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# Nutrient Release Pattern and Soil Enzyme Activities in Calcareous and Non-calcareous Soils as Influenced by PROM

**S. V. Bhosale [a], A. B. Gosavi [a*], A. B. Jadhav [a], D. H. Phalke [a] and A. V. Patil [a]**

[a] *Mahatma Phule Krishi Vidyapeeth, Division of Soil Science, College of Agriculture, Pune (MS), India.*

***Authors' contributions***

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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## ABSTRACT

A study was conducted at the Post Graduate Laboratory, Division of Soil Science, College of Agriculture, Pune to study the nutrient release pattern and soil enzyme activities in calcareous and non-calcareous soils as influenced by various levels of phosphorus using organic and inorganic sources. An incubation experiment was laid out in a factorial completely randomized design with seven treatments three replications. The study demonstrated that applying 100% $P_2O_5$ through Phosphate Rich Organic Manure (PROM) significantly improved soil nutrient status, particularly in non-calcareous soils, with the reduction in soil pH and calcium carbonate, improving organic carbon, and enhancing the availability of macronutrients (nitrogen, phosphorus, potassium). Enzyme activities, *viz.* urease, dehydrogenase and alkaline phosphatase were higher in non-

**Corresponding author: E-mail: gosaviavi@rediffmail.com;*

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calcareous soils, correlating positively with nutrient availability, particularly with phosphorus and micronutrients such as iron and manganese. Phosphate Rich Organic Manure (PROM) application was more effective than chemical fertilizers in nutrient mobilization and improving soil enzyme activity over 120 days of incubation. Strong correlations were observed between enzyme activity and micronutrient availability, especially in treatments receiving 100% $P_2O_5$ through PROM, indicating its potential to enhance both nutrient cycling and soil fertility.

*Keywords: Calcareous soil; enzyme activities; incubation; interaction and correlation; nutrient release; PROM.*

## 1. INTRODUCTION

Phosphorus is the second most essential macronutrient for plants, after nitrogen (Reddy *et al*., 2004) and is crucial for root development, nodulation, nitrogen fixation, and various physiological functions such as energy transfer, cell division, and photosynthesis. However, in calcareous and alkaline soils, phosphorus availability is limited due to its fixation with calcium, iron, and aluminium, leading to poor plant uptake. The lack of phosphorus in calcareous and alkaline soils is a major global problem. In calcareous/alkaline soils, phosphorus availability to plants is restricted by its reduced mobility in soils and higher fixation (Shen *et al*., 2011). Phosphorus solubilizing microorganisms (PSB) and soil enzymes like phosphatase play a crucial role in converting immobilized P into plant-available forms, especially in phosphorus-deficient soils. Non-traditional phosphorus fertilizers like Phosphate Rock (PR) and Phosphate Rich Organic Manure (PROM) offer sustainable alternatives to conventional chemical fertilizers by maintaining phosphorus availability for longer periods and enhancing crop productivity. PROM, in particular, has been recognized as an eco-friendly solution, approved by the Government of India, to improve phosphorus availability in calcareous soils. Despite India's significant rock phosphate reserves, only a fraction is suitable for fertilizer production. Therefore, further research on optimizing PROM's production and its effects on phosphorus availability in different soil types is essential for improving phosphorus use efficiency and sustainable agriculture.

## 2. METHODOLOGY

An incubation study was conducted in the Post Graduate Laboratory, Division of Soil Science, College of Agriculture, Pune, Maharashtra. The soil samples for the incubation study were collected from the PG Instructional Farm, College of Agriculture, Pune. The $CaCO_3$ of less than 5 % and more than 10 % were selected as non-calcareous and calcareous soils for study.

The recommended dose of fertilizer (50:75:45 kg $ha^{-1}$ of N, $P_2O_5$ and $K_2O$) of soybean (*Glycine max*) was used for this incubation study except for absolute control treatment. The recommended dose of phosphorus was applied through PROM (organic), DAP and SSP (inorganic) sources. The nitrogen and potassium were supplied through urea and muriate of potash, respectively. The PROM was prepared at the Division of Soil Science, College of Agriculture, Pune and its proximate analysis was done before the start of incubation.

**List 1. Treatments details**

| Tr.No. | Factor 'A' (Soil type) | Tr.No. | Factor 'B' (Nutrient management) |
|---|---|---|---|
| 1 | Highly calcareous soil ($A_1$) | 1 | Absolute control |
| 2 | Low calcareous soil ($A_2$) | 2 | Recommended dose of fertilizers through DAP (50:75:45 N: $P_2O_5$: $K_2O$ kg $ha^{-1}$) |
| | | 3 | Recommended dose of fertilizers through SSP (50:75:45 N: $P_2O_5$: $K_2O$ kg $ha^{-1}$) |
| | | 4 | 25 % $P_2O_5$ through PROM |
| | | 5 | 50 % $P_2O_5$ through PROM |
| | | 6 | 75 % $P_2O_5$ through PROM |
| | | 7 | 100 % $P_2O_5$ through PROM |

*Note:1. The recommended dose of soybean (50:75:45 N: $P_2O_5$:$K_2O$ kg $ha^{-1}$) was used for the incubation study. 2. The uniform dose of N and $K_2O$ was common to all the treatments except absolute control.*

