

Article

Applying Limestone or Basalt in Combination with Bio-Fertilizer to Sustain Rice Production on an Acid Sulfate Soil in Malaysia

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Abstract: A study was conducted to determine the efficacy of applying ground magnesium limestone (GML) or ground basalt in combination with bio-fertilizer to sustain rice production on an acid sulfate soil in Malaysia. Soils from Kelantan Plains, Malaysia, were treated with GML, ground basalt, bio-fertilizer, GML + bio-fertilizer, and ground basalt + bio-fertilizer (4 t ha⁻¹ each). Results showed that soil fertility was improved by applying the soil amendments. GML and basalt contain some Zn and Cu; thus, application of these amendments would increase their contents in the soil needed for the healthy growth of rice. Basalt applied in combination with bio-fertilizer appeared to be the best agronomic option to improve the fertility of acid sulfate soils for sustainable rice production in the long run. In addition to increasing Ca, Mg, Zn, and Cu reserves in the soil, water pH increased and precipitated Al³⁺ and/or Fe²⁺. Ground basalt is cheaper than GML, but basalt dissolution in the acidic soil was slow. As such, its ameliorative effects could only be seen significantly from the second season onwards. The specially-formulated bio-fertilizer for alleviating the infertility of acid sulfate soil could also enhance rice growth. The use of the bio-fertilizer fortified with N₂-fixing bacteria is a green technology that would help reduce NO₃⁻ and/or NO₂⁻ pollution and reduce the cost of rice production. The phosphate-solubilizing bacteria (PSB) present in the bio-fertilizer not only increased the available P, but also helped release organic acids that would inactivate Al³⁺ and/or Fe²⁺ via the process of chelation.

Keywords: acid sulfate soil; Al and Fe chelation; organic acids; rice cultivation; soil amendments

1. Introduction

The demand for rice is going up annually with the increase in human population worldwide; as it is, more than one billion people depend on rice cultivation as their main source of income [1]. Considering the importance of rice and its economic role, rice production in the world should be increased substantially. To open up new land areas for rice cultivation in most Association of Southeast Asian Nations (ASEAN) countries is not practical due to the scarcity of fertile land. Therefore, using the available, but less productive lands/soils in these countries is one of the options to increase rice production.

Malaysia intends to raise its rice self-sufficiency level from 73% to 86% by the year 2020. Hence, it needs to expand the area for rice cultivation, improve rice productivity, or a combination of both.

Less productive acid sulfate soils, which are sporadically distributed in the coastal plains of Malaysia, can be ameliorated via innovative agronomic practices.

Acid sulfate soils are characterized by a pH of <3.5 and the presence of pyrite (FeS_2) formed when coastal sediments are inundated by seawater [1]. This pedogenic pyrite can easily be oxidized on exposure to the air, releasing sulfuric acid and Al and/or Fe into the soil environment where, under anaerobic conditions, these metals mostly exist as Al^{3+} and Fe^{2+} , respectively. In the end, a yellowish mineral called jarosite ($\text{KFe}_3(\text{SO})_2(\text{OH})_6$) is formed [2]. Based on the physicochemical properties, as defined by the Soil Survey Staff, most of the acid sulfate soils in the ASEAN region used for agriculture can be classified as sulfaquepts [3].

The low pH and high Al and/or Fe concentration can be detrimental to rice. Al inhibits cell elongation that curtails the growth of rice roots [4]. Al^{3+} and Fe^{2+} are attracted to the negatively-charged cell wall of rice roots, inhibiting their cell division and elongation. The consequence of this phenomenon is reduction in nutrient uptake, leading to reduced rice yield.

Among the agronomic practices recommended to improve the fertility of acid sulfate soils acid sulfate soils are lime application [5], submergence, leaching, and application of manganese dioxide [6], application of basalt [7], or employment of bio-fertilizer [8,9]. Depending on the rate applied, adding lime or basalt to the soils would increase their pH. When soil pH rises to a level above 5, Al is precipitated as inert Al-hydroxides; hence, it is no longer a threat to rice growth. Additionally, Ca and Mg released by the amendments are made available for rice production.

An innovative agronomic option is to apply ground magnesium limestone (GML) or basalt in combination with bio-fertilizer [8]. The presence of organic bio-fertilizer in a flooded acid sulfate soil could enhance the reduction process [10], leading to the release of Fe^{2+} [11], which is toxic to rice plants [12]. This study was conducted to determine the efficacy of applying GML or ground basalt in combination with bio-fertilizer to sustain rice production and improve acid sulfate soils in Malaysia. The information obtained from this study could be extended to the farming communities of the ASEAN region with similar soils that currently crop rice.

2. Experimental Section

2.1. Experimental Site

This study was conducted on an acid sulfate soil area in Semerak, Kelantan, Malaysia. The site was about 5 m above sea level, located at the latitude of $5^\circ 52'208''$ N and longitude of $102^\circ 28'501''$ E (Figure 1).

