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Effect of pH on inorganic Phosphate solubilisation by microorganisms isolated from Trans Himalayan Region of Himachal Pradesh

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Abstract

Various Phosphate solubilising microorganisms (PSM) were isolated from the natural habitats of *Populus alba* from Lahaul and Spiti valleys of Himachal Pradesh. The PSB isolated belonged to genera *Bacillus* and *Micrococcus* while the PSF isolated frequently belonged to species of *Aspergillus* and *Penicillium*. The isolates with high efficiency for tricalcium phosphate solubilisation, were further tested for their capacities to solubilise Mussoorie Rock Phosphate (MRP), Udaipur Rock Phosphate (URP) and North Carolina Rock Phosphate (NCRP) at different pH ranges. The bacterial isolate PBC2 (*Bacillus*) was reported to be highly efficient under alkaline conditions while amongst fungi PFC6 (*Aspergillus niger*) was solubilising maximum insoluble phosphate under acidic conditions.

Keywords: PSM, *Populus alba*, Lahaul and Spiti Valley, pH ranges

Introduction

Phosphorus (P) plays a crucial role in various biochemical processes of plant viz. photosynthesis, respiration, cell division etc. Chemical fertilizers in the form of soluble inorganic phosphate are immobilised generally to greater extent after their introduction in soils and hence become unavailable to plants (Dadarwal *et al.*, 1997) [3]. A number of phosphate solubilising microbes (PSM) are known to produce acidic products which either by change of soil pH or by chelation of metal cations, release insoluble phosphorus in soluble form (Reyes *et al.*, 2002; Chung *et al.*, 2005; Gupta *et al.*, 2007) [9, 2, 5]. These phosphate solubilising biofertilizers have gained importance due to increase in the cost of fertilizers and environmental hazards posed by them.

Keeping all these points in view, the solubilisation efficiencies of screened microbial isolates were tested using various sources of phosphates at different pH values varying from acidic (pH 5.0) to alkaline (pH 9.0).

2. Materials and Methods

Six phosphate solubilising bacteria (PSB) viz. viz PBC1 (*Bacillus* sp.), PBC2 (*Bacillus* sp.), PBC3 (Unidentified), PBC4 (*Micrococcus*), PBC5 (Unidentified), PBC6 (*Bacillus* sp.) and seven Phosphate solubilising fungi (PSF) viz. PFC1 (Unidentified), PFC2 (*Aspergillus niger*), PFC3 (*Penicillium* sp.), PFC4 (Unidentified), PFC5 (*Aspergillus fumigatus*), PFC6 (*Aspergillus niger*) and PFC7 (Unidentified) isolated from Kukumseri and Udaipur areas of Lahaul and Spiti valleys were tested for their efficacies to solubilise tri calcium phosphate (TCP) at different pH values (5-9). Out of these three PSB and three PSF were screened to test their efficacies to dissolve various rock phosphates (RP) viz. North Carolina Rock Phosphate (NCRP), Mussoorie Rock Phosphate (MRP) and Udaipur rock phosphate (URP) at different pH values (5-9) in Pikovskaya (PVK) (Pikovskaya, 1948) [7] and National Botanical Research Institute (NBRIP) broth (Nautiyal, 1999) [6], respectively. The quantitative estimation of P dissolution by these isolates was done by following Ascorbic acid method (Watanabe and Olsen, 1965) [10]. The intensity of blue colour was measured on spectrophotometer at 730 nm and quantity of P solubilised was expressed as µg/ml. The pH of the filtrate was recorded at the end of the experiment.

3. Results and Discussion

Bacterial isolates PBC1-PBC6 were tested for their efficacies to solubilise TCP at various pH values (5-9) i.e. from acidic to alkaline range. Out of these isolates PBC2 (*Bacillus* sp.) represented maximum TCP dissolution (79.5 µg/ml in NBRIP and 76.4 µg/ml in PVK broth) followed by PBC5 (Unidentified) (69.2 µg/ml in NBRIP and 67.9 µg/ml in PVK broth) and

