their abilities to solubilise various sources of phosphates at different temperature ranges (9°, 12°, 18°, 24°, 35° and 40°C). Strains of both bacteria PBC1 (*Bacillus* sp.), PBC2 (*Bacillus* sp.), PBC5 (Unidentified) and Fungi PFC3 (*Penicillium* sp.), PFC5 (*Aspergillus fumigatus*) and PFC6 (*Aspergillus niger*) were employed to solubilise the phosphate against the varied samples obtained from different habitats, the dessert of Udaipur, Rajasthan subtropical highland climate of Mussoorie, Uttarakhand and humid subtropical climate of North Carolina. Out of these Phosphate solubilises amongst bacteria PBC2 (*Bacillus* sp.) was solubilising maximum insoluble phosphate while amongst Phosphate solubilising fungi, PFC6 (*A. niger*) was reported to be highly efficient. The maximum Phosphate solubilisation was observed while solubilising TCP followed by NCRP, MRP and URP, clearly indicating that NCRP has the highest efficacy in making phosphorous dissolve into the soil environment. This potential of the PSB isolated from NCRP can thus be further employed to make phosphorous readily available to the plants as an alternative to chemical fertilizers at varied temperatures and different climatic conditions, habitat.

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INTRODUCTION

In alkaline soils, the availability of Phosphorus from Rock phosphate (RP) is very less. Moreover, the anions of Phosphate are rapidly immobilised after their application in soil and hence become unavailable to plants (Narsian and Patel, 2000). Chemical fertilizers are very expensive and cause a lot of soil environmental pollution. Thus, there is a need to find the possibility of other alternative source to dissolve rock phosphate so as to provide phosphate in available form to plants. Plant growth promoting microbial formulations in the form of P solubilising microbes (PSM) can overcome this

problem by dissolving these insoluble products in eco-friendly manner. The efficiency of these beneficiary microbes depend on various abiotic factors such as temperature, pH and salt concentration of the system. Important genera of Phosphate solubilising bacteria are *Bacillus*, *Pseudomonas* and *Micrococcus*. Certain strains of Rhizobia can also solubilise both organic and inorganic phosphates (Abd-Alla, 1994). Filamentous fungi such as *Aspergillus* and *Penicillium* can also solubilise phosphates under in vitro conditions (Seshadri et al, 2004; Wakelin et al, 2004). The solubilisation of inorganic phosphates in soils is due to acidification, chelation and exchange reactions (Fox and Comerford, 1990; Vazquez et al, 2000). *Populus Alba* (White Popler) is an important medicinal plant in Lahaul and Spiti valleys. It is used as anti-inflammatory, antiseptic and as a tonic.

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Table 1. Effect of temperature on tricalcium phosphate solubilisation ($\mu\text{g/ml}$) by bacterial isolates in Pikovskaya and NBRIP broth in *Populus alba*

Bacteria	Temperature ($^{\circ}\text{C}$)											
	9		12		18		24		35		40	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	28.5 \pm 0.500	30.2 \pm 0.264	37.2 \pm 0.251	39.1 \pm 0.208	40.2 \pm 0.321	41.4 \pm 0.458	44.1 \pm 0.100	49.4 \pm 0.450	40.2 \pm 0.251	42.7 \pm 1.24	16.2 \pm 0.251	18.2 \pm 0.200
PBC2 (<i>Bacillus</i> sp.)	39.6 \pm 0.556	41.4 \pm 0.264	50.6 \pm 0.793	54.4 \pm 0.608	62.4 \pm 0.458	65.5 \pm 0.814	70.3 \pm 0.321	75.2 \pm 0.321	54.7 \pm 0.818	56.4 \pm 0.585	27.6 \pm 0.793	30.8 \pm 1.00
PBC5 (Unidentified)	32.5 \pm 0.173	35.0 \pm 0.100	46.3 \pm 0.321	47.2 \pm 0.264	55.2 \pm 0.200	57.2 \pm 0.378	58.3 \pm 0.493	61.4 \pm 0.458	47.3 \pm 0.321	49.2 \pm 0.321	21.2 \pm 0.305	22.3 \pm 0.550