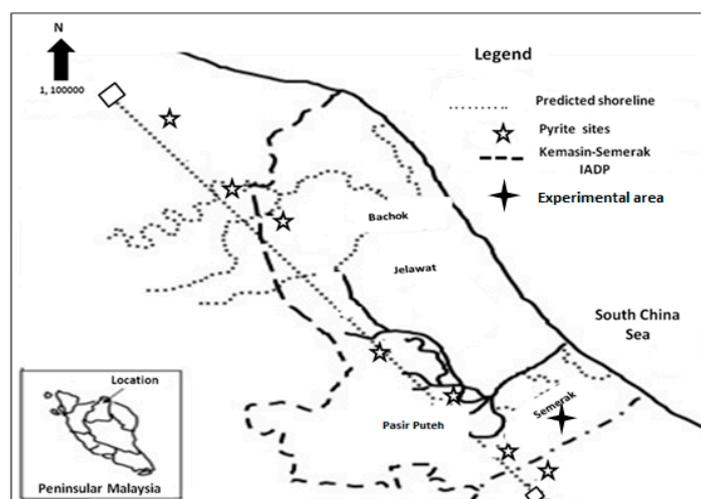


Figure 1. The predicted shoreline in the Kelantan Plains, Malaysia, about 4300 years before present (BP), modified from Enio et al. [1].

2.2. Experimental Design

GML and basalt with or without bio-fertilizer were broadcasted into the soil 15 days before sowing rice seedlings. The treatments were: Control, GML ($4 \text{ t}^{-1} \cdot \text{ha}$), ground basalt ($4 \text{ t} \cdot \text{ha}^{-1}$), bio fertilizer ($4 \text{ t} \cdot \text{ha}^{-1}$), GML + bio-fertilizer ($4 \text{ t} \cdot \text{ha}^{-1}$ each), and ground basalt + bio-fertilizer ($4 \text{ t} \cdot \text{ha}^{-1}$ each) with $5 \times 5 \text{ m}$ size of each plot. They were arranged in randomized complete block design (RCBD) with four replications. Nitrogen (N), phosphorus (P), and muriate of potash (KCl) were applied every season including control at 120, 30, and $60 \text{ kg} \cdot \text{ha}^{-1}$, respectively. The trial was run for three seasons consecutively, but the amendments were only applied during the first season. In Malaysia most rice is cultivated more than two times in a year, consequently.

2.2.1. Biochemical Properties of the Basalt, Ground Magnesium Limestone (GML) and Bio-Fertilizer

The basalt is composed of SiO_2 51.62%, MnO 0.20%, MgO 5.77%, CaO 9.15%, K_2O 0.97%, P_2O_5 15%, and Na_2O 3.14%. GML contains: CaCO_3 54%, CaO 30%, MgCO_3 44% and MgO 20%. However, the bio-fertilizer was fortified with fixing bacteria (*Stenotrophomonas maltophilia*) and phosphate-solubilizing bacteria (*Bacillus* sp., *Burkholderia thailandensis*, *Sphingomonas pituitosa*, and *Burkholderia seminalis*) at the population of 1×10^{-8} colony forming unit (cfu). The former can fix $52 \text{ kg} \cdot \text{N}^{-1} \cdot \text{ha}^{-1} \cdot \text{year}^{-1}$ from the air, while the latter can release phytohormones (indoleacetic acid), organic acids, and enzymes, and is furthermore able to dissolve insoluble phosphates [8]. The organic matter source used in the formulation of the bio-fertilizer was oil palm empty fruit bunch (EFB) and peat soil containing 48% carbon.

2.2.2. Rice Seedlings and Transplanting

The rice variety tested was MR 219 (wetland rice). The seeds were surface sterilized [13] and sown in a plastic tray. The 21-day-old seedlings were transplanted ($20 \times 20 \text{ cm}$) onto the $5 \times 5 \text{ m}$ experimental plots in the rice field on 6 November 2012. Second transplanting was started on 13 April 2013, and third transplanting was started on 27 October 2013.

2.3. Soil and Plant Analysis

Soil samples were taken at the surface level (0–6 inches) for analysis before and after experimentation. Soil pH was determined in water (1:2.5) using a Soil pH was determined in water (1:2.5) using a PHM210 standard pH meter (MAXTECH Corporation, Minneapolis, MN, USA) at $25 \text{ }^\circ\text{C}$ [14] and electrical conductivity (EC) was determined [14]. Total N was determined by the Kjeldahl digestion method [15]. Available soil P was determined according to Bray and Kurtz [16], while plant tissue P was analyzed by the wet digestion method [17]. Exchangeable Ca, Mg, and K were extracted using $1 \text{ M NH}_4\text{OAc}$ [18] and the cations in the extracts were determined by Atomic Absorption Spectroscopy (AAS). Exchangeable Al was extracted by 1 M KCl and the Al in the extract was determined by AAS [19]. Total carbon was analyzed by LECO CR-412 Carbon Analyzer (Leco Corporation, St. Joseph, MI, USA). Micronutrients in the soil were dissolved by double acid method ($0.05 \text{ M HCl} + 0.0125 \text{ M H}_2\text{SO}_4$) and the metals in the solution were determined using inductively coupled plasma atomic emission spectrometry (ICP-AES). The cation-exchange capacity (CEC) of the soil was determined by Kitsopoulos [20].