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PBC1 (*Bacillus* sp.) (54.2 µg/ml in NBRIP and 51.7 µg/ml in PVK broth) at pH 9 (Table 1). The highest decrease in pH of filtrate was also reported at pH 9 in PBC2 (*Bacillus* sp.) (7.76 in NBRIP and 7.79 in PVK broth) followed by PBC5 (Unidentified) (7.79 in NBRIP and 7.83 in PVK broth) and PBC1 (*Bacillus* sp.) (7.82 in NBRIP and 7.86 in PVK broth) (Table 2). These screened bacterial isolates viz. PBC2 (*Bacillus* sp.), PBC5 (Unidentified) and PBC1 (*Bacillus* sp.) were further tested for their abilities to dissolve three different types of rock phosphates (NCRP, MRP and URP) in NBRIP and PVK broths respectively. The highest RP solubilisation was observed by PBC2 (*Bacillus* sp.) while solubilising NCRP (51.2 µg/ml in NBRIP and 46.9 µg/ml in PVK broth) followed by MRP (49.0 µg/ml in NBRIP and 46.2 µg/ml in PVK broth) and URP (43.7 µg/ml in NBRIP and 42.5 in PVK broth) at pH 9 (Table 3). The maximum decrease in pH of filtrate was reported in PBC2 (*Bacillus* sp.) during NCRP solubilisation (7.8 in NBRIP and 7.9 in PVK broth) followed by MRP (7.84 in NBRIP and 7.93 in PVK broth) and URP solubilisation (7.88 in NBRIP and 7.98 in PVK broth) at pH 9 (Table 4). Fungal isolates (PFC1-PFC7) were also tested for their abilities to solubilise TCP in NBRIP and PVK broths respectively at different pH ranges (5-9). Out of these fungal isolates PFC6 (*Aspergillus niger*) was reported to be highly efficient with maximum TCP solubilisation (116.1 µg/ml in NBRIP and 112 µg/ml in PVK broth) followed by PFC5 (*Aspergillus fumigatus*) (109.7 µg/ml in NBRIP and 104.9 µg/ml in PVK broth) and PFC3 (*Penicillium* sp.) (93.4 µg/ml in NBRIP and 91.2 µg/ml in PVK broth) at pH 5 (Table 5).

The highest fall in pH of filtrate was observed by PFC6 (*A. niger*) (4.23 in NBRIP and 4.28 in PVK broth) followed by PFC5 (*A. fumigatus*) (4.26 in NBRIP and 4.31 in PVK broth) and PFC3 (*Penicillium* sp.) (4.34 in NBRIP and 4.40 in PVK broth) (Table 6). The three screened fungal isolates viz. PFC6 (*A. niger*), PFC5 (*A. fumigatus*) and PFC3 (*Penicillium* sp.) were further tested for their capabilities to solubilise three different rock phosphates i.e. NCRP, MRP and URP in NBRIP and PVK broth respectively at different pH ranges (5-9). The PFC6 (*A. niger*) isolate represented highest RP dissolution during NCRP solubilisation (67.3 µg/ml in NBRIP and 61.2 µg/ml in PVK broth) followed by MRP (59.3 µg/ml in NBRIP and 57.5 µg/ml in PVK broth) and URP (54.7 µg/ml in NBRIP and 53.2 µg/ml in PVK broth) at pH 5 (Table 7). The highest decrease in pH of filtrate was also observed in PFC6 (*A. niger*) during NCRP solubilisation (4.83 in NBRIP and 4.88 in PVK broth) followed by MRP (4.89 in NBRIP and 4.96 in PVK broth) and URP solubilisation (4.91 in NBRIP and 4.98 in PVK broth) at pH 5 (Table 8). The results indicate that the pH of a nutrient medium affects the performance of P solubilisers (Gaur, 1990) [4] and organic acids produced by these organisms chelate Calcium ions to bring P into solution (Randall *et al.*, 2001). The lower pH values and more available P were also reported due to inoculation of these P solubilisers in soil (Caravaca *et al.*, 2004) [1]. The PSB exhibited highest P solubilising abilities from neutral to alkaline values (pH 7-9) while PSF from neutral to acidic values (pH 7-5) irrespective of individual cultures.