**Table 1. Proximate analysis of PROM**

| Sr. No | Parameters | Unit | PROM |
|---|---|---|---|
| 1 | pH (1:10) | - | 7.18 |
| 2 | EC | (dS $m^{-1}$) | 1.74 |
| 3 | Moisture | (%) | 24.02 |
| 4 | Organic Carbon | (%) | 21.06 |
| 5 | Total N | (%) | 0.78 |
| 6 | Total P | (%) | 14.57 |
| 7 | Total K | (%) | 0.37 |
| 8 | Total Fe | (mg $kg^{-1}$) | 12.70 |
| 9 | Total Mn | (mg $kg^{-1}$) | 0.67 |
| 10 | Total Zn | (mg $kg^{-1}$) | 2.69 |
| 11 | Total Cu | (mg $kg^{-1}$) | 0.46 |
| 12 | C:N ratio | - | 24:1 |
| 13 | C:P ratio | - | 1.44:1 |

Plastic bowls of 2 kg capacity were filled with 1kg soil (2 mm) in one hundred sixty eight bowls. The calculated quantity of PROM, DAP, SSP, urea and MOP was added as per treatment. The moisture in the soil was maintained at field capacity. The set of experiments was incubated for 30, 60, 90 and 120 days in three replications using discarding method. The incubation study was conducted by using a factorial completely randomized design. (Panse and Sukhatme,1985).

## 3. RESULTS AND DISCUSSION

### 3.1 Chemical Properties of Soil

Low soil pH was observed in non-calcareous soil and with 100% $P_2O_5$ applied through PROM. The lowest pH (7.60) was recorded at 120 days with non-calcareous soil and 100% $P_2O_5$ through PROM. Organic matter played an important role in reducing the pH of both soils as negative correlation between pH and organic matter is reported by Talashilkar *et al*. (2000). Soil EC in calcareous soil was higher, with slight improvement for RDF through DAP and SSP. At 120 days, the highest EC (0.51 dS $m^{-1}$) was observed in non-calcareous soil with RDF applied through DAP, showing significant interaction effects. Interaction effects with the combination of non-calcareous soil and RDF through DAP recorded higher EC (0.51 $dSm^{-1}$) at 120 days.

Soil organic carbon was higher in non-calcareous soil, increasing from 0.71% at 30 days to 0.80% at 120 days. The higher organic carbon content was observed due to the application of 100% $P_2O_5$ through PROM, with significant interaction at 30 days only. The application of rock phosphate with organic materials in gypsiferous soils increased the organic carbon and phosphorous status of the soil. (Muhawish and Al-Kafaje, 2017). Non-calcareous soil recorded lower $CaCO_3$ than calcareous soil, decreasing from 4.12% at 30 days to 3.55% at 120 days with 100% $P_2O_5$ through PROM. Significant reductions were observed across all PROM treatments (100%, 75%, and 50%) over time.

### 3.2 Influence of PROM ON Soil Macro-nutrients

The higher available nitrogen (250.51–260.16 kg $ha^{-1}$) was recorded in non-calcareous soil than in calcareous soil (135.50–145.42 kg $ha^{-1}$) over the incubation period. Application of RDF through DAP recorded higher nitrogen levels, followed by SSP and 100% $P_2O_5$ through PROM, with non-significant interaction effects. Pandey *et al*. (2024) reported that ammonical nitrogen, the first available organic form of nitrogen for plants and microorganisms, was significantly higher in the incubation study compared to the control.

The soil available phosphorus content (21.50–25.54 kg $ha^{-1}$) was observed higher in non-calcareous soil compared to calcareous soil (12.50–15.07 kg $ha^{-1}$) over the incubation period. The highest phosphorus (28.24 kg $ha^{-1}$ at 120 days) was observed with 100% $P_2O_5$ through PROM in non-calcareous soil, showing significant interaction effects. Kumar *et al*. (2015) reported that the increased availability of phosphorus in organically amended soils results from a large reduction in phosphorus sorption.

The higher available potassium content (515.21–525.67 kg $ha^{-1}$) was observed in non-calcareous

**Table 2. Influence of soil calcareousness and PROM on periodical changes in pH of soil**

| Days ➡ | 30 | | | 60 | | | 90 | | | 120 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Levels of $P_2O_5$ ⬇** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** |
| $B_1$ | 8.21 | 7.79 | 8 | 8.17 | 7.73 | 7.95 | 8.17 | 7.73 | 7.95 | 8.15 | 7.69 | 7.92 |
| $B_2$ | 8.18 | 7.76 | 7.97 | 8.16 | 7.7 | 7.93 | 8.15 | 7.7 | 7.93 | 8.13 | 7.67 | 7.90 |
| $B_3$ | 8.19 | 7.77 | 7.98 | 8.17 | 7.69 | 7.93 | 8.17 | 7.69 | 7.93 | 8.14 | 7.64 | 7.89 |
| $B_4$ | 8.21 | 7.73 | 7.98 | 8.19 | 7.68 | 7.94 | 8.16 | 7.68 | 7.92 | 8.15 | 7.64 | 7.9 |
| $B_5$ | 8.19 | 7.72 | 7.96 | 8.16 | 7.69 | 7.93 | 8.14 | 7.69 | 7.92 | 8.13 | 7.62 | 7.88 |
| $B_6$ | 8.18 | 7.72 | 7.95 | 8.13 | 7.69 | 7.91 | 8.13 | 7.69 | 7.91 | 8.12 | 7.62 | 7.87 |
| $B_7$ | 8.17 | 7.72 | 7.95 | 8.12 | 7.68 | 7.9 | 8.12 | 7.68 | 7.9 | 8.1 | 7.6 | 7.85 |
| **Mean A** | **8.19** | **7.74** | **--** | **8.16** | **7.7** | **--** | **8.15** | **7.69** | **--** | **8.13** | **7.64** | **--** |
| Factors | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) |
| C.D. at (5%) | 0.006 | 0.011 | 0.016 | 0.006 | 0.011 | 0.015 | 0.004 | 0.008 | 0.011 | 0.008 | 0.015 | 0.021 |
| SE(m)± | 0.002 | 0.004 | 0.006 | 0.002 | 0.004 | 0.005 | 0.001 | 0.003 | 0.004 | 0.003 | 0.005 | 0.007 |

