Soil Acidity Indices, Nutrient Availability and Plant Growth through Amelioration Practices in Adjacent Coal-mine Paddy Soil

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Authors’ contributions
This work was carried out in collaboration among all authors. Author MSL conducted the study, performed the statistical analysis, wrote the protocol, managed the literature searches and wrote the first draft of the manuscript. Author NJS designed the study. Authors DT, VR, LIPR and KMS give suggestion and ideas on improvisation of the study. All authors read and approved the final manuscript.

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ABSTRACT
Potential adjacent coal mine paddy soils often endure low soil and plant productivity through unscientific mining activities causing acid mine drainage. But the extent of its effect to soil is not known, therefore the study was taken to characterize coal mine affected lowland fields on the basis of soil acidity, identify the best amelioration practices and evaluate the performance on rice productivity at farmers’ field level. An experiment with a completely randomised block design (5 replicates) was performed to determine the effects of poultry manure (PM), compost (C), lime (L), paper mill sludge (PMS) and microbial consortium (MC) with their suitable combination through pot experimentation at College of Post Graduate Studies followed by the preeminent selected practices at field trials at Khliehriat, Meghalaya. The factors used were PM and C (10 t ha⁻¹), L as CaCO₃, PMS (250 and 500 kg ha⁻¹) and MC were incorporated at appropriate rates. On categorization, two
locations were found to exhibit extremely pH acid soil (pH 4.51 ± 0.51) i.e. Moonlakhep (L1) and ultra pH soil (pH 3.14 ± 0.23) i.e. Ladrymbai (L2). Integration of practices showed significant increase in soil acidic indices such as soil pH by 6% to 23% and significant decrease in exchangeable acidity by 49% to 18% with T₄ at both locations. Confined increases of soil organic carbon by 12% to 40% with enhanced available soil nutrients by 40% at high optimum rates were noticed. Yield attributes were significantly influenced by different treatments. Highest plant height (83.58 cm and 81.32 cm), grain yield (3436 kg ha⁻¹ and 3120 kg ha⁻¹) were recorded under the practices of T₄. However, stover yield (7875 kg ha⁻¹) was noticed in T₈ at L1 and at L2 maximum in T₄ (7420 kg ha⁻¹). Soil acidic indices, nutrient and crop growth were influenced at high optimum rates of soil amendment and enhanced with PM amended soil.

Keywords: Coal mine paddy soil; amelioration practices; soil acidity indices; soil productivity; plant growth.

1. INTRODUCTION

Scenario of adjacent paddy fields near coal mine excavation has resulted in adverse effect on soil dynamics through the phenomenon of toxic/heavy metals flows with accumulate rainfall during monsoon season through the phenomenon known as acid mine drainage. These soil exhibits reduce capacity for plant growth and low soil productivity. In Meghalaya, India, the inception of the National Tribunal Congress (NGT) has made the coal extraction activities to a standstill and presently for income generation adjacent paddy field near these areas has been mostly concentrated for lease farmers but the ill affect from the past years has already deteriorate the soil resulting farmers in search of alternatives as to make the soil productive in nature.

As agriculture is turning into organic state, one such alternative is the use of amelioration practices which confer many benefits in reducing the metal toxicity as well as acidity in soil. Among the organic materials, Compost (C) is currently widely use as an inexpensive source [1] as it exhibits valuable effect to soil acidity [2,3]. Secondly, Lime (L) application to acid soils is well known especially in pyrite containing minerals coal mine soil [4,5] but the accountability on its rate of application must be taken [6]. Recent inputs as Poultry Manure (PM) adds advantage to reduce leaching of ammonium and nitrate [7], several studies has shows increase in soil pH, soil organic carbon and available nutrient through its application at high doses [8-11] while industrial wastes such as Paper Mill Sludge (PMS) acts as soil conditioner and sources of nutrient [12] and has demonstrated it application in soils to grain crops enhanced crop growth at agronomic rates [13-15], whereas in coal mines areas it application at high rate is advocated [16,17] resulting in elevating the pH, decline in pyrite oxidation and metal solubility [18]. One must take into consideration the vital role of microorganisms, but solely effect often shows not to the mark results, therefore to enhance growth, one such sources recently incorporated in soils is the microbial consortium (MC) where suitable combination of rhizosphere microbe are merge by artificial culturing [19]. In acid soil, P is most limiting so incorporation of microbe through PSB will helps plant in uptake through the rhizosphere region through root dip [20].