Table 1: Effect of pH on tricalcium phosphate solubilisation (µg/ml) by bacterial isolates in Pikovskaya and NBRIP broth in *Populus alba* (Mean±SE)

Bacteria	pH Values									
	5		6		7		8		9	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	40.1 ± 0.115	43.5 ± 0.503	43.3 ± 0.305	49.4 ± 0.513	47.2 ± 0.264	49.4 ± 0.461	47.9 ± 0.152	49.1 ± 0.305	51.7 ± 0.251	54.2 ± 0.251
PBC2 (<i>Bacillus</i> sp.)	65.9 ± 0.152	69.3 ± 0.300	69.3 ± 0.264	72.5 ± 0.550	73.5 ± 0.550	79.1 ± 0.100	74 ± 0.251	78.9 ± 0.152	76.4 ± 0.513	79.5 ± 0.556
PBC3 (Unidentified)	35.1 ± 0.360	37.6 ± 0.400	36.2 ± 0.251	38.3 ± 0.305	38.3 ± 0.264	39.2 ± 0.208	41 ± 0.200	43.2 ± 0.251	44.3 ± 0.264	49.4 ± 0.378
PBC4 (<i>Micrococcus</i>)	37.2 ± 0.251	40.1 ± 0.208	41.2 ± 0.200	44.6 ± 0.400	44.8 ± 0.152	49.2 ± 0.251	47.3 ± 0.208	49.4 ± 0.493	49.2 ± 0.200	53.2 ± 0.264
PBC5 (Unidentified)	54.7 ± 0.251	57.2 ± 0.251	57.9 ± 0.152	59.4 ± 0.416	62.2 ± 0.251	66.5 ± 0.305	65.8 ± 0.321	69.4 ± 0.568	67.9 ± 0.100	69.2 ± 0.251
PBC6 (<i>Bacillus</i> sp.)	22.8 ± 0.264	23.3 ± 0.208	24.5 ± 0.513	28.6 ± 0.321	28.0 ± 0.057	31.3 ± 0.321	29.7 ± 0.519	34.5 ± 0.896	36.2 ± 0.264	38.2 ± 0.251

Table 2: Effect of pH on pH of filtrate in bacterial isolates in Pikovskaya and NBRIP broth in *Populus alba* (Mean±SE)

Bacteria	pH Values									
	5		6		7		8		9	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	4.81 ± 0.015	4.76 ± 0.011	4.96 ± 0.030	4.91 ± 0.020	5.95 ± 0.015	5.89 ± 0.015	6.91 ± 0.015	6.88 ± 0.015	7.86 ± 0.010	7.82 ± 0.020
PBC2 (<i>Bacillus</i> sp.)	4.72 ± 0.025	4.69 ± 0.005	4.81 ± 0.00	4.77 ± 0.015	5.90 ± 0.036	5.87 ± 0.015	6.82 ± 0.020	6.77 ± 0.015	7.79 ± 0.036	7.76 ± 0.005
PBC3 (Unidentified)	4.89 ± 0.025	4.80 ± 0.017	5.05 ± 0.020	5.01 ± 0.020	6.10 ± 0.015	6.03 ± 0.025	7.05 ± 0.020	7.00 ± 0.010	8.01 ± 0.010	7.93 ± 0.130
PBC4 (<i>Micrococcus</i>)	4.85 ± 0.055	4.76 ± 0.015	4.99 ± 0.005	4.96 ± 0.025	6.07 ± 0.020	6.01 ± 0.020	7.01 ± 0.011	6.99 ± 0.015	7.98 ± 0.011	7.91 ± 0.017
PBC5 (Unidentified)	4.74 ± 0.015	4.70 ± 0.011	4.92 ± 0.015	4.88 ± 0.015	5.93 ± 0.011	5.91 ± 0.010	6.87 ± 0.011	6.83 ± 0.017	7.83 ± 0.010	7.79 ± 0.010
PBC6 (<i>Bacillus</i> sp.)	4.91 ± 0.015	4.88 ± 0.015	5.22 ± 0.017	5.11 ± 0.025	6.18 ± 0.015	6.12 ± 0.020	7.13 ± 0.026	7.09 ± 0.015	8.08 ± 0.026	8.01 ± 0.015

Table 3: Effect of pH on rock phosphate solubilisation ($\mu\text{g/ml}$) by bacterial isolates in Pikovskaya and NBRIP broth in *Populus alba* (Mean \pm SE)