Table 2. Effect of temperature on pH of filtrate in bacterial isolates in Pikovskaya and NBRIP broth in *Populus alba*

Bacteria	Temperature ($^{\circ}\text{C}$)											
	9		12		18		24		35		40	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	6.22 \pm 0.020	6.18 \pm 0.017	6.17 \pm 0.023	6.14 \pm 0.015	6.15 \pm 0.010	6.10 \pm 0.005	6.10 \pm 0.005	6.08 \pm 0.011	6.11 \pm 0.005	6.08 \pm 0.015	6.30 \pm 0.005	6.25 \pm 0.015
PBC2 (<i>Bacillus</i> sp.)	6.12 \pm 0.010	6.08 \pm 0.015	6.07 \pm 0.010	6.03 \pm 0.010	6.01 \pm 0.015	6.00 \pm 0.005	5.98 \pm 0.010	5.95 \pm 0.011	6.01 \pm 0.015	5.99 \pm 0.015	6.17 \pm 0.020	6.15 \pm 0.010
PBC5 (Unidentified)	6.20 \pm 0.005	6.18 \pm 0.005	6.15 \pm 0.010	6.12 \pm 0.015	6.09 \pm 0.010	6.05 \pm 0.011	6.02 \pm 0.020	5.99 \pm 0.015	6.08 \pm 0.010	6.01 \pm 0.015	6.24 \pm 0.010	6.19 \pm 0.015

Table 3. Effect of temperature on rock phosphate solubilisation ($\mu\text{g/ml}$) by bacterial isolates in Pikovskaya and NBRIP broth in *Populus alb*

Bacteria	Rock phosphate	Temperature ($^{\circ}\text{C}$)											
		9		12		18		24		35		40	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	NCRP	5.2 \pm 0.251	5.9 \pm 0.152	7.7 \pm 0.152	8.3 \pm 0.351	10.1 \pm 0.152	11.2 \pm 0.300	17.0 \pm 0.115	18.9 \pm 0.208	9.0 \pm 0.100	10.2 \pm 0.321	2.7 \pm 0.1	3.2 \pm 0.251
	MRP	5.1 \pm 0.100	5.43 \pm 0.152	5.4 \pm 0.2	5.8 \pm 0.152	9.2 \pm 0.251	10.1 \pm 0.360	14.2 \pm 0.305	16.2 \pm 0.251	8.4 \pm 0.152	9.2 \pm 0.251	2.5 \pm 0.1	2.8 \pm 0.115
	URP	2.2 \pm 0.251	2.8 \pm 0.152	3.4 \pm 0.152	4.2 \pm 0.251	3.9 \pm 0.152	5.0 \pm 0.152	9.6 \pm 0.200	10.2 \pm 0.251	6.1 \pm 0.152	6.7 \pm 0.230	1.4 \pm 0.057	1.9 \pm 0.200
PBC2 (<i>Bacillus</i> sp.)	NCRP	13.5 \pm 0.115	14.0 \pm 0.152	23.2 \pm 0.321	25.3 \pm 0.472	30.2 \pm 0.264	32.1 \pm 0.500	35.9 \pm 0.251	38.0 \pm 0.100	26.2 \pm 0.2	26.9 \pm 0.450	8.1 \pm 0.100	8.7 \pm 0.230
	MRP	13.2 \pm 0.251	14.0 \pm 0.416	21.3 \pm 0.300	23.2 \pm 0.264	28.4 \pm 0.400	30.7 \pm 0.378	32.2 \pm 0.200	34.4 \pm 0.264	25.3 \pm 0.305	26.3 \pm 0.208	8.1 \pm 0.321	8.3 \pm 0.152
	URP	9.33 \pm 0.208	10.2 \pm 0.251	13.5 \pm 0.100	14.2 \pm 0.200	19.2 \pm 0.251	19.8 \pm 0.321	28.4 \pm 0.360	29.9 \pm 0.378	15.4 \pm 0.2	16.1 \pm 0.152	6.1 \pm 0.152	7.0 \pm 0.173
PBC5 (Unidentified)	NCRP	9.1 \pm 0.100	10.2 \pm 0.251	20.1 \pm 0.141	23.7 \pm 0.568	12.2 \pm 0.264	13.5 \pm 0.351	17.1 \pm 0.100	18.3 \pm 0.300	21.2 \pm 0.251	22.3 \pm 0.305	3.2 \pm 0.321	4.1 \pm 0.305
	MRP	8.9 \pm 0.100	9.5 \pm 0.152	17.2 \pm 0.355	18.1 \pm 0.305	10.1 \pm 0.100	11.4 \pm 0.251	15.3 \pm 0.305	17.1 \pm 0.802	19.3 \pm 0.493	21.0 \pm 0.513	2.7 \pm 0.200	3.3 \pm 0.305
URP	4.2 \pm 0.321	4.9 \pm 0.305	9.4 \pm 0.173	10.2 \pm 0.305	7.2 \pm 0.200	7.8 \pm 0.230	10.4 \pm 0.360	11.8 \pm 0.208	10.2 \pm 0.251	11.8 \pm 0.208	1.6 \pm 0.152	2.0 \pm 0.152	URP