2.4. Determination of Yield Parameters and Nutrient Concentration in Plant

The crop was harvested at maturity and a one meter square plot was used for the determination of the grain and straw yield, while filled grains were isolated from the unfilled ones followed the method described by Seizo [21]. Chlorophyll content using a MINOLTATM SPAD-502 meter (Konica Minolta, Tokyo, Japan) plant height, root length, tiller/plant, number of panicles per plant, and plant nutrient uptake were measured by the dry-ashing method [22,23].

2.5. Determination of Aluminum Form in the Soil

Exchangeable Al was extracted with 1 M KCl at 1:10 (soil/solution ratio) by shaking for 24 h, while the weakly organically-bound Al form was extracted with 0.3 M CuCl_2 at 1:10 (soil/solution ratio) by shaking for 2 h. The strongly organically-bound Al form was extracted with 0.1 M $\text{Na}_4\text{P}_2\text{O}_7$ at 1:10 (soil/solution ratio) by shaking for 24 h. In all phases, the supernatant was isolated by centrifugation for 20 min at 13,500 rpm and, when necessary, they were further purified by filtration. The amount of the organically-bound Al in the soil was determined as the difference between $\text{Na}_4\text{P}_2\text{O}_7$ extracted-Al and CuCl_2 extracted-Al [24]. The Al in the solution was analyzed using ICP-AES.

2.6. Average Weather Conditions at the Study Site

The average monthly rainfall data taken from the studied area during the experimental period are shown in Figure 2a. There is monsoon season from October to December every year in Malaysia, which is why there was high rainfall. The day temperature remained at 28–32 °C, whereas night temperature was 22–24 °C throughout the year (Figure 2b).

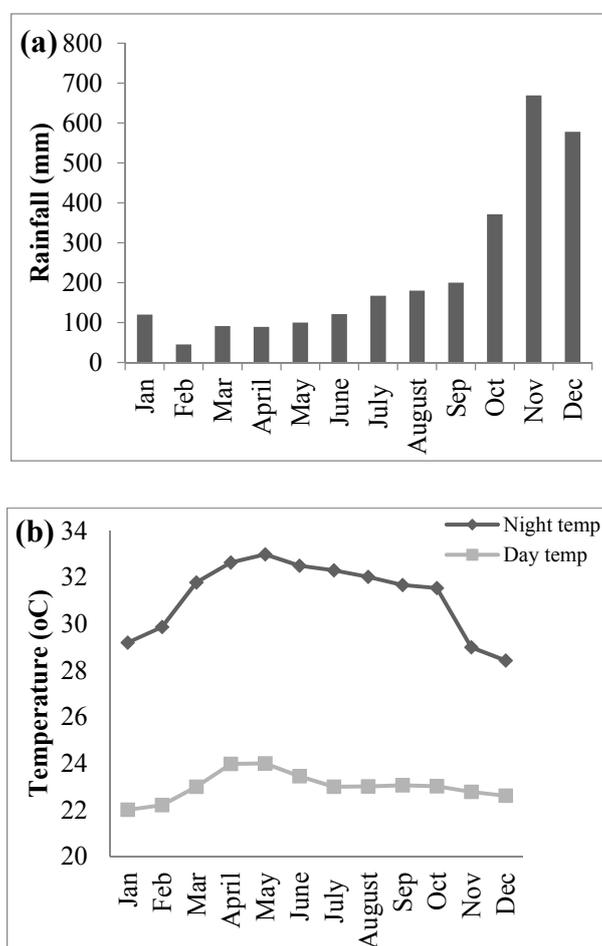


Figure 2. Monthly rainfall (a) and monthly temperature (b) at the experimental site [25].

2.7. Statistical Analysis

All data were statistically subjected to analysis of variance (ANOVA) using SAS Software program version 9.3. The treatments means were compared by Tukey's test at a 5% level of confidence [26].

3. Results

3.1. Initial Chemical Properties of the Soil

Chemical properties of the topsoil before treatment were as follows: soil pH was (3.78), CEC ($7.15 \text{ cmol}_c \text{ kg}^{-1}$), total C (2.07%), total N (0.14%), and available P (18.32 mg/kg); however, exchangeable K, Al, Ca, and Mg were 0.06, 5.21, 0.56, and $0.64 \text{ cmol}_c \text{ kg}^{-1}$, respectively. Due to a low pH, coupled with high exchangeable Al and low exchangeable basic cations, as well as low available P, the soil was unsuitable for rice cultivation without proper amelioration. The presence of yellowish jarosite within the top 50 cm of the soil profile proved that it was an acid sulfate soil that can be classified as sulfaquept [27].