Bacteria	Rock phosphate	pH Values									
		5		6		7		8		9	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	NCRP	13.3 \pm 0.208	19.2 \pm 0.208	16.2 \pm 0.230	21.6 \pm 0.585	20.3 \pm 0.321	22 \pm 0.665	23.8 \pm 0.472	27.7 \pm 0.493	25.1 \pm 0.100	29.7 \pm 0.611
	MRP	10.2 \pm 0.251	13.1 \pm 0.305	14.3 \pm 0.321	17.6 \pm 0.416	16.9 \pm 0.251	19.5 \pm 0.550	22.2 \pm 0.264	24.2 \pm 0.305	24.2 \pm 0.251	26.3 \pm 0.321
	URP	3.4 \pm 0.378	7.7 \pm 0.360	6.2 \pm 0.173	8.63 \pm 0.513	9.86 \pm 0.152	12.2 \pm 0.346	17.3 \pm 0.321	19.0 \pm 0.200	19.1 \pm 0.152	22.2 \pm 0.305
PBC2 (<i>Bacillus</i> sp.)	NCRP	37.5 \pm 0.200	46.6 \pm 0.400	40.0 \pm 0.200	47.8 \pm 0.305	44.3 \pm 0.550	45.7 \pm 0.854	45.7 \pm 0.435	48.6 \pm 0.493	46.9 \pm 0.305	51.2 \pm 0.251
	MRP	33.3 \pm 0.200	37.6 \pm 0.251	38.1 \pm 0.305	41.7 \pm 0.378	41.9 \pm 0.351	44.1 \pm 0.100	44.5 \pm 0.503	46.4 \pm 0.503	46.2 \pm 0.321	49 \pm 0.200
	URP	17.1 \pm 0.305	19.5 \pm 0.404	22.5 \pm 0.556	25.2 \pm 0.230	30.2 \pm 0.264	29.4 \pm 1.92	39.2 \pm 0.251	41.3 \pm 0.351	42.5 \pm 0.416	43.7 \pm 0.802
PBC5 (Unidentified)	NCRP	17.7 \pm 0.251	21.6 \pm 0.585	18.9 \pm 0.208	23.7 \pm 0.458	22.3 \pm 0.300	25.5 \pm 0.472	25.8 \pm 0.264	28.3 \pm 0.321	26.7 \pm 0.264	31.5 \pm 0.503
	MRP	11.6 \pm 0.600	14.2 \pm 0.200	15.9 \pm 0.208	17.9 \pm 0.100	21.1 \pm 0.288	23.5 \pm 0.529	24.7 \pm 0.251	26.6 \pm 0.360	24.9 \pm 0.264	27.9 \pm 0.251
	URP	10.6 \pm 0.152	14.4 \pm 0.793	13.2 \pm 0.351	14.4 \pm 0.152	16.8 \pm 0.152	18.9 \pm 0.305	20.2 \pm 0.321	21.2 \pm 0.251	21.9 \pm 0.251	25.3 \pm 0.305

Table 4: Effect of pH on pH of filtrate during rock phosphate solubilisation by bacterial isolates in Pikovskaya and NBRIP broth in *Populus alba* (Mean \pm SE)