Table 4. Effect of temperature on pH of filtrate during rock phosphate solubilisation by bacterial isolates in Pikovskaya and NBRIP broth in *Populus alba*

Bacteria	Rock phosphate	Temperature (°C)											
		9		12		18		24		35		40	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	NCRP	6.60 ± 0.00	6.57 ± 0.020	6.56 ± 0.011	6.52 ± 0.020	6.52 ± 0.010	6.47 ± 0.020	6.46 ± 0.015	6.42 ± 0.025	6.49 ± 0.020	6.48 ± 0.015	6.61 ± 0.015	6.59 ± 0.005
	MRP	6.61 ± 0.005	6.58 ± 0.015	6.56 ± 0.015	6.56 ± 0.010	6.53 ± 0.011	6.48 ± 0.015	6.48 ± 0.015	6.44 ± 0.026	6.53 ± 0.035	6.52 ± 0.011	6.63 ± 0.000	6.62 ± 0.010
	URP	6.63 ± 0.015	6.60 ± 0.015	6.62 ± 0.020	6.58 ± 0.020	6.58 ± 0.010	6.54 ± 0.015	6.59 ± 0.011	6.51 ± 0.020	6.62 ± 0.025	6.61 ± 0.015	6.66 ± 0.010	6.64 ± 0.015
PBC2 (<i>Bacillus</i> sp.)	NCRP	6.53 ± 0.010	6.48 ± 0.015	6.36 ± 0.015	6.35 ± 0.017	6.30 ± 0.015	6.23 ± 0.020	6.19 ± 0.005	6.14 ± 0.020	6.29 ± 0.005	6.39 ± 0.262	6.58 ± 0.010	6.51 ± 0.015
	MRP	6.55 ± 0.015	6.51 ± 0.010	6.37 ± 0.025	6.35 ± 0.015	6.31 ± 0.011	6.28 ± 0.015	6.22 ± 0.020	6.18 ± 0.015	6.31 ± 0.015	6.28 ± 0.026	6.58 ± 0.015	6.54 ± 0.011
	URP	6.57 ± 0.011	6.52 ± 0.025	6.48 ± 0.011	6.44 ± 0.015	6.38 ± 0.020	6.33 ± 0.015	6.28 ± 0.011	6.24 ± 0.005	6.41 ± 0.015	6.36 ± 0.015	6.63 ± 0.011	6.59 ± 0.015
PBC5 (Unidentified)	NCRP	6.56 ± 0.015	6.55 ± 0.020	6.41 ± 0.005	6.38 ± 0.0152	6.31 ± 0.020	6.28 ± 0.015	6.25 ± 0.015	6.22 ± 0.023	6.34 ± 0.005	6.32 ± 0.020	6.59 ± 0.000	6.55 ± 0.015
	MRP	6.58 ± 0.020	6.57 ± 0.010	6.45 ± 0.035	6.39 ± 0.015	6.33 ± 0.037	6.30 ± 0.015	6.27 ± 0.015	6.18 ± 0.015	6.37 ± 0.015	6.36 ± 0.011	6.61 ± 0.010	6.57 ± 0.028
	URP	6.62 ± 0.020	6.59 ± 0.020	6.52 ± 0.023	6.48 ± 0.015	6.39 ± 0.015	6.37 ± 0.020	6.34 ± 0.015	6.29 ± 0.005	6.48 ± 0.017	6.42 ± 0.025	6.62 ± 0.005	6.61 ± 0.026