3.2. Effects of Applying Amendments on Soil pH

Applying GML, basalt, or bio-fertilizer alone or in combination increased the soil pH (Table 1). The highest soil pH of 5.27 was recorded in the first season of the trial by applying GML in combination with bio-fertilizer. In the following two seasons GML and basalt, either alone or in combination with bio-fertilizer, increased pH to a level above 5.

Table 1. Effects of treatments on soil pH.

Treatments	Soil pH at Harvest			
	Before Treatment	1st Season	2nd Season	3rd Season
Control	3.78a	3.91d	3.86d	3.85e
GML	3.78a	4.75b	4.79b	4.62d
Basalt	3.78a	4.49c	5.07a	4.88b
Bio-fertilizer	3.78a	4.17c	4.14c	4.11d
GML + bio-fertilizer	3.78a	5.27a	5.12a	4.85c
Basalt + bio-fertilizer	3.78a	4.32c	5.29a	5.15a

GML = ground magnesium limestone. Means within the same column followed by the same letters are not significantly different at $p < 0.05$.

3.3. Effects of Treatments on Plant Nutrients

Table 2 shows that the highest macronutrient concentration in soil was observed in the GML or basalt treatment in combination with bio-fertilizer. Significantly higher N (0.22%), P ($33.74 \text{ mg} \cdot \text{kg}^{-1}$), and Mg ($2.97 \text{ cmol}_c \text{ kg}^{-1}$) were found in the GML treatment combined with bio-fertilizer compared to that of the control. In season three, exchangeable Ca and Mg were higher in the basalt compared to those of the GML treatment. It was found that Zn and Cu contents in the soil were increased due to the treatments (Table 2).

Table 2. Effects of applying GML and basalt with, or without, bio-fertilizer on nutrient content in the soil.

Treatments	Total N (%)	Av. P ($\text{mg} \cdot \text{kg}^{-1}$)	Exchangeable Cations				Micronutrients			
			K	Al	Ca	Mg	Fe	Zn	Mn	Cu
			(cmol _c kg ⁻¹)				(mg · kg ⁻¹)			
Control	0.11c	18.64d	0.14d	5.03a	0.65e	0.7e	189a	1.70b	6.13c	1.87e
GML	0.16b	19.83c	0.17c	0.93c	1.01d	1.27d	86c	2.00a	10.03a	3.14c
Basalt	0.20a	20.16c	0.26b	0.86c	2.14b	2.12b	79c	2.16a	9.87b	3.84b
Bio-fertilizer	0.21a	29.23b	0.24b	1.85b	0.96d	1.18d	141b	2.10a	8.21b	2.03d
GML + bio-fertilizer	0.22a	33.74a	0.31a	0.78d	2.25a	2.98a	67d	2.03a	10.71a	4.12a
Basalt + bio-fertilizer	0.20a	30.26ab	0.29a	0.69d	1.13c	1.68c	76c	2.01a	10.87a	3.81b

GML = ground magnesium limestone. Means within the same column followed by the same letters are not significantly different at $p < 0.05$.

3.4. Effects of Treatments on Chlorophyll Content

The application of amendments increased the chlorophyll content SPAD values (Table 3). The highest SPAD values of 42.33 and 45.50 were found in the GML plus bio-fertilizer treatment in the first and second seasons, respectively, followed by basalt plus bio-fertilizer treatment (44.90) in the third season.

Table 3. Effects of treatments on chlorophyll content (SPAD values) in the rice leaf.

Treatments	Chlorophyll Content (SPAD Values)		
	1st Season	2nd Season	3rd Season
	(after 90 days of sowing)		
Control	34.53d	36.2d	33.90d
GML	41.73b	43.60b	41.50c
Basalt	40.27c	42.60c	43.60b
Bio-fertilizer	42.30a	42.70c	41.3c
GML + bio-fertilizer	42.33a	45.50a	44.40a
Basalt + bio-fertilizer	41.43b	44.20b	44.90a

Means within the same column followed by the same letters are not significantly different at $p < 0.05$.

3.5. Effects of Treatments on Plant Height, Root Length, and Tiller Number

GML, basalt and bio-fertilizer application had ameliorative effects on the growth of rice (Table 4). Among the treatments, higher plant height (104 cm) and root length (23.65 cm) were found in the GML combined with bio-fertilizer treatment for the first season compared to others while, for the second and third season, there were no significant effects on the two agronomic parameters. A similar trend was observed for the tiller numbers.

3.6. Effects of Treatments on Panicle Number, Panicle Size, Unfilled Grain and Harvest Index

Application of GML and basalt with or without bio-fertilizer increased the number and size of panicles, filled grains, and harvest index (Table 5). Among the treatments, application of GML and basalt, in combination with bio-fertilizer, gave the highest panicles and harvest index; on the other hand, these treatments gave the lowest unfilled grains for all the three seasons.