Bacteria	Rock phosphate	pH Values									
		5		6		7		8		9	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PBC1 (<i>Bacillus</i> sp.)	NCRP	4.90 \pm 0.010	4.84 \pm 0.011	5.12 \pm 0.023	5.06 \pm 0.025	6.12 \pm 0.025	6.06 \pm 0.017	7.29 \pm 0.015	7.12 \pm 0.025	8.01 \pm 0.010	7.99 \pm 0.025
	MRP	4.90 \pm 0.005	4.87 \pm 0.020	5.20 \pm 0.015	5.12 \pm 0.025	6.16 \pm 0.025	6.12 \pm 0.025	7.32 \pm 0.030	7.28 \pm 0.015	8.02 \pm 0.020	7.95 \pm 0.025
	URP	4.97 \pm 0.020	4.93 \pm 0.020	5.30 \pm 0.005	5.17 \pm 0.030	6.34 \pm 0.010	6.19 \pm 0.020	7.37 \pm 0.015	7.33 \pm 0.025	8.17 \pm 0.020	8.02 \pm 0.025
PBC2 (<i>Bacillus</i> sp.)	NCRP	4.80 \pm 0.020	4.75 \pm 0.025	4.88 \pm 0.011	4.83 \pm 0.010	6.00 \pm 0.011	5.96 \pm 0.025	7.02 \pm 0.030	6.97 \pm 0.015	7.9 \pm 0.020	7.8 \pm 0.020
	MRP	4.87 \pm 0.015	4.82 \pm 0.015	4.91 \pm 0.015	4.87 \pm 0.020	6.07 \pm 0.030	5.96 \pm 0.020	7.01 \pm 0.015	6.99 \pm 0.005	7.93 \pm 0.020	7.84 \pm 0.00
	URP	4.90 \pm 0.015	4.86 \pm 0.025	4.99 \pm 0.020	4.90 \pm 0.017	6.12 \pm 0.020	6.05 \pm 0.032	7.10 \pm 0.00	7.01 \pm 0.037	7.98 \pm 0.005	7.88 \pm 0.005
PBC5 (Unidentified)	NCRP	4.87 \pm 0.020	4.81 \pm 0.015	5.13 \pm 0.030	4.97 \pm 0.011	6.06 \pm 0.030	6.01 \pm 0.015	7.27 \pm 0.040	7.00 \pm 0.023	7.99 \pm 0.010	7.89 \pm 0.025
	MRP	4.88 \pm 0.005	4.83 \pm 0.011	5.15 \pm 0.026	5.02 \pm 0.035	6.10 \pm 0.020	6.05 \pm 0.010	7.27 \pm 0.015	7.22 \pm 0.025	8.06 \pm 0.020	7.87 \pm 0.015
	URP	4.91 \pm 0.011	4.89 \pm 0.00	5.23 \pm 0.035	5.14 \pm 0.045	6.18 \pm 0.020	6.12 \pm 0.005	7.32 \pm 0.030	7.02 \pm 0.020	8.10 \pm 0.020	7.91 \pm 0.025

Table 5: Effect of pH on tricalcium phosphate solubilisation ($\mu\text{g/ml}$) by fungal isolates in Pikovskaya and NBRIP broth in *Populus alba* (Mean \pm SE)

Fungi	pH Values									
	5		6		7		8		9	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC1 (Unidentified)	44.0 \pm 0.404	46.1 \pm 0.152	41.1 \pm 0.152	43.2 \pm 0.305	35.5 \pm 0.152	37.0 \pm 0.500	30.3 \pm 0.321	33.2 \pm 0.251	27.4 \pm 0.450	28.2 \pm 0.251
PFC2 (<i>A. niger</i>)	40.3 \pm 0.321	42.0 \pm 0.200	39.2 \pm 0.305	40.7 \pm 0.200	27.7 \pm 0.200	30.0 \pm 0.152	20.8 \pm 0.115	22.2 \pm 0.305	16.4 \pm 0.360	17.4 \pm 0.152
PFC3 (<i>Penicillium</i> sp.)	91.2 \pm 0.251	93.4 \pm 0.450	85.2 \pm 0.200	89.2 \pm 0.251	70.6 \pm 0.781	74.0 \pm 0.152	63.1 \pm 0.208	64.6 \pm 0.100	53.3 \pm 0.305	54.9 \pm 0.100
PFC4 (Unidentified)	72.4 \pm 0.450	76.4 \pm 0.400	69.3 \pm 0.321	71.2 \pm 0.251	58.7 \pm 0.321	59.2 \pm 0.550	50.2 \pm 0.305	53.4 \pm 0.251	47.3 \pm 0.351	50.2 \pm 0.305
PFC5 (<i>A.fumigatus</i>)	104.9 \pm 0.152	109.7 \pm 1.02	99.9 \pm 0.305	103.0 \pm 0.493	90.5 \pm 0.458	92.2 \pm 0.305	72.2 \pm 0.321	75.0 \pm 0.152	60.4 \pm 0.472	61.3 \pm 0.100
PFC6 (<i>A. niger</i>)	112.0 \pm 0.208	116.1 \pm 0.360	109.2 \pm 0.321	110.3 \pm 0.321	95.2 \pm 0.305	96.3 \pm 0.351	80.1 \pm 0.100	81.8 \pm 0.230	68.8 \pm 0.288	70.2 \pm 0.321
PFC7 (Unidentified)	67.8 \pm 0.321	68.9 \pm 0.351	63.8 \pm 0.152	65.0 \pm 0.208	50.1 \pm 0.208	51.3 \pm 0.208	39.2 \pm 0.251	40.0 \pm 0.100	35.2 \pm 0.264	38.4 \pm 0.529