Table 5. Effect of temperature on tricalcium phosphate solubilisation (µg/ml) by fungal isolates in Pikovskaya and NBRIP broth in *Populus alba*

Fungi	Temperature (°C)											
	9		12		18		24		35		40	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC3 (<i>Penicillium</i> sp.)	30.4 ± 0.529	32.2 ± 0.321	61.4 ± 0.585	64.3 ± 0.550	80.5 ± 0.416	82.9 ± 0.709	86.2 ± 0.200	88.4 ± 0.665	64.4 ± 0.404	68.3 ± 0.435	26.5 ± 0.400	33.3 ± 0.305
PFC5 (<i>A.fumigatus</i>)	42.6 ± 0.568	45.4 ± 0.493	68.3 ± 0.264	73.0 ± 0.115	91.2 ± 0.251	94.5 ± 0.450	99.4 ± 0.450	100.7 ± 0.264	69.3 ± 0.264	70.4 ± 0.665	30.2 ± 0.200	34.2 ± 0.264
PFC6 (<i>A. niger</i>)	49.2 ± 0.305	54.4 ± 0.568	73.8 ± 0.264	74.6 ± 0.500	100.2 ± 0.251	105.6 ± 0.500	107.9 ± 0.655	110.3 ± 0.416	78.5 ± 0.550	80.4 ± 0.665	32.9 ± 1.05	37.5 ± 0.400

Table 6. Effect of temperature on pH of filtrate in fungal isolates in Pikovskaya and NBRIP broth in *Populus alba*

Fungi	Temperature (°C)											
	9		12		18		24		35		40	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC3 (<i>Penicillium</i> sp.)	6.14 ± 0.010	6.09 ± 0.015	5.52 ± 0.015	5.48 ± 0.015	5.25 ± 0.014	5.22 ± 0.020	5.02 ± 0.005	4.99 ± 0.011	5.20 ± 0.015	5.17 ± 0.010	6.12 ± 0.011	6.09 ± 0.005
PFC5 (<i>A.fumigatus</i>)	6.08 ± 0.005	6.05 ± 0.010	5.38 ± 0.010	5.35 ± 0.010	5.18 ± 0.011	5.14 ± 0.005	4.91 ± 0.015	4.89 ± 0.005	5.12 ± 0.015	5.09 ± 0.005	6.09 ± 0.011	6.07 ± 0.005
PFC6 (<i>A. niger</i>)	6.02 ± 0.015	5.99 ± 0.015	5.25 ± 0.011	5.22 ± 0.020	5.04 ± 0.015	5.00 ± 0.010	4.82 ± 0.000	4.79 ± 0.010	5.01 ± 0.005	4.99 ± 0.011	6.02 ± 0.005	5.99 ± 0.015

Table 7 Effect of temperature on rock phosphate solubilisation (µg/ml) by fungal isolates in Pikovskaya and NBRIP broth in *Populus alba*