Overall, the organic amendments displayed many advantage but the need of identifying appropriate combination is necessary [21] therefore in these adjacent paddy areas, some studies on their soil nutrient and heavy metal status had been done, but covering of mostly the potential areas of paddy fields near coal mining activities and the use of the above said organic materials has been not been practiced. Therefore, with this background, this study intend to determine the categorization of soil on the basis of soil acidity of 10 potential paddy soil locations, identification the suitable practices on the categories soil and lastly demonstrated the best management practices of the adjacent paddy fields under field trials.

2. MATERIALS AND METHODS

2.1 Detailed Plan of the Work

2.1.1 Study site

Khliehriat is located in East Jaintia Hills district of Meghalaya, India (25°21′31.2" N latitude and 92°22′11.5" E longitude) situated at 1172 m
above mean sea level. The research plan was carried out through pot experiment at the College of Post Graduate Studies Central Agricultural University, Umiam, Meghalaya. The location is at altitude of 250 m (41°01’91”N latitude & 91°54’46.24”E longitude). It is situated within tropical forest zone with bimodal rainfall.

2.1.2 Experimental details, design and treatments allocation under pot experiment

Soil bulk samples were collected at 0-15cm from identified locations, air dried, passed through 2mm sieved and analyzed for soil acidity indices as based on the acidity categorization [22]. For pot experiment large volume soil were collected from selected categories soil indices location and thoroughly mixed with respective treatments. Each pot 7 kg capacities were filled with 5 kg treated soil. Test crop namely CAU-R3 (Rice variety) was first sown in nursery bed (2 x 5 ft); 20 days old seedlings uprooted and dipped in MC overnight. During transplanting two (2) seedlings were incorporated to each pot. The gap filling were carried out after 5-10 days of transplanting.

The experiment was arranged in a completely randomized block design with 5 no. of replications. The numbers of treatment as listed below were allocated to soils of different acidity indices (Fig. 2.).

- **T1**: Control
- **T2**: C + L @ 500 kg/ha+ MC
- **T3**: C + L @ 250 kg/ha+ MC
- **T4**: PM + L @ 500 kg/ha+ MC
- **T5**: PM + L @ 250 kg/ha+ MC
- **T6**: C+ PMS @ 500 kg/ha+ MC
- **T7**: C+ PMS @ 250 kg/ha+ MC
- **T8**: PM + PMS @ 500 kg/ha+ MC
- **T9**: PM + PMS @ 250 kg/ha+ MC
- **T10**: MC

2.2 Soil Routine Analysis

The soil samples were processed using standard methodology [23] and analysed for different soil acidity indices and nutrient status of soil.

2.3 Field Trials

On scrutinizing the results from the pot experiment, the best management amelioration practices was conducted in paddy field of certain locations based on their selected acidities under the Khliehriat Block, East Jaintia Hills, Meghalaya comprising of 3 fields of 20 m x 20 m each.

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**Fig. 1. Flow chart of plan of work**
3. RESULTS AND DISCUSSION

3.1 Initial Readings of 10 Locations Soils

After observation of the 10 locations paddy sampled soil, a conclusion was drawn to categorise soil on the basis of acidity. However, due to similar soil geology and environments, it became extremely difficult to categorise these soil into three component of high, medium and low pH soil, therefore a conclusion was drawn to categorise soil only on the basis of ultra pH for L2 and extremely acid pH for L1 areas soil based as per USDA guidelines (Table 2). Therefore, the said locations were selected as two reference soil for pot experiment and amelioration practices (Table 1) were incorporated in these soil.