3.7. Effects of Treatments on Rice Yield

Application of GML and basalt with, or without, bio-fertilizer significantly increased the grain yield (Figure 3a). The highest grain yield of $6.82 \text{ t} \cdot \text{ha}^{-1}$ was obtained for the GML plus bio-fertilizer treatment, followed by bio-fertilizer alone ($5.39 \text{ t} \cdot \text{ha}^{-1}$) in the first season. For the former, the yield was further increased in the following season ($6.07 \text{ t} \cdot \text{ha}^{-1}$). However, this was not the case for the latter treatment ($5.6 \text{ t} \cdot \text{ha}^{-1}$). In the third season, the highest rice yield of 5.94 and $5.81 \text{ t} \cdot \text{ha}^{-1}$ was recorded for the GML plus bio-fertilizer and basalt plus bio-fertilizer treatment, respectively. The straw yield followed almost the same pattern as that of the grain yield (Figure 3b).

Table 4. Effects of treatments on plant height, root length, and tiller number.

Treatments	Plant Height			Root Length			Tillers Plant ⁻¹		
	(cm)								
	1st Season	2nd Season	3rd Season	1st Season	2nd Season	3rd Season	1st Season	2nd Season	3rd Season
Control	89d	75.33d	70.36e	19.66d	17.52e	16.30d	9c	14c	26d
GML	96.41c	83.13b	78.15c	21.34b	20.67d	19.00c	19a	16b	30b
Basalt	95.31c	81.67c	76.67d	20.13c	22.53b	21.12b	16b	16b	30b
Bio-fertilizer	99.33b	81.00c	76.03d	21.46b	21.33c	20.00c	20a	15b	29c
GML + bio-fertilizer	104.00a	86.54a	81.33a	23.65a	24.41a	23.31a	21a	18a	31a
Basalt + bio-fertilizer	99.33b	85.34a	80.23b	22.30b	23.04a	22.32a	19a	17a	31a

Means within the same column followed by the same letters are not significantly different at $p < 0.05$.

Table 5. Effects of treatments on panicle number, panicle size, unfilled grain, and harvest index.

Treatments	Number of Panicle Plant ⁻¹			Size Panicle ⁻¹			Unfilled Grains (%)			Harvest Index		
	1st Season	2nd Season	3rd Season	1st Season	2nd Season	3rd Season	1st Season	2nd Season	3rd Season	1st Season	2nd Season	3rd Season
Control	7c	11d	22e	17.83e	17.67c	17.39d	26.21a	24.69a	23.77a	0.40e	0.41d	0.46d
GML	15a	13b	26c	22.60b	20.00b	20.00b	18.31d	19.46c	18.92c	0.45b	0.50c	0.49c
Basalt	16a	13b	27b	18.33d	19.87b	20.11ab	20.45b	21.59c	19.29c	0.41c	0.51b	0.49c
Bio-fertilizer	14b	12c	25d	20.10c	19.52b	19.67c	16.12f	22.97b	20.38b	0.35d	0.51b	0.49c
GML + bio-fertilizer	15a	16a	28a	24.23a	21.71a	21.65a	17.82e	15.76d	13.89d	0.55a	0.53a	0.53a
Basalt + bio-fertilizer	15a	15a	27b	23.00b	21.16a	21.37a	19.24c	15.28d	14.13d	0.47b	0.52b	0.50b

Means within the same column followed by the same letters are not significantly different at $p < 0.05$.