Fungi	Rock phosphate	Temperature (°C)											
		9		12		18		24		35		40	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC3 (<i>Penicillium</i> sp.)	NCRP	8.1 ± 0.152	9.0 ± 0.152	21.2 ± 0.251	23.4 ± 0.416	35.3 ± 0.251	36.5 ± 0.452	51.2 ± 0.305	53.3 ± 0.321	23.4 ± 0.251	24.7 ± 0.264	4.6 ± 0.152	5.06 ± 0.057
	MRP	7.7 ± 0.152	7.9 ± 0.057	20.1 ± 0.152	23.5 ± 0.404	35.3 ± 0.264	37.4 ± 0.208	50.3 ± 0.300	50.6 ± 0.611	23.2 ± 0.251	23.5 ± 0.503	4.4 ± 0.173	4.7 ± 0.200
	URP	2.9 ± 0.152	4.6 ± 0.208	18.3 ± 0.3	19.1 ± 0.208	25.5 ± 0.351	27.2 ± 0.264	41.2 ± 0.208	43.4 ± 0.378	17.4 ± 0.208	18.0 ± 0.152	2.5 ± 0.152	2.7 ± 0.152
PFC5 (<i>A.fumigatus</i>)	NCRP	9.5 ± 0.057	9.9 ± 0.152	27.7 ± 0.251	29.2 ± 0.251	39.7 ± 0.416	42.1 ± 0.100	57.2 ± 0.100	60.2 ± 0.251	30.1 ± 0.100	31.8 ± 0.321	6.2 ± 0.251	7.0 ± 0.152
	MRP	9.1 ± 0.100	9.4 ± 0.115	25.3 ± 0.321	26.2 ± 0.251	39.2 ± 0.251	41.2 ± 0.251	54.1 ± 0.100	56.3 ± 0.208	28.2 ± 0.208	29.2 ± 0.305	6.0 ± 0.057	6.5 ± 0.200
	URP	4.6 ± 0.057	5.1 ± 0.100	20.2 ± 0.264	22.9 ± 0.624	29.3 ± 0.305	31.2 ± 0.251	43.3 ± 0.321	44.9 ± 0.378	18.2 ± 0.208	19.3 ± 0.200	3.1 ± 0.152	3.6 ± 0.200
PFC6 (<i>A. niger</i>)	NCRP	12.6 ± 0.251	13.1 ± 0.173	30.2 ± 0.264	32.7 ± 0.709	42.5 ± 0.500	43.9 ± 0.264	60.1 ± 0.100	61.7 ± 0.378	33.6 ± 0.152	35.4 ± 0.529	8.1 ± 0.152	8.8 ± 0.100
	MRP	10.4 ± 0.208	12.5 ± 0.602	28.3 ± 0.200	30.3 ± 0.152	41.6 ± 0.208	43.6 ± 0.6	59.4 ± 0.360	60.2 ± 0.305	30.1 ± 0.100	32.1 ± 0.152	8.0 ± 0.000	8.3 ± 0.503
	URP	5.9 ± 0.200	6.4 ± 0.208	22.3 ± 0.152	23.6 ± 0.550	30.2 ± 0.208	31.2 ± 0.351	45.3 ± 0.264	47.2 ± 0.251	22.2 ± 0.251	24.2 ± 0.305	4.0 ± 0.152	4.6 ± 0.152

Table 8. Effect of temperature on pH of filtrate during rock phosphate solubilisation by fungal isolates in Pikovskaya and NBRIP broth in *Populus alba*