Table 6: Effect of pH on pH of filtrate during TCP solubilisation in fungal isolates in Pikovskaya and NBRIP broth in *Populus alba* (Mean±SE)

Fungi	pH Values									
	5		6		7		8		9	
	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC1 (Unidentified)	4.68 ± 0.005	4.64 ± 0.015	5.13 ± 0.010	5.10 ± 0.010	6.03 ± 0.010	5.98 ± 0.015	7.72 ± 0.010	7.69 ± 0.011	8.77 ± 0.011	8.72 ± 0.010
PFC2 (<i>A. niger</i>)	4.71 ± 0.015	4.65 ± 0.011	5.27 ± 0.020	5.22 ± 0.010	6.08 ± 0.015	6.03 ± 0.015	7.78 ± 0.005	7.75 ± 0.011	8.78 ± 0.015	8.74 ± 0.010
PFC3 (<i>Penicillium</i> sp.)	4.40 ± 0.015	4.34 ± 0.011	4.71 ± 0.005	4.62 ± 0.025	4.91 ± 0.020	4.85 ± 0.011	7.59 ± 0.011	7.56 ± 0.010	8.63 ± 0.010	8.59 ± 0.017
PFC4 (Unidentified)	4.60 ± 0.005	4.57 ± 0.010	4.97 ± 0.020	4.88 ± 0.015	5.01 ± 0.015	4.95 ± 0.020	7.63 ± 0.005	7.58 ± 0.015	8.69 ± 0.015	8.63 ± 0.015
PFC5 (<i>A.fumigatus</i>)	4.31 ± 0.017	4.26 ± 0.010	4.62 ± 0.005	4.57 ± 0.020	4.75 ± 0.015	4.7 ± 0.010	7.56 ± 0.017	7.47 ± 0.020	8.58 ± 0.010	8.54 ± 0.020
PFC6 (<i>A. niger</i>)	4.28 ± 0.010	4.23 ± 0.015	4.49 ± 0.015	4.46 ± 0.010	4.61 ± 0.015	4.58 ± 0.025	7.51 ± 0.017	7.46 ± 0.010	8.57 ± 0.005	8.52 ± 0.020
PFC7 (Unidentified)	4.64 ± 0.010	4.60 ± 0.577	5.05 ± 0.015	4.98 ± 0.015	5.12 ± 0.020	5.06 ± 0.030	7.68 ± 0.010	7.64 ± 0.015	8.72 ± 0.005	8.68 ± 0.015

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

Fungi	Rock phosphate	pH Values									
		5		6		7		8		9	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC3 (<i>Penicillium</i> sp.)	NCRP	53.3 ± 0.305	58.2 ± 0.321	51.3 ± 0.264	54.3 ± 0.416	44.7 ± 0.251	46.3 ± 0.360	40.6 ± 0.208	43.3 ± 0.300	32.0 ± 0.057	34.3 ± 0.321
	MRP	49.1 ± 0.907	51.8 ± 0.208	44.1 ± 0.100	47.3 ± 0.264	39.9 ± 0.360	43.2 ± 0.305	36.9 ± 0.702	40.1 ± 0.152	29.3 ± 0.305	30.6 ± 0.305
	URP	42.2 ± 0.305	45.8 ± 0.152	34.5 ± 0.550	37.1 ± 0.115	28.7 ± 0.251	30.2 ± 0.251	22.2 ± 0.321	25.6 ± 0.404	19.4 ± 0.608	22.3 ± 0.321
PFC5 (<i>A.fumigatus</i>)	NCRP	59.6 ± 0.692	65.2 ± 0.305	58.9 ± 1.13	61.6 ± 0.208	50.4 ± 0.503	54.6 ± 0.450	44.7 ± 0.208	47.9 ± 0.305	39.4 ± 0.360	43.6 ± 0.458
	MRP	54.3 ± 0.351	56.2 ± 0.173	50.1 ± 0.152	53.4 ± 0.321	45.9 ± 0.057	48.2 ± 0.251	40.1 ± 0.100	43.2 ± 0.321	38.2 ± 0.264	41.6 ± 0.503
	URP	49.2 ± 0.264	53.2 ± 0.305	44.1 ± 0.152	46.5 ± 0.200	38.9 ± 0.173	41.4 ± 0.264	36.0 ± 0.100	38.4 ± 0.264	27.8 ± 0.321	32.9 ± 0.680
PFC6 (<i>A. niger</i>)	NCRP	61.2 ± 0.251	67.3 ± 0.300	59.9 ± 0.529	62.8 ± 0.305	54.6 ± 0.568	57.7 ± 0.360	48.6 ± 0.450	53.6 ± 0.458	44.0 ± 0.115	47.5 ± 0.300
	MRP	57.5 ± 0.351	59.3 ± 0.251	51.0 ± 0.200	54.2 ± 0.251	48.7 ± 0.288	53.4 ± 0.200	46.9 ± 0.305	49.5 ± 0.550	37.8 ± 0.264	42.3 ± 0.472
	URP	53.2 ± 0.321	54.7 ± 0.400	47.1 ± 0.230	49.1 ± 0.351	43.2 ± 0.305	44.8 ± 0.435	39.1 ± 0.360	41.9 ± 0.611	30.2 ± 0.305	32.0 ± 0.450