3.2 Effect of Amelioration Practices on Soil Acidity Indices under Pot Experiment

The application of all amelioration practices on the adjacent paddy field of both location soils increased the soil pH and base saturation (BS) and decrease the exchangeable acidity as compared to its natural state (Table 3) [24,25]. Among the amelioration practices, the high rate of application exhibited favourable effect in both location with pronounce effect in L treated plot with an increase of ≥ 10% to 23% at L1 and ≥ 6% to 20% at L2 in amalgamation with PM amended soil, which might associate with its high relatively content of Ca [26,27] and its improved nutrient status [28-30] while L as a source of basic cations (Ca\(^{2+}\) and Mg\(^{2+}\)) [31] and anions (CO\(_3^{2-}\)) are able to exchange H\(^{+}\) from exchange sites and eliminate Al\(^{3+}\) toxicity, which is a common stress factor in acidic soil with pH < 4 [32,33]. In L1, soil pH was highest under T\(_4\) (pH 5.54) followed by T\(_2\) (pH 5.25) with an increase of about 23% and 16% respectively while ultra acid soil of L2 registered an increase of 20% and 13% as compared to control T\(_1\). C effect on these soils was noticed as reported by [34] but effectiveness of manure on soil pH depends on its quality [35]. PMS amended plot T\(_6\), T\(_7\), T\(_8\) and T\(_9\) on the other hand registered increase of merely 4% to 15% and 3% to 10% respectively which might be due to its ineffectiveness at the primary stage [36-38], and need long term studies (above 15 years) as demonstrated [39,40] to illustrate results especially in acid soils. T\(_{10}\) showed negligible effect on soil acidity indices [41].

Table 1. Nutrient content of soil amelioration practices

| Amelioration practices | Soil pH | OC (%) | N (%) | P (%) | K (%) | S (%) | Ca (%) | Mg (%) |
|------------------------|---------|--------|-------|-------|-------|-------|--------|--------|
| PM                     | 7.09    | 13.26  | 1.16  | 1.12  | 0.74  | 0.31  | 14.02  | 0.41   |
| C                      | 7.29    | 20.19  | 0.72  | 0.24  | 0.39  | 0.41  | 2.21   | 0.13   |
| PMS                    | 7.32    | 22.00  | 0.054 | 0.58  | 0.01  | --    | 0.30   | 0.18   |
| L                      | 11.02   | --     | --    | --    | --    | --    | 37.59  | 15.34  |
Table 2. The soil acidities parameters of East Jaintia hill district, Meghalaya

| Sl. no. | Location          | pH     | Ex. acidity (meq/100g) | Ex. Al (meq/100g) | ECEC    | LR (t/ha) | BS (%)   |
|---------|-------------------|--------|------------------------|-------------------|---------|-----------|----------|
| 1       | Rymbai            | 4.27 ± 0.31 | 2.21 ± 0.46             | 3.16 ± 0.32       | 3.16 ± 0.07 | 12.57 ± 1.91 | 31.40 ± 0.39 |
| 2       | Dentrum           | 4.20 ± 0.27 | 2.34 ± 0.54             | 3.58 ± 0.28       | 3.34 ± 0.13 | 10.26 ± 1.39 | 31.27 ± 0.21 |
| 3       | Khiesarang        | 4.34 ± 0.44 | 2.21 ± 0.37             | 3.24 ± 0.24       | 3.10 ± 0.18 | 10.15 ± 1.10 | 30.45 ± 0.19 |
| 4       | Lad Rymbai        | 3.14 ± 0.23 | 3.31 ± 0.40             | 4.33 ± 0.29       | 3.13 ± 0.10 | 19.64 ± 0.89 | 30.80 ± 0.11 |
| 5       | Bataw             | 4.15 ± 0.32 | 2.32 ± 0.13             | 3.36 ± 0.28       | 3.09 ± 0.07 | 12.01 ± 1.46 | 31.38 ± 0.32 |
| 6       | Lumshnong         | 4.29 ± 0.48 | 2.84 ± 0.19             | 2.28 ± 0.19       | 3.13 ± 0.07 | 11.48 ± 1.34 | 31.88 ± 0.16 |
| 7       | Moonlakhep        | 4.51 ± 0.51 | 1.81 ± 0.17             | 2.07 ± 0.12       | 3.39 ± 0.21 | 10.15 ± 2.15 | 32.18 ± 0.40 |
| 8       | Dakhiah west      | 3.57 ± 0.24 | 3.13 ± 0.22             | 3.54 ± 0.33       | 3.21 ± 0.11 | 14.11 ± 1.02 | 30.49 ± 0.15 |
| 9       | Latyrke           | 4.30 ± 0.16 | 3.20 ± 0.35             | 3.19 ± 0.41       | 3.18 ± 0.12 | 10.59 ± 0.96 | 31.16 ± 0.24 |
| 10      | Khliehriat East   | 4.34 ± 0.35 | 2.25 ± 0.28             | 3.18 ± 0.40       | 3.19 ± 0.16 | 11.67 ± 1.11 | 30.74 ± 0.15 |