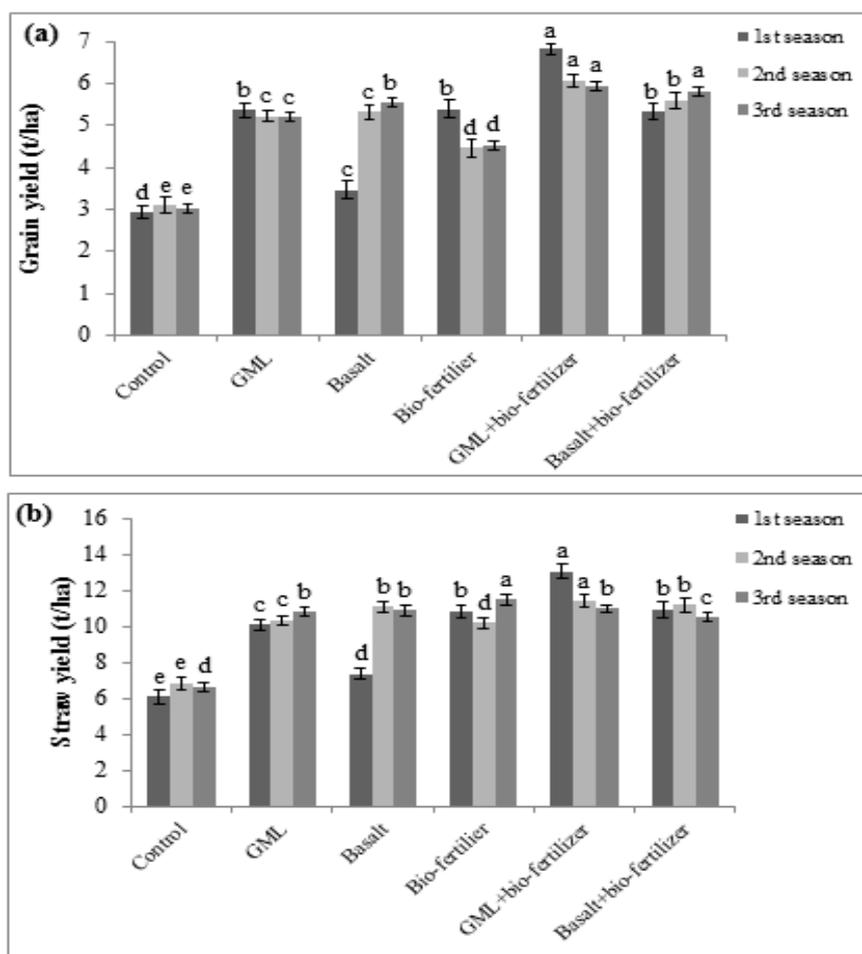


Figure 3. Effects of applying GML and basalt with, or without, bio-fertilizer on rice growth: (a) grain yield and (b) straw yield. GML = ground magnesium limestone. Means within the same column followed by the same letters are not significantly different at $p < 0.05$ ($n = 18$).

3.8. Effects of Treatments on Nitrogen, Phosphorus, Potash (NPK) and Protein Content in the Tissue or Grain

Nitrogen, phosphorus, and potash (NPK) contents in the plant tissue and/or grain in season three were significantly higher due to the application of the amendments compared to those without (Table 6). The highest contents of NPK in the tissue and rice grain as well as protein content in the grain were due the application GML and basalt with bio-fertilizer.

Table 6. Effects of treatments on nitrogen, phosphorus, and potash (NPK) and protein content in the tissue or grain.

Treatments	N		P		K		Protein Content in Grain
	Plant	Grain	Plant	Grain	Plant	Grain	
Control	0.46c	0.93c	0.12c	0.15c	1.31c	0.22d	5.53c
GML	0.66b	0.98b	0.15a	0.28b	1.48b	0.28b	5.83b
Basalt	0.68a	0.97b	0.16a	0.29b	1.49b	0.28b	5.77b
Bio-fertilizer	0.67b	0.97b	0.14b	0.27b	1.45b	0.25c	5.77b
GML + bio-fertilizer	0.71a	1.06a	0.17a	0.33a	1.54a	0.31a	6.30a
Basalt + bio-fertilizer	0.70a	1.03a	0.17a	0.32a	1.56a	0.32a	6.12a

GML = ground magnesium limestone. Means within the same column followed by the same letters are not significantly different at $p < 0.05$.

3.9. Effects of Treatments on Al, Fe, Ca, Mg, and Si Content in the Plant Tissue

In the third season, the highest Al content of 0.05% and Fe of 0.11% were found in the control treatment (Table 7). Applying the treatments increased Ca and Mg content in the tissue, with the highest value of 0.24% and 0.62% being reported for the GML and basalt with bio-fertilizer, respectively. Si content was increased by basalt application, with the highest value of 0.56% found in the basalt plus bio-fertilizer treatment.

Table 7. Effects of treatments on Al, Fe, Ca, Mg, and Si content in plant tissue.

Treatments	Metal Concentration in the Plant				
	Al	Fe	Ca	Mg	Si
Control	0.05a	0.11a	0.08d	0.29d	0.15d
GML	0.03b	0.07b	0.17b	0.43b	0.25c
Basalt	0.02b	0.09b	0.14c	0.47b	0.53a
Bio-fertilizer	0.03b	0.09b	0.13c	0.35c	0.28c
GML + bio-fertilizer	0.02b	0.05b	0.23a	0.62a	0.49b
Basalt + bio-fertilizer	0.02b	0.06b	0.24a	0.56a	0.56a

GML = ground magnesium limestone. Means within the same column followed by the same letters are not significantly different at $p < 0.05$.

3.10. Effects of Treatments on Al Form in Soil

Applying GML, basalt and bio-fertilizer, either alone or in combination, decreased exchangeable Al (Figure 4). Significantly higher exchangeable Al of 5.03 and weakly-bound Al of 3.04 $\text{cmol}_c \text{kg}^{-1}$ were observed in the control treatment. In the third season, the highest strongly-bound Al of 9.34 $\text{cmol}_c \text{kg}^{-1}$ was recorded in the GML with bio-fertilizer, which was not significantly different from that of the basalt with bio-fertilizer treatment (8.67 $\text{cmol}_c \text{kg}^{-1}$).