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

**Table 3. Influence of soil calcareousness and PROM on periodical changes in electrical conductivity of soil**

| Days ➡ | 30 | | | 60 | | | 90 | | | 120 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Levels of $P_2O_5$ ⬇** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** |
| $B_1$ | 0.37 | 0.35 | 0.36 | 0.39 | 0.38 | 0.38 | 0.4 | 0.43 | 0.41 | 0.41 | 0.44 | 0.42 |
| $B_2$ | 0.44 | 0.37 | 0.41 | 0.46 | 0.42 | 0.44 | 0.49 | 0.46 | 0.48 | 0.51 | 0.47 | 0.49 |
| $B_3$ | 0.43 | 0.38 | 0.4 | 0.46 | 0.43 | 0.45 | 0.48 | 0.45 | 0.47 | 0.49 | 0.48 | 0.49 |
| $B_4$ | 0.39 | 0.36 | 0.37 | 0.4 | 0.39 | 0.39 | 0.41 | 0.41 | 0.41 | 0.43 | 0.4 | 0.42 |
| $B_5$ | 0.4 | 0.37 | 0.39 | 0.41 | 0.4 | 0.41 | 0.43 | 0.4 | 0.42 | 0.44 | 0.43 | 0.44 |
| $B_6$ | 0.41 | 0.37 | 0.39 | 0.43 | 0.41 | 0.42 | 0.44 | 0.43 | 0.43 | 0.46 | 0.44 | 0.45 |
| $B_7$ | 0.41 | 0.38 | 0.39 | 0.44 | 0.42 | 0.43 | 0.46 | 0.44 | 0.45 | 0.48 | 0.46 | 0.47 |
| **Mean A** | **0.41** | **0.37** | **--** | **0.43** | **0.41** | **--** | **0.44** | **0.43** | **--** | **0.46** | **0.45** | **--** |
| Factors | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) |
| C.D. at (5%) | 0.009 | 0.017 | NS | 0.01 | 0.019 | NS | 0.01 | 0.019 | 0.028 | 0.011 | 0.02 | 0.029 |
| SE(m)± | 0.003 | 0.006 | 0.008 | 0.003 | 0.006 | 0.009 | 0.004 | 0.007 | 0.009 | 0.004 | 0.007 | 0.01 |

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

**Table 4. Influence of soil calcareousness and PROM on periodical changes of organic carbon in soil**

| **Days ➡** | 30 | | | 60 | | | 90 | | | 120 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Levels of $P_2O_5$ ⬇** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** |
| $B_1$ | 0.55 | 0.66 | 0.61 | 0.58 | 0.68 | 0.63 | 0.63 | 0.7 | 0.67 | 0.64 | 0.73 | 0.69 |
| $B_2$ | 0.67 | 0.68 | 0.68 | 0.59 | 0.71 | 0.65 | 0.65 | 0.73 | 0.69 | 0.66 | 0.75 | 0.71 |
| $B_3$ | 0.65 | 0.7 | 0.67 | 0.62 | 0.73 | 0.67 | 0.66 | 0.75 | 0.71 | 0.68 | 0.77 | 0.73 |
| $B_4$ | 0.56 | 0.71 | 0.64 | 0.64 | 0.75 | 0.7 | 0.68 | 0.77 | 0.72 | 0.69 | 0.79 | 0.74 |
| $B_5$ | 0.59 | 0.73 | 0.66 | 0.66 | 0.77 | 0.71 | 0.7 | 0.78 | 0.74 | 0.71 | 0.82 | 0.77 |
| $B_6$ | 0.61 | 0.75 | 0.68 | 0.69 | 0.79 | 0.74 | 0.71 | 0.79 | 0.75 | 0.73 | 0.84 | 0.79 |
| $B_7$ | 0.63 | 0.77 | 0.7 | 0.7 | 0.82 | 0.76 | 0.73 | 0.83 | 0.78 | 0.75 | 0.88 | 0.82 |
| **Mean A** | **0.61** | **0.71** | **--** | **0.64** | **0.75** | **--** | **0.68** | **0.76** | **--** | **0.70** | **0.80** | **--** |
| Factors | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) |
| C.D. at (5%) | 0.01 | 0.018 | 0.026 | 0.009 | 0.017 | NS | 0.009 | 0.016 | NS | 0.009 | 0.017 | NS |
| SE(m)± | 0.003 | 0.006 | 0.009 | 0.003 | 0.006 | 0.008 | 0.003 | 0.006 | 0.008 | 0.003 | 0.006 | 0.008 |