Note: mean values of 10 composite soil samples of each location ± std. deviation. Ex - exchangeable, ECEC – effective CEC, LR-lime requirement, BS-base saturation
Table 3. Effect of amelioration practices/treatments on chemical properties of extremely acidic soil of Moonlakhep (L1) and ultra acidic soil of Lad Rymbai (L2)

| Treatments | pH  | SOC (%) | Ex. Acidity (meq/100g) | BS(%) |
|------------|-----|---------|------------------------|-------|
|            | L1  | L2      | L1                     | L2    |
| T1         | 4.51| 3.14    | 1.82                   | 1.53  |
| T2         | 5.25| 3.64    | 2.36                   | 1.84  |
| T3         | 4.95| 3.34    | 2.13                   | 1.64  |
| T4         | 5.54| 3.85    | 2.57                   | 2.00  |
| T5         | 5.09| 3.43    | 2.25                   | 1.73  |
| T6         | 5.02| 3.48    | 2.22                   | 1.72  |
| T7         | 4.69| 3.30    | 1.96                   | 1.68  |
| T8         | 5.18| 3.58    | 2.27                   | 1.92  |
| T9         | 4.77| 3.36    | 2.16                   | 1.71  |
| T10        | 4.54| 3.15    | 1.83                   | 1.56  |

SE (m)±
- 0.019
- 0.013
- 0.024
- 0.016
- 0.020
- 0.030
- 0.072
- 0.090

CD (p≤0.05)
- 0.095
- 0.075
- 0.144
- 0.098
- 0.121
- 0.183
- NS
- NS

Table 4. Effect of amelioration practices/treatments on soil available nutrient of extremely acidic soil of Moonlakhep (L1) and ultra acidic soil of Lad Rymbai (L2)

| Treatments | Av. N (kg/ha) | Av. P (kg/ha) | Av. K (kg/ha) |
|------------|---------------|---------------|---------------|
|            | L1            | L2            | L1            | L2            | L1            | L2            |
| T1         | 201.56        | 164.11        | 12.43         | 9.16          | 183.81        | 160.26        |
| T2         | 246.06        | 205.11        | 16.38         | 12.05         | 245.83        | 210.61        |
| T3         | 229.03        | 183.43        | 14.06         | 11.27         | 229.58        | 182.35        |
| T4         | 281.99        | 234.19        | 18.41         | 12.95         | 254.87        | 232.44        |
| T5         | 251.13        | 211.15        | 15.57         | 11.93         | 236.43        | 196.06        |
| T6         | 228.70        | 194.83        | 14.41         | 11.69         | 234.87        | 189.19        |
| T7         | 216.56        | 180.47        | 13.94         | 10.52         | 217.52        | 174.33        |
| T8         | 249.92        | 210.39        | 16.09         | 12.10         | 241.04        | 200.86        |
| T9         | 229.19        | 192.27        | 14.80         | 11.08         | 223.33        | 179.72        |
| T10        | 206.31        | 168.03        | 12.75         | 9.43          | 191.53        | 162.13        |

SE (m)±
- 1.370
- 1.226
- 0.052
- 0.033
- 1.303
- 1.266

CD (p≤0.05)
- 8.207
- 7.350
- 0.348
- 0.200
- 7.807
- 7.586

3.3 Effect of Amelioration Practices on Soil Nutrients

3.3.1 Soil organic carbons

Soil organic carbon (SOC) was significantly (p < 0.05) influenced by amelioration practices in the pot experiment (Table 4). Soil amendment with the optimum rates resulting in higher organic carbon in which L amended soil registered higher organic carbon than PMS amended soil but the difference were not so vast i.e. 22% to 40% at L1 and 12% to 31% at L2, fact being that soil organic carbon take time to amplified in the soil [42,43]. PM showed higher SOC citing the effect of low C:N ratio [44] and enhanced physical condition of the soil [45], that result in proliferation soil microbial biomass and their activity in the soil [42]. C and PMS as an organic input did showed some effect on organic carbon, this may be attributed to the fact of their mineralization rates.