Fungi	Rock phosphate	Temperature (°C)											
		9		12		18		24		35		40	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC3 (<i>Penicillium</i> sp.)	NCRP	6.60 ± 0.010	6.56 ± 0.010	6.50 ± 0.010	6.45 ± 0.015	6.28 ± 0.010	6.24 ± 0.015	6.02 ± 0.015	5.98 ± 0.015	6.41 ± 0.025	6.33 ± 0.015	6.62 ± 0.025	6.59 ± 0.017
	MRP	6.58 ± 0.020	6.56 ± 0.010	6.51 ± 0.010	6.47 ± 0.025	6.31 ± 0.005	6.28 ± 0.015	6.05 ± 0.015	6.01 ± 0.015	6.44 ± 0.011	6.37 ± 0.025	6.64 ± 0.011	6.62 ± 0.00
	URP	6.65 ± 0.010	6.58 ± 0.015	6.58 ± 0.015	6.55 ± 0.015	6.37 ± 0.050	6.32 ± 0.025	6.18 ± 0.010	6.08 ± 0.020	6.58 ± 0.00	6.52 ± 0.020	6.68 ± 0.010	6.62 ± 0.015
PFC5 (<i>A.fumigatus</i>)	NCRP	6.47 ± 0.010	6.44 ± 0.020	6.35 ± 0.015	6.31 ± 0.015	6.15 ± 0.010	6.10 ± 0.015	5.88 ± 0.015	5.85 ± 0.020	6.28 ± 0.005	6.22 ± 0.020	6.59 ± 0.017	6.55 ± 0.015
	MRP	6.48 ± 0.010	6.43 ± 0.011	6.38 ± 0.005	6.35 ± 0.015	6.18 ± 0.005	6.12 ± 0.020	5.92 ± 0.015	5.87 ± 0.025	6.32 ± 0.005	6.27 ± 0.025	6.61 ± 0.010	6.58 ± 0.015
	URP	6.52 ± 0.010	6.49 ± 0.011	6.39 ± 0.015	6.37 ± 0.010	6.19 ± 0.015	6.18 ± 0.020	6.10 ± 0.015	6.05 ± 0.025	6.44 ± 0.025	6.38 ± 0.015	6.65 ± 0.003	6.62 ± 0.005
PFC6 (<i>A. niger</i>)	NCRP	6.41 ± 0.011	6.37 ± 0.015	6.30 ± 0.010	6.26 ± 0.011	6.06 ± 0.010	6.02 ± 0.015	5.84 ± 0.011	5.77 ± 0.025	6.23 ± 0.010	6.19 ± 0.005	6.57 ± 0.010	6.54 ± 0.010
	MRP	6.44 ± 0.005	6.38 ± 0.015	6.33 ± 0.010	6.28 ± 0.015	6.10 ± 0.00	6.05 ± 0.015	5.89 ± 0.005	5.85 ± 0.020	6.27 ± 0.010	6.22 ± 0.025	6.59 ± 0.005	6.55 ± 0.017
	URP	6.49 ± 0.011	6.45 ± 0.015	6.37 ± 0.005	6.33 ± 0.020	6.20 ± 0.015	6.15 ± 0.020	6.08 ± 0.005	6.05 ± 0.020	6.38 ± 0.002	6.34 ± 0.025	6.62 ± 0.005	6.59 ± 0.011

¹PVK-pikovskaya agar, MPVK-Modified Pиковskaya agar, NBRIP-National Botanical Research Institute Agar
NCRP-North Carolina Rock Phosphate, MRP- Mussoorie rock Phosphate, URP- Udaipur Rock Phosphate

Plantation of this tree has been taken to avoid erosion and degradation in the fragile mountainous system in these valleys. The present paper reports to study the efficacy of different PSMs to solubilise various sources of phosphates at different temperature regimes (9°, 12°, 18°, 24°, 35° and 40°C).

MATERIALS AND METHODS

The highly efficient strains of Phosphate solubilising bacteria (PSB) viz. PBC1 (*Bacillus* sp.), PBC2 (*Bacillus* sp.), PBC5 (Unidentified) and Phosphate solubilising fungi (PSF) viz. PFC3 (*Penicillium* sp.), PFC5 (*Aspergillus fumigatus*) and PFC6 (*Aspergillus niger*) were isolated from the soils of Kukumseri and Udaipur of Lahaul and Spiti Valley of Trans Himalayan region of Himachal Pradesh.