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

**Table 5. Influence of soil calcareousness and PROM on periodical changes of $CaCO_3$ in soil**

| **Days ➡** | 30 | | | 60 | | | 90 | | | 120 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Levels of $P_2O_5$ ⬇** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** |
| $B_1$ | 12.45 | 4.21 | 8.33 | 12.37 | 4.17 | 8.27 | 12.3 | 4.08 | 8.19 | 12.24 | 4.01 | 8.13 |
| $B_2$ | 12.33 | 4.07 | 8.2 | 12.23 | 4.12 | 8.18 | 11.81 | 3.98 | 7.9 | 10.75 | 3.82 | 7.29 |
| $B_3$ | 12.36 | 4.08 | 8.22 | 12.08 | 4.14 | 8.11 | 11.75 | 4.01 | 7.88 | 10.58 | 3.98 | 7.28 |
| $B_4$ | 12.28 | 4.17 | 8.22 | 11.82 | 4.09 | 7.96 | 11.33 | 3.82 | 7.58 | 10.5 | 3.77 | 7.14 |
| $B_5$ | 12.15 | 4.14 | 8.15 | 11.33 | 4.08 | 7.71 | 11 | 3.77 | 7.39 | 10.08 | 3.68 | 6.88 |
| $B_6$ | 11.91 | 4.1 | 8.01 | 11.31 | 4.06 | 7.69 | 10.75 | 3.68 | 7.22 | 10.07 | 3.57 | 6.82 |
| $B_7$ | 11.42 | 4.09 | 7.75 | 11.25 | 4.04 | 7.64 | 10.58 | 3.57 | 7.08 | 9.8 | 3.55 | 6.68 |
| **Mean A** | **12.13** | **4.12** | **--** | **11.77** | **4.1** | **--** | **11.36** | **3.85** | **--** | **10.58** | **3.77** | **--** |
| Factors | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) |
| C.D. at (5%) | 0.124 | 0.233 | 0.329 | 0.128 | 0.24 | 0.339 | 0.118 | 0.221 | 0.312 | 0.172 | 0.321 | 0.454 |
| SE(m)± | 0.043 | 0.08 | 0.113 | 0.044 | 0.082 | 0.116 | 0.04 | 0.076 | 0.107 | 0.059 | 0.11 | 0.156 |

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

**Table 6. Influence of soil calcareousness and PROM on release of nitrogen in soil**

| **Days ➡** | **30** | | | **60** | | | **90** | | | **120** | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Levels of $P_2O_5$ ⬇** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** |
| $B_1$ | 116.6 | 237.5 | 177.1 | 119 | 242.4 | 180.7 | 120.4 | 247.3 | 183.8 | 123.6 | 248.4 | 186 |
| $B_2$ | 145.5 | 265.3 | 205.4 | 146.3 | 266.3 | 206.3 | 152.2 | 262.1 | 207.2 | 159 | 268.3 | 213.6 |
| $B_3$ | 142.6 | 259.1 | 200.8 | 143.3 | 260.3 | 201.8 | 147.8 | 268.3 | 208 | 152.6 | 271.2 | 211.9 |
| $B_4$ | 126.3 | 242.4 | 184.3 | 137.8 | 247.3 | 192.6 | 139.4 | 248.4 | 193.9 | 140.4 | 253.9 | 197.2 |
| $B_5$ | 137.8 | 247.3 | 192.6 | 139.4 | 248.6 | 194 | 140.4 | 253.9 | 197.2 | 142.8 | 255.2 | 199 |
| $B_6$ | 139.4 | 248.6 | 194 | 140.4 | 254.4 | 197.4 | 142.8 | 255.2 | 199 | 143.3 | 257.1 | 200.2 |
| $B_7$ | 140.3 | 253.3 | 196.8 | 142.8 | 256.5 | 199.6 | 150.2 | 261.6 | 205.9 | 156.3 | 267 | 211.7 |
| **Mean A** | **135.5** | **250.5** | **--** | **138.4** | **253.7** | **--** | **141.9** | **256.7** | **--** | **145.4** | **260.2** | **--** |
| Factors | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) |
| C.D. at (5%) | 7.455 | 13.947 | NS | 7.409 | 13.862 | NS | 5.425 | 10.15 | NS | 4.546 | 8.504 | NS |
| SE(m)$\pm$ | 2.56 | 4.79 | 6.774 | 2.545 | 4.76 | 6.732 | 1.863 | 3.486 | 4.929 | 1.561 | 2.921 | 4.13 |