3.3.2 Soil available nitrogen (N), phosphorus (P) and potassium (K)

Compared to T1, the primary nutrients (N, P, K) in T4 showed an increase by about 40% in N, P and K at both respectively locations (Table 4). Nitrogen was highly significant in PM amended soils might be due to the composition in the part of PM component [46-50] with increasing rate of L increases soil available N [51,52]. PMS showed low soil available N owing to its low levels of nitrogenous matter [53]. Overall, residue effect of urea applied previously in these region soils has make N to be high [54]. Phosphorus was recorded low with less variation among the treatments in both locations; this is because it requires time to release soil fixed P for
mineralization to take place. [50,55]. L effect was insignificant thereby confirming that these soils deficient in available P [56] PMS resulted low P on account for its breakdown through hydrolysis in soil [53]. MC contribution might be possible that the insoluble P have been made available [57] by the effective microbes which involve P cycle [58]. Potassium on the other hand, showed promising results through the amelioration practices such as C and PM due to its solubility, retentionbility and reduce leaching loss [59]. Manure combined with L increased K [24]. Moreover, in soils with pH dependent charges, increase in pH with liming enhances the CEC [60]. Combination of animal manures with MC has been reported to increase K content [61]. Since MC aids in reimburse for the deficiency of the immobile nutrients, the integrated application of PM/C with L/ PMS with microorganism can be measured as a useful practice.

3.4 Plant Parameters

3.4.1 Plant height and stover yield

In the soil of L1, adjacent paddy soil near coal mine showed plant height (in cm) to be greater in the treatment T4 (83.6 cm) suggesting that PM, which contains total N (> 4%), supplied a more balanced mix of NPK nutrients that was synchronized to the needs of the rice plants throughout the crop period [62,63]. From the Table 5 it is observed that PM amended soil give superior readings with both L i.e. T4 and T5 (increase of 22 to 28%) as well as with PMS i.e. T8 and T9 (increase of 16% to 23%) application compared to control. Meanwhile, C did not exert significant effect as organic matter with low nitrogen content decomposes slowly. In L2 soil, plant height ranged from 64.4 to 82.3 cm. (Fig. 3). The application of L might have decreased acidity, increased availability of plant nutrients and enhanced microbial activities [64,65]. Increase in rice plant height through application of PMS from the same source was reported [53].

The amelioration practices of PM and PMS, dry matter accumulation was privileged Table 5. All practices showed better results at higher rates of application in both location soils. In L1, the increase of >13 to 19% was noticed in T8 and T9. L2 biomass recorded low and lesser than L1. Among treatment, L in amalgamation with PM showed an increase of 9% to 18% and 7% to 11% was observed in T2 and T3 soils comprising of C. Stover yield was observed highest in PM might be attributed to the easily and faster mineralization [66,67]. PMS due to its composition has resulted in substantial stover yield in rice. MC is known to have useful approach to encourage plant growth [68,69].
Table 5. Effect of amelioration practices/treatments on plant growth of extremely acidic soil of Moonlakhep (L1) and ultra acidic soil of Lad Rymbai (L2)

| Treatments | Plant height (cm) | Grain yield (kg/ha) | Stover weight (kg/ha) |
|------------|------------------|---------------------|----------------------|
|            | L1               | L2                  | L1                   | L2                  |
| T1         | 65.40            | 64.41               | 2489                 | 2277                | 6596 | 6304 |
| T2         | 79.03            | 75.92               | 3208                 | 2967                | 7317 | 7055 |
| T3         | 74.22            | 72.12               | 2839                 | 2665                | 7012 | 6718 |
| T4         | 83.58            | 81.32               | 3436                 | 3120                | 7673 | 7420 |
| T5         | 80.15            | 78.91               | 2968                 | 2850                | 6950 | 6595 |
| T6         | 74.89            | 73.18               | 3114                 | 2749                | 7257 | 6946 |
| T7         | 72.58            | 71.39               | 2732                 | 2514                | 6950 | 6595 |
| T8         | 80.68            | 78.47               | 3224                 | 2837                | 7875 | 7283 |
| T9         | 76.47            | 75.32               | 2936                 | 2606                | 7473 | 6794 |
| T10        | 67.31            | 65.02               | 2528                 | 2292                | 6610 | 6316 |
| SE (m)±    | 0.333            | 0.321               | 17.579               | 15.665              | 24.104 | 21.08 |
| CD (p ≤0.05) | 1.99       | 1.922               | 105.34               | 93.876              | 144.44 | 126.33 |