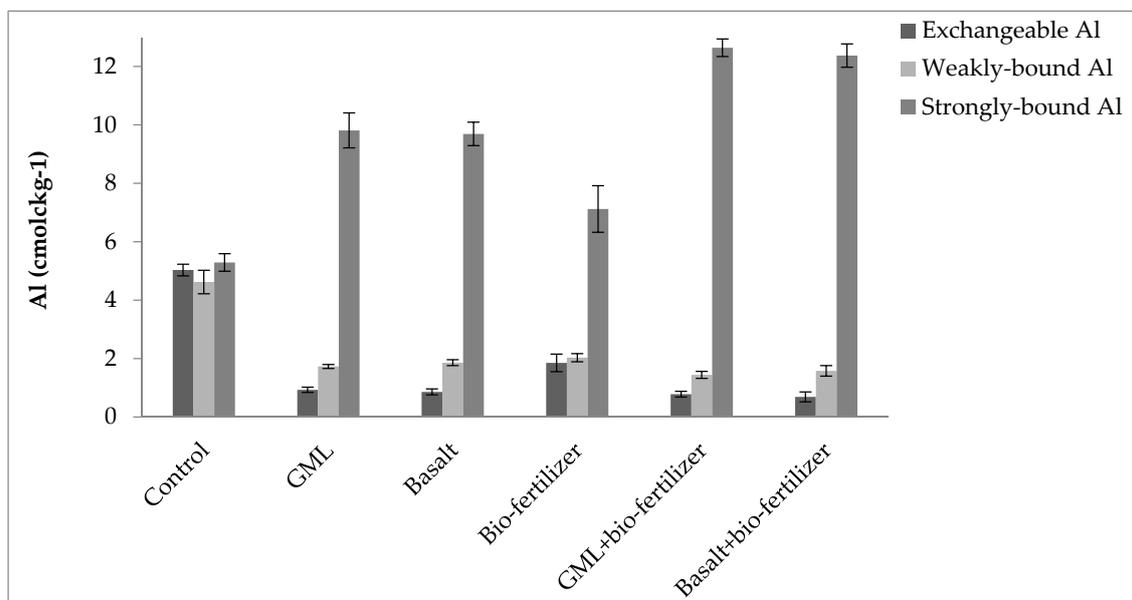


Figure 4. Effects of treatments on the form of Al in soil. GML = ground magnesium limestone. Means within the same column followed by the same letters are not significantly different at $p < 0.05$ ($n = 18$).

3.11. Relationship between pH with Ca, Mg, Al, and Fe

There were positive linear correlations between soil pH and exchangeable Ca ($Y = 3.112 + 0.689x$; $R^2 = 0.715$) and exchangeable Mg ($Y = 3.217 + 0.723x$; $R^2 = 0.765$) (Figure 5a,b). Applying GML and basalt with or without bio-fertilizer increased Ca and Mg content in the soil, and this increase partly contributes to the pH increase because they are basic metals.

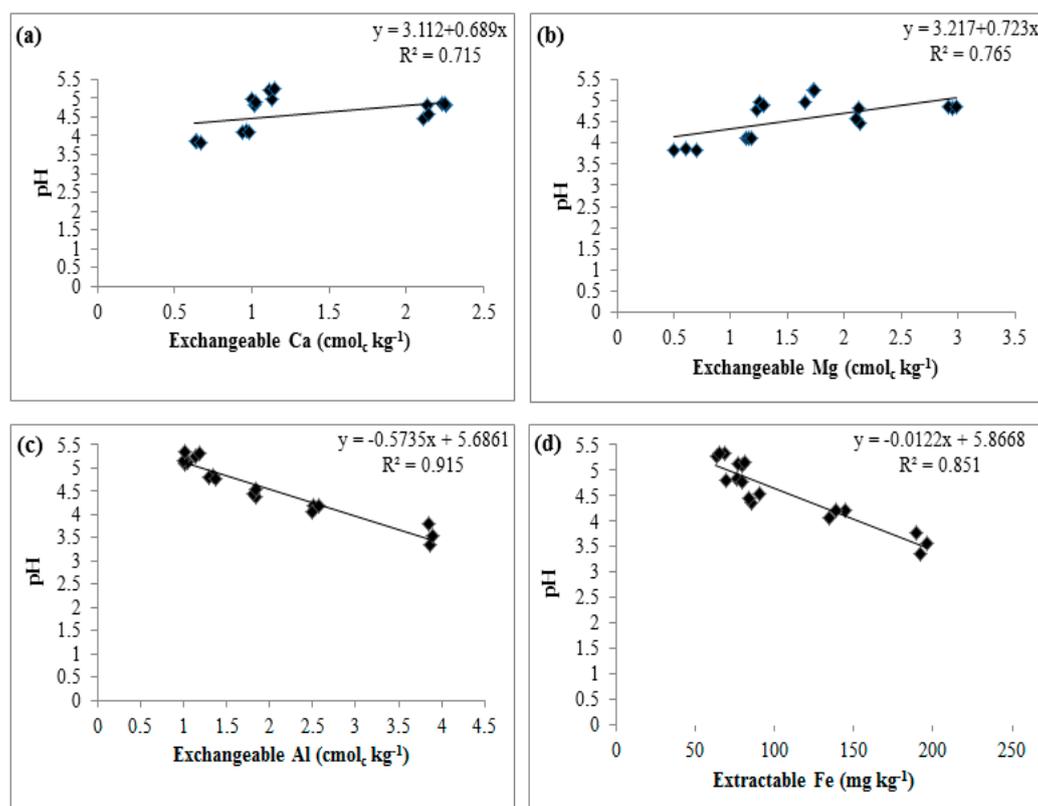
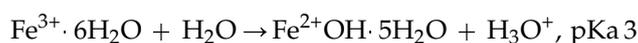
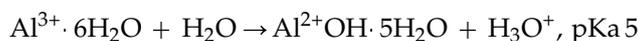