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

**Table 7. Influence of soil calcareousness and PROM on the release of phosphorous in soil**

| **Days ➡** | **30** | | | **60** | | | **90** | | | **120** | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Levels of $P_2O_5$ ⬇** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** |
| $B_1$ | 11.95 | 20.39 | 16.17 | 12.17 | 21.6 | 16.89 | 12.43 | 22.02 | 17.23 | 12.68 | 22.06 | 17.37 |
| $B_2$ | 12.67 | 21.59 | 17.13 | 13.02 | 22.45 | 17.74 | 15.71 | 25.14 | 20.43 | 16.11 | 26.25 | 21.18 |
| $B_3$ | 12.37 | 22.07 | 17.22 | 12.48 | 23.04 | 17.76 | 14.97 | 26.11 | 20.54 | 15.28 | 28.02 | 21.65 |
| $B_4$ | 12.22 | 20.68 | 16.45 | 12.25 | 21.8 | 17.02 | 12.97 | 22.06 | 17.52 | 14.03 | 22.65 | 18.34 |
| $B_5$ | 12.27 | 21.38 | 16.83 | 12.37 | 22.01 | 17.19 | 13.34 | 22.65 | 18 | 14.78 | 23.34 | 19.06 |
| $B_6$ | 12.99 | 22.18 | 17.59 | 14.28 | 23.05 | 18.67 | 15.76 | 26.18 | 20.97 | 16.31 | 28.21 | 22.26 |
| $B_7$ | 13 | 22.2 | 17.6 | 14.3 | 23.12 | 18.71 | 15.84 | 26.25 | 21.04 | 16.33 | 28.24 | 22.28 |
| **Mean A** | **12.5** | **21.5** | **--** | **12.98** | **22.44** | **--** | **14.43** | **24.35** | **--** | **15.07** | **25.54** | **--** |
| Factors | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) |
| C.D. at (5%) | 0.011 | 0.021 | 0.03 | 0.068 | 0.127 | 0.18 | 0.049 | 0.092 | 0.131 | 0.018 | 0.034 | 0.048 |
| SE(m)$\pm$ | 0.004 | 0.007 | 0.01 | 0.023 | 0.044 | 0.062 | 0.017 | 0.032 | 0.045 | 0.006 | 0.012 | 0.016 |

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

**Table 8. Influence of soil calcareousness and PROM on the release of potassium in soil**

| **Days ➡** | **30** | | | **60** | | | **90** | | | **120** | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Levels of $P_2O_5$ ⬇** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** | **$A_1$** | **$A_2$** | **Mean B** |
| $B_1$ | 482.2 | 511.2 | 496.7 | 483.3 | 512.4 | 497.8 | 486.3 | 512.9 | 499.6 | 486.2 | 515.3 | 500.7 |
| $B_2$ | 490.4 | 519 | 504.7 | 493.9 | 522.3 | 508.1 | 499 | 526.7 | 512.9 | 502.4 | 530.3 | 516.4 |
| $B_3$ | 489.8 | 519 | 504.4 | 493.8 | 522.1 | 507.9 | 496.1 | 528.3 | 512.2 | 500.1 | 532.4 | 516.2 |
| $B_4$ | 484.4 | 511.9 | 498.1 | 486.3 | 515.9 | 501.1 | 487.4 | 518.5 | 503 | 488.8 | 521.3 | 505.1 |
| $B_5$ | 486.3 | 512.4 | 499.3 | 487.4 | 517.3 | 502.3 | 488.8 | 519.2 | 504 | 490.1 | 522.3 | 506.2 |
| $B_6$ | 487.4 | 515.9 | 501.7 | 488.8 | 518.5 | 503.7 | 491 | 521.3 | 506.2 | 491.3 | 524.3 | 507.8 |
| $B_7$ | 488.8 | 517.3 | 503 | 491.3 | 521.1 | 506.2 | 495.7 | 528.8 | 512.3 | 498.4 | 533.9 | 516.2 |
| **Mean A** | **487.1** | **515.2** | **--** | **489.3** | **518.5** | **--** | **492.1** | **522.3** | **--** | **493.9** | **525.7** | **--** |
| Factors | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) | (A) | (B) | (AXB) |
| C.D. at (5%) | 0.24 | 0.449 | 0.635 | 0.176 | 0.329 | 0.465 | 1.127 | 2.109 | 2.983 | 0.932 | 1.744 | 2.466 |
| SE(m)± | 0.082 | 0.154 | 0.218 | 0.06 | 0.113 | 0.16 | 0.387 | 0.724 | 1.024 | 0.32 | 0.599 | 0.847 |

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

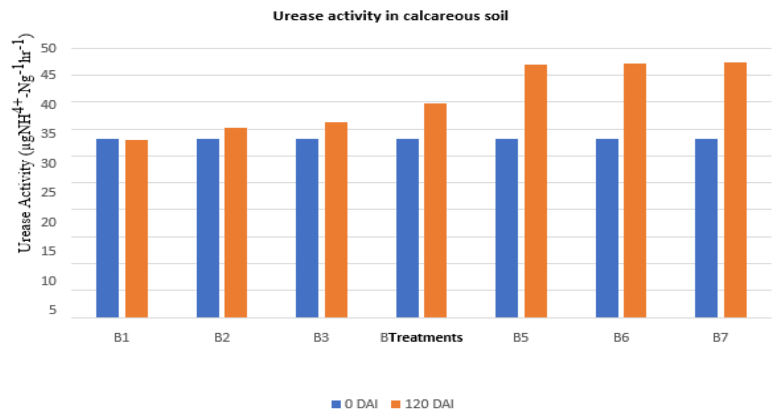
soil than in calcareous soil (487.05–493.90 kg ha$^{-1}$) over the incubation period. RDF through DAP recorded the highest potassium levels, with significant interaction effects, as non-calcareous soil with RDF through DAP reached 533.88 kg ha$^{-1}$ at 120 days. Potassium content is more readily available in manured soil than in unmanured soil was also observed by Brar *et al*. (2015).

### 3.3 Urease Activity

At 0 days, non-calcareous soil reported higher urease activity (40.03 µg $NH_4^+$-N g$^{-1}$ hr$^{-1}$) than calcareous soil (33.07 µg $NH_4^+$-N g$^{-1}$ hr$^{-1}$), with non-significant interaction. By 120 days, non-calcareous soil maintained higher activity (48.85 µg $NH_4^+$-N g$^{-1}$ hr$^{-1}$), and 100% $P_2O_5$ through PROM resulted in the highest activity (53.01 µg $NH_4^+$-N g$^{-1}$ hr$^{-1}$). Significantly higher activity (58.74 µg $NH_4^+$-N g$^{-1}$ hr$^{-1}$) at 120 days of incubation was reported for interaction $A_2B_7$ (non-calcareous soil + 100% $P_2O_5$ through PROM). R. Srinivasan *et al*. (2016) found that the addition of organic sources is a good source of energy and carbon to heterotrophs causing an increase in enzymatic activity and a rise in population.

### 3.4 Dehydrogenase Activity

At 0 days, non-calcareous soil reported higher dehydrogenase activity (16.05 µg TPF g$^{-1}$ 24 hr$^{-1}$) than calcareous soil (14.34 µg TPF g$^{-1}$ 24 hr$^{-1}$), with non-significant interaction, while at 120 days, non-calcareous soil recorded higher activity (18.76 µg TPF g$^{-1}$ 24 hr$^{-1}$). Application of 100% $P_2O_5$ through PROM recorded higher dehydrogenase activity (18.18 µg TPF g$^{-1}$ 24 hr$^{-1}$). The interaction effect was significant at 120 days and a combination of $A_2B_7$ (non-calcareous soil + 100% $P_2O_5$ through PROM) found higher activity (20.11 µg TPF g$^{-1}$ 24 hr$^{-1}$).



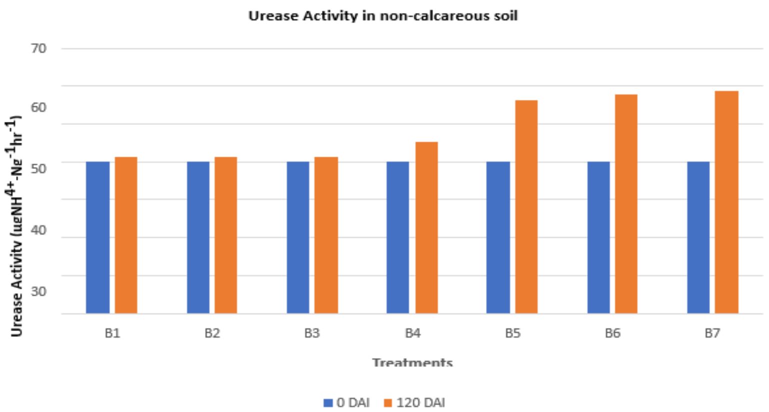




**Fig. 1. Periodical changes in urease activity due to P application**

*Note-$A_1$- Calcareous soil, $A_2$- Non-calcareous soil, $B_1$- Absolute control, $B_2$- RDF through DAP, $B_3$- RDF through SSP, $B_4$-25 % $P_2O_5$ through PROM, $B_5$-50 % $P_2O_5$ through PROM, $B_6$-75 % $P_2O_5$ through PROM, $B_7$-100 % $P_2O_5$ through PROM*

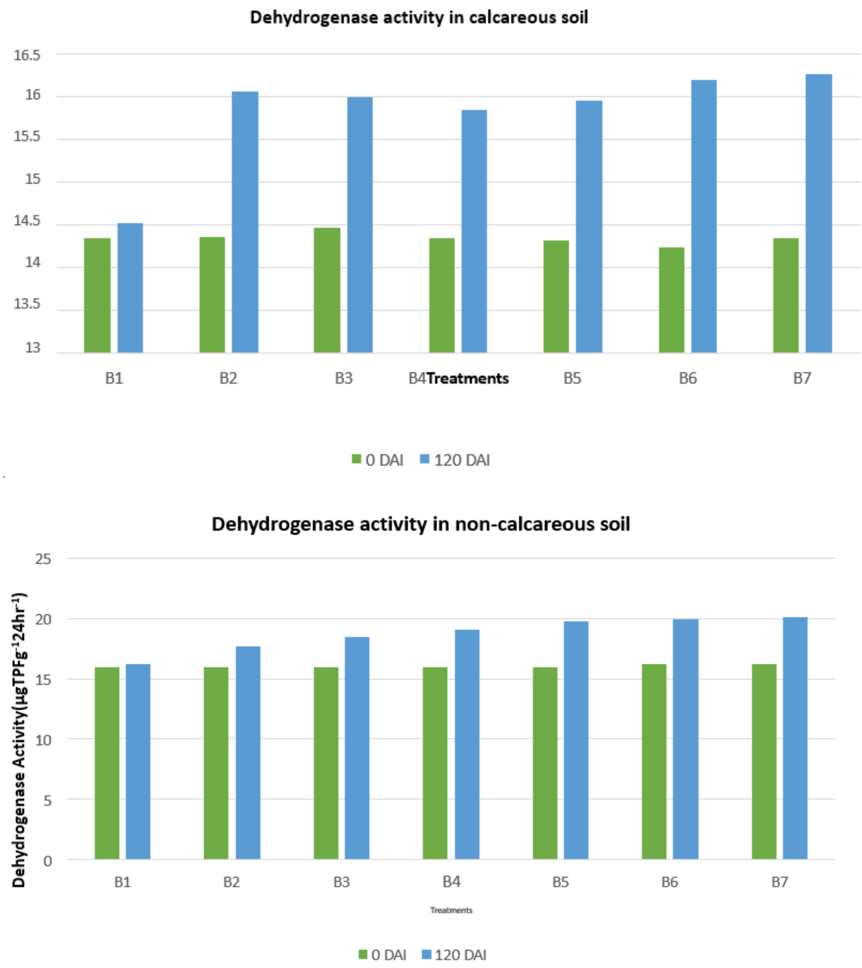



**Fig. 2. Periodical changes of dehydrogenase activity due to P application**

### 3.5 Alkaline Phosphatase Activity

At 0 days of incubation, alkaline phosphatase activity (9.66 µg PNP $g^{-1}$ $hr^{-1}$) of calcareous soil was higher than non-calcareous soil (8.51 µg PNP $g^{-1}$ $hr^{-1}$), with non-significant interaction. At 120 days, non-calcareous soil showed higher activity (16.66 µg PNP $g^{-1}$ $hr^{-1}$), with 100% $P_2O_5$ through PROM leading to the highest activity (18.08 µg PNP $g^{-1}$ $hr^{-1}$). The significant interaction effect at 120 days for $A_2B_7$ (non-calcareous soil + 100% $P_2O_5$) recorded higher alkaline phosphatase activity (18.44 µg PNP $g^{-1}$ $hr^{-1}$). Waldrip *et al*. (2012) reported that organic manure may enhance phosphatase activity by providing soil microbes with carbon, nitrogen, and phosphorus.

### 3.6 Correlation between Soil Enzyme Activities and Nutrient Release

At 0 days, soil urease activity recorded a significant positive correlation with nitrogen (r=0.880) and a non-significant negative correlation with phosphorus (r = -0.619) and potassium (r = 0.277). At 120 days, urease activity had a positive significant correlation with nitrogen (r = 0.645) and non-significant positive correlations with phosphorus (r = 0.404) and potassium (r = 0.034).

For micronutrients, at 0 days, it was negatively correlated with iron, zinc, and copper, while at 120 days, it showed significant positive correlations with all micronutrients: iron (r = 0.981), manganese (r = 0.945), zinc (r = 0.972), and copper (r = 0.959). Kaptanoglu Berber *et al*. (2014) reported in the study that there was not a statistically significant relationship between organic matter and urease enzyme activity.

At 0 days, soil dehydrogenase activity showed a significant positive correlation with phosphorus (r=0.847) and a non-significant negative correlation with nitrogen (r = -0.353). Potassium had a non-significant positive correlation (r=0.498), while zinc showed a significant negative correlation (r=-0.636). At 120 days, phosphorus continued to show a significant positive correlation (r=0.636), with nitrogen (r=0.516) and potassium (r=0.423) showing non-significant positive correlations. Non-significant negative correlations were found with iron (r=-0.437) and copper (r=-0.177), and a non-significant positive correlation with manganese (r=0.212). Phosphorus is a key factor affecting dehydrogenase activity over time. It was observed that the increase in dehydrogenase activity and microbial biomass was proportional to the addition of the number and quantity of nutrients (Manjaiah and Singh, 2001).

At 0 days, soil alkaline phosphatase activity showed a significant positive correlation with phosphorus (r = 0.787) and potassium (r = 0.844), but a non-significant correlation with nitrogen (r = -0.015). At 120 days, it remained positively correlated with phosphorus (r = 0.736), with non-significant correlations to nitrogen (r = 0.300) and potassium (r = 0.217). At 0 days, alkaline phosphatase also showed a significant positive correlation with manganese (r = 0.730) and non-significant correlations with iron (r = 0.186), zinc (r = 0.048), and copper (r = 0.433). At 120 days, it showed significant correlations with iron (r = 0.942) and manganese (r = 0.929) and non-significant correlations with zinc (r = 0.239) and copper (r = 0.137). Tarafdar *et al*. (1987) reported a significant correlation between organic P depletion and phosphatase activity in wheat (r= 0.99**) and clover (0.97**) rhizosphere soils.