Phosphate Dissolving Efficiency of Culture Isolates

- 10⁶ bacterial cells and 3X10⁶ fungal spores/ml were inoculated in 100 ml Pikovskaya (PVK) and National Botanical Research Institute (NBRIP) broth supplemented with tri calcium phosphate (TCP) (18.5% P₂O₅).
- The microbes were incubated for 3 days in case of bacteria and 6 days for fungi under shake at 250 rpm at different temperature ranges (9°, 12°, 18°, 24°, 35° and 40°C). Uninoculated broth served as control.
- The solubilised P was determined in clear filtrate using Ascorbic acid method (Watanabe and Olsen, 1965).
- The intensity of blue colour was measured on spectrophotometer at 730 nm and the quantity of solubilised phosphorus was expressed as µg/ml. The final pH of culture filtrate was also determined.
- The highly efficient PSM were further tested for their efficacy to solubilise other complex sources of Phosphate viz North Carolina Rock Phosphate (NCRP), Mussoorie Rock Phosphate (MRP) and Udaipur Rock Phosphate (URP) in

Pikovskaya (PVK) and National Botanical Research Institute (NBRIP) broth respectively at different temperature ranges (9°, 12°, 18°, 24°, 35° and 40°C).

RESULTS AND DISCUSSION

Three bacteria and three fungal isolates were tested for their capabilities to solubilise TCP at various temperature ranges. A diverse level of P dissolution was observed at higher temperature of 35° and 40°C. The dissolution potentials of microbes were found to be highest at 24° C temperature. The bacterial isolate PBC2 (*Bacillus* sp.) solubilised maximum TCP (75.2 µg/ml in NBRIP and 70.3 µg/ml in PVK broth) (Table 1) and also represented maximum fall in pH of filtrate (Table 2). The solubilisation capacity of this isolate was at par at 18° C temperature. These bacterial isolates were further tested for their efficacies to solubilise NCRP, MRP and URP at different temperature regimes. PBC2 (*Bacillus* sp.) was reported to solubilise maximum NCRP (38 µg/ml in NBRIP and 35.9 µg/ml in PVK broth) followed by MRP (34.4 µg/ml in NBRIP and 32.2 µg/ml in PVK broth) and URP (29.9 µg/ml in NBRIP and 28.4 µg/ml in PVK broth) at 24° C (Table 3). The highest decrease in pH was also observed by this isolate while solubilising NCRP (6.57 µg/ml in NBRIP, 6.17 µg/ml in PVK) followed by MRP and URP (Table 4). Out of three PSFs, PFC6 (*A. niger*) solubilised maximum TCP (110.3 µg/ml in NBRIP and 107.9 µg/ml in PVK broth) at 24° C followed by 18° C and 35° C. Out of three fungal isolates, PFC6 (*Aspergillus niger*) was found to be highly efficient at 24° C (Table 5) While testing their efficacies to dissolve three different kinds of rock phosphates, PFC6 (*A. niger*) solubilised maximum NCRP (61.7 µg/ml in NBRIP and 60.1 µg/ml in PVK broth) followed by MRP (60.2 µg/ml in NBRIP and 59.4 µg/ml in PVK broth) and URP (47.2 µg/ml in NBRIP and 45.3 µg/ml in PVK broth) at 24° C (Table 7). The fall in the pH of filtrate was observed at 24° C in all the fungal isolates. PFC6 (*A. niger*) showed maximum decrease in pH of filtrate (5.77 in NBRIP and 5.84 in PVK broth) while solubilising NCRP followed by MRP (5.85 in NBRIP and 5.89 in PVK broth) and URP (6.05 in NBRIP and 6.08 in PVK broth). The fungal isolates were observed to be the best P solubilisers than the bacterial isolates.

Conclusion

This work will result in development of technology, package for mass multiplication and field delivery application of different phosphate solubilizers for restoring vegetation in varied temperature areas. These plant growth promoters will find practical utility in federal and non-government agencies involved in agricultural programmes.

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