Figure 5. Relationship between soil pH and exchangeable Ca (a); soil pH and exchangeable Mg (b); soil pH and exchangeable Al (c); and soil pH and extractable Fe (d).

The correlation between soil pH and exchangeable Al or extractable Fe was negative, and the respective equations representing the relationship are given by $Y = -0.573x + 5.686$ ($R^2 = 0.915$) and $Y = -0.012x + 5.866$ ($R^2 = 0.851$) (Figure 5c,d). If both of these metals were present in high concentration in the soils, water pH would be adjusted accordingly to the level close to their respective pKa values [26]:



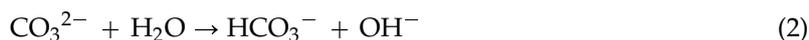
Hence, without applying GML, basalt, or bio-fertilizer, the pH of the water in the experimental plots would be around 3–4. This was, in fact, the case for the pH of water in the paddy fields throughout Peninsular Malaysia [3].

4. Discussion

4.1. Effects of Applying GML on Soil Properties

Applying GML into the soil resulted in the following reactions:

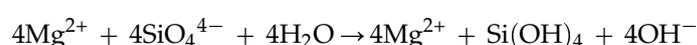




Soil pH increased readily upon reaction of GML with water. When pH was above 5, Al in the water would precipitate as inert Al-hydroxides, making it unavailable. This is because the pKa of Al is 5. The pKa of Fe is 3; hence, when water pH was above 3, Fe would precipitate as Fe hydroxides. This means applying GML at the rate proposed in this study would result in the decrease of Fe and Al to the minimal level and so they were no longer a threat to rice growth. Even if soil pH was slightly lower than 5, rice would grow as it could defend itself against Al^{3+} and/or Fe^{2+} toxicity via a special mechanism. Furthermore, under the stress of Al^{3+} and/or Fe^{2+} , rice roots can secrete organic acids that eventually inactivated the toxic ions via a chelation mechanism [28].

4.2. Effects of Basalt Application on Soil Properties

Basalt, an igneous rock, is composed of olivine, pyroxene, amphibole, and feldspars. Olivine disintegrated and eventually dissolved according to the following reaction [3,27]:



This is not a balanced reaction; however, Alia et al. [28] reported that if olivine were to dissolve in soils, the plausible reaction would be as above. This statement is supported by the study of [7,29]. The infertility of the soil had been ameliorated by basalt application. Not only was water pH increased by the hydrolysis of SiO_4^{4-} to the level higher than that of the CO_3^{2-} , Mg was made available. The dissolution of basalt would add other macronutrients, such as Ca, K, and P into the soil.

However, silicic acid released by the dissolution of basalt would be taken up by the rice plants [30] that resulted in prevention of rice blast [31]. This phenomenon was confirmed by the study of Massey and Hartley [32]. Basalt contained 30%–40% silica and so it would increase soluble Si in the soil that could be taken up by rice (Table 7).

4.3. Effects of Applying Bio-Fertilizer on Soil

The bio-fertilizer was fortified with N_2 -fixing and phosphate-solubilizing bacteria (PSB). Its application would result in the fixation of some nitrogen from the air, which was otherwise needed to be supplied from inorganic N-fertilizer sources. Hence, using this bio-fertilizer, to a certain extent, might reduce the cost of rice production. This agronomic practice is the kind of green technology that we are looking for in rice production in the tropics.

High Al and/or Fe concentration in the water not only caused toxicity to rice, but also reduced the availability of P [33]. The low available P in the acid sulfate soil could be somewhat alleviated by the PSB, which were able to increase soil pH to a level above 5 [8]. The PSB were also able to dissolve Al-Fe-phosphate, making P more available for rice production than it would be otherwise [8] (Figure 3a).

Applying GML and basalt in combination with bio-fertilizer resulted in the increase of the strongly-bound Al (Figure 4). This means that Al^{3+} present in the water would be fixed by the organic acids released by the PSB and was subsequently deactivated, and this was also true for the case of Fe^{2+} (Figure 6). Hence, this bio-fertilizer could potentially increase the productivity of rice planted in the acid sulfate soils in Malaysia (Kelantan), Thailand (Bangkok Plains), Vietnam (Mekong Delta), and Indonesia (Kalimantan).