Table 8: Effect of pH on pH of filtrate during rock phosphate solubilisation by fungal isolates in Pikovskaya and NBRIP broth in *Populus alba* (Mean±SE)

Fungi	Rock phosphate	pH Values									
		5		6		7		8		9	
		PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP	PVK	NBRIP
PFC3 (<i>Penicillium</i> sp.)	NCRP	4.92 ± 0.025	4.84 ± 0.020	5.00 ± 0.015	4.85 ± 0.037	6.03 ± 0.030	5.98 ± 0.015	7.09 ± 0.020	7.06 ± 0.015	8.19 ± 0.00	8.15 ± 0.015
	MRP	4.98 ± 0.010	4.89 ± 0.026	5.05 ± 0.015	4.93 ± 0.010	6.09 ± 0.020	6.03 ± 0.015	7.17 ± 0.020	7.09 ± 0.020	8.21 ± 0.010	8.18 ± 0.011
	URP	4.99 ± 0.011	4.95 ± 0.010	5.08 ± 0.010	4.97 ± 0.015	6.15 ± 0.005	6.07 ± 0.020	7.25 ± 0.011	7.15 ± 0.005	8.31 ± 0.005	8.21 ± 0.010
PFC5 (<i>A.fumigatus</i>)	NCRP	4.82 ± 0.025	4.75 ± 0.026	4.87 ± 0.011	4.79 ± 0.017	5.98 ± 0.00	5.88 ± 0.015	7.07 ± 0.020	7.00 ± 0.015	8.11 ± 0.015	8.04 ± 0.020
	MRP	4.85 ± 0.011	4.78 ± 0.015	4.91 ± 0.015	4.84 ± 0.015	5.98 ± 0.017	5.89 ± 0.025	7.09 ± 0.005	7.06 ± 0.015	8.16 ± 0.010	8.08 ± 0.020
	URP	4.89 ± 0.005	4.85 ± 0.015	4.98 ± 0.005	4.89 ± 0.010	6.00 ± 0.015	5.94 ± 0.011	7.14 ± 0.015	7.09 ± 0.015	8.21 ± 0.011	8.16 ± 0.010
PFC6 (<i>A. niger</i>)	NCRP	4.88 ± 0.015	4.83 ± 0.020	4.99 ± 0.005	4.94 ± 0.005	6.01 ± 0.015	5.96 ± 0.020	7.04 ± 0.011	6.99 ± 0.015	8.09 ± 0.020	8.01 ± 0.017
	MRP	4.96 ± 0.011	4.89 ± 0.015	4.99 ± 0.020	4.88 ± 0.015	6.08 ± 0.020	5.97 ± 0.015	7.08 ± 0.015	7.03 ± 0.011	8.11 ± 0.010	8.07 ± 0.017
	URP	4.98 ± 0.005	4.91 ± 0.00	5.02 ± 0.015	4.97 ± 0.015	6.09 ± 0.00	6.02 ± 0.020	7.22 ± 0.017	7.11 ± 0.015	8.18 ± 0.005	8.11 ± 0.017

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5. References

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