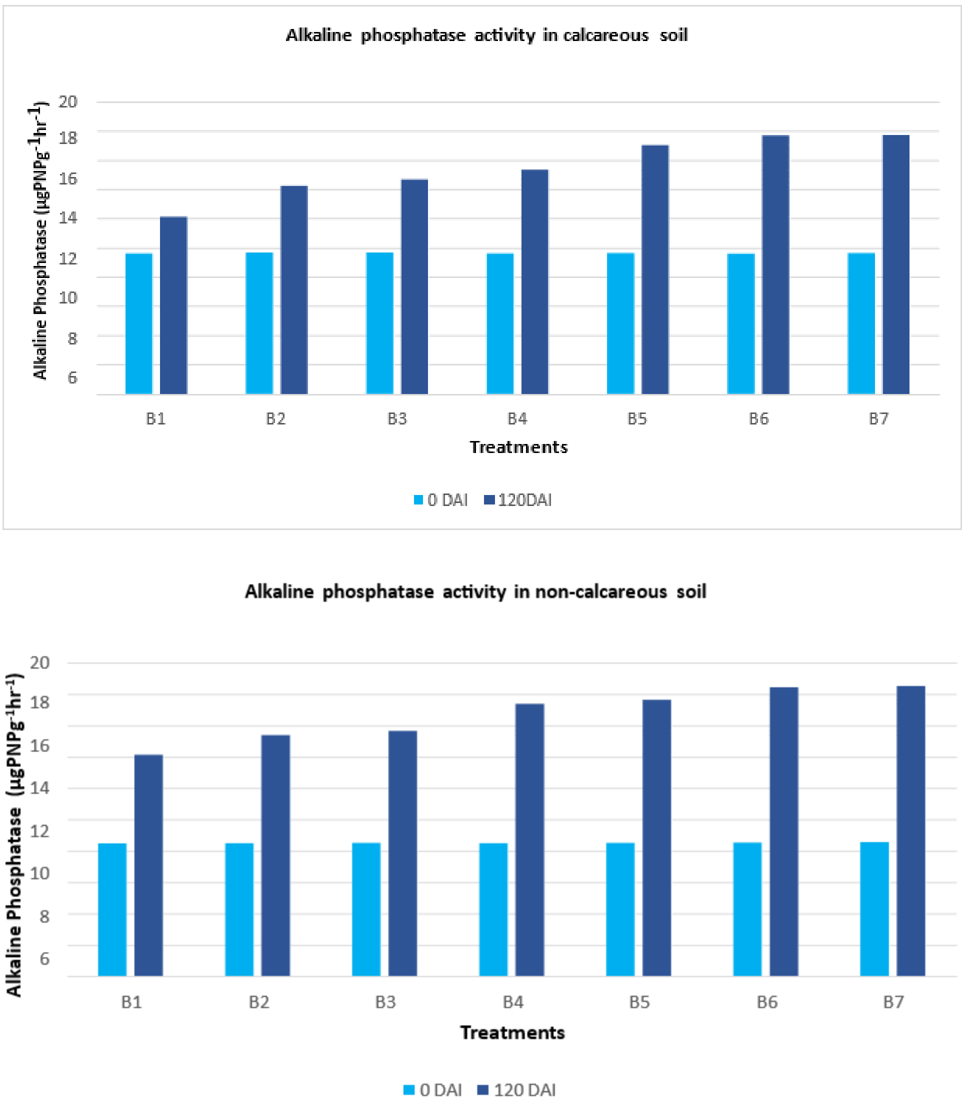



**Fig. 3. Periodical changes of alkaline phosphatase activity due to P application**

**Table 9. Correlation between soil enzyme activities and nutrient release at 0 days of incubation**

| 0 DAI | N | P | K | Fe | Mn | Zn | Cu | Urease | Dehydrogenase | Alkaline phosphatase |
|---|---|---|---|---|---|---|---|---|---|---|
| N | 1 | | | | | | | | | |
| P | 0.400 | 1 | | | | | | | | |
| K | 0.025 | -0.381 | 1 | | | | | | | |
| Fe | 0.448 | 0.445 | 0.525 | 1 | | | | | | |
| Mn | 0.066 | -0.127 | 0.854** | 0.697* | 1 | | | | | |
| Zn | 0.407 | -0.695* | 0.263 | 0.766** | 0.324 | 1 | | | | |
| Cu | -0.026 | 0.232 | 0.392 | 0.528 | 0.756* | 0.419 | 1 | | | |
| Urease | 0.880** | -0.619 | 0.277 | -0.227 | 0.020 | -0.553 | -0.500 | 1 | | |
| Dehydro genase | -0.353 | 0.847** | 0.498 | -0.437 | 0.212 | -0.636* | -0.177 | 0.673* | 1 | |
| Alkaline Phosphatase | -0.015 | 0.787** | 0.844** | 0.186 | 0.730* | 0.048 | 0.433 | 0.343 | 0.633* | 1 |

**Table 10. Correlation between soil enzyme activities and nutrient release at 120 days of incubation**

| 0 DAI | N | P | K | Fe | Mn | Zn | Cu | Urease | Dehydrogenase | Alkaline phosphatase |
|---|---|---|---|---|---|---|---|---|---|---|
| N | 1 | | | | | | | | | |
| P | 0.212 | 1 | | | | | | | | |
| K | 0.687 * | 0.818** | 1 | | | | | | | |
| Fe | 0.014 | 0.328 | -0.051 | 1 | | | | | | |
| Mn | -0.043 | 0.213 | -0.127 | 0.984** | 1 | | | | | |
| Zn | 0.010 | -0.248 | -0.091 | 0.990** | 0.998** | 1 | | | | |
| Cu | -0.014 | 0.267 | -0.090 | 0.995** | 0.994** | 0.298 | 1 | | | |
| Urease | 0.645* | 0.404 | 0.034 | 0.981** | 0.945** | 0.959** | 0.972** | 1 | | |
| Dehydrogenase | 0.516 | 0.636* | 0.423 | 0.801**. | 0.299 | 0.811** | 0.237 | 0.156 | 1 | |
| Alkaline Phosphatase | 0.300 | 0.736* | 0.217 | 0.942**. | 0.929** | 0.239 | 0.137 | 0.239 | 0.254 | 1 |

## 4. CONCLUSION

The application of Phosphate Rich Organic Manure (PROM) reduced soil calcium carbonate levels in both calcareous and non-calcareous soils significantly enhancing the availability of macronutrients (nitrogen, phosphorus, and potassium) compared to inorganic fertilizers. Over the 120-day incubation period, all three enzymes-urease, dehydrogenase, and alkaline phosphatase exhibited a gradual increase in activity, indicating active microbial processes and ongoing soil organic matter decomposition; notably, the increase in dehydrogenase activity suggests enhanced organic matter breakdown, which can improve soil structure and nutrient availability. Additionally, the correlation analysis highlighted the essential role of these enzymes in nutrient cycling, particularly the significant positive correlation between alkaline phosphatase and phosphorus at 120 days, underscoring its potential contribution to phosphorus mineralization and cycling within the soil ecosystem.

## DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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