Table 6. Field trails results from three plots of Moonlakhep (L1) and ultra acidic soil of Lad Rymbai (L2)

| Parameters                      | R1                             | R2                             | R3 | Mean |
|---------------------------------|---------------------------------|---------------------------------|----|------|
|                                 | L1                             | L2                             | L1 | L2   |
| pH                              | 5.61                           | 3.41                           | 5.79| 3.37 | 5.89 | 3.11 | 5.76 | 3.36 |
| Ex. Acidity (meq/100g)         | 0.83                           | 0.87                           | 0.74| 0.91 | 0.77 | 1.05 | 0.78 | 0.94 |
| Base Saturation (%)             | 41.08                          | 34.16                          | 42.53| 34.11| 39.44| 33.94| 41.02| 34.07 |
| SOC (%)                         | 2.46                           | 2.19                           | 2.58| 2.24 | 2.67 | 2.27 | 2.57 | 2.23 |
| Av.N (kg/ha)                    | 288.09                         | 263.05                         | 273.94| 251.82| 274.85| 254.37| 278.96| 256.41 |
| Av.P (kg/ha)                    | 19.05                          | 15.06                          | 18.26| 14.64| 17.94| 14.33| 18.42| 14.68 |
| Av.K (kg/ha)                    | 266.71                         | 228.26                         | 248.13| 223.4| 256.83| 231.75| 257.22| 227.80 |
| Plant Height (cm)               | 85                             | 82.3                           | 89 | 77.6 | 81    | 86.3 | 85.00| 82.00 |
| Stover Yield (kg/ha)            | 8216                           | 7948                           | 7851| 7647 | 8109  | 7264 | 8058.67| 7619.67 |
| Grain Yield (kg/ha)             | 3416                           | 2400                           | 3394| 2219 | 3489  | 2344 | 3433.00| 2256.00 |

R=Replication

3.4.2 Grain yield

Table 5 depicts the effect of amelioration practices on grain yield in the extremely acid pH paddy soil of L1 areas. Yield was significantly affected by all practices, among the practices, T₄ registered the highest yield (3436 kg/ha) and elevated rate application of L and PMS resulted...
in higher yield compared to 50% application soil. In L2, grains yield varied from 2277 to 3120 kg/ha where $T_4$ registered the highest yield with over 37% increase in comparison with $T_1$. Surprisingly, $T_2$ showed an increase of 30% in grain yield. The results suggest that PM offered better nutritional quality and favourable balance of nutrients when supplemented with L and PMS [70,71]. In our study, the yield increment suggests a long term study is needed [72,73]. The increase in yield through L in combination with both manures may be due its part to the neutralization of exchangeable $\text{Al}^{3+}$ ions and in combination with PM the $\text{Ca}^{2+}$, aids in the grain filling [74]. PMS role might be due to its significant increase of soil pH [75] while many studies [76,77,78] has reported that high C:N ratio in PMS has affect yield.

3.5 Field Trials

Observation as notice in the two locations reveal that the treatment of $T_4$ i.e PM+L+MC was the best amelioration practices with all soil and plant parameters showing it dominate effect in these soil. Therefore, field trials was conducted on both location (Figs. 4 and 5) with incorporation of this practices in adjacent to farmers practices as depicted in Table 6.

4. CONCLUSION

All amelioration practices neutralised the soil acidity with increased in soil pH and exchangeable cations. Soil available nutrients showed promising results especially N and K with almost all the added combination practices while PM amended soil with L i.e. $T_4$ improvise better in these soil. Plant parameters was influenced mostly by L and PMS amended plot with PM i.e. $T_4$, $T_5$, $T_8$, $T_9$ in terms of plant height and stover yield, but grain yield was maximum at $T_4$ thus showing it dominant in terms of output entity. All soil and plant parameters was enhanced by high optimum rates of soil amendment of L and PMS especially with PM compared to C amended soils which displayed inferior effect in these high acidic soils. Hence the use of PM, L and MC helps in soil productivity and crop growth in these damaged adjacent coal mine soil which in turn will helps farmer to gain crop productivity and also reduce the environmental deterioration on soil-plant ecosystem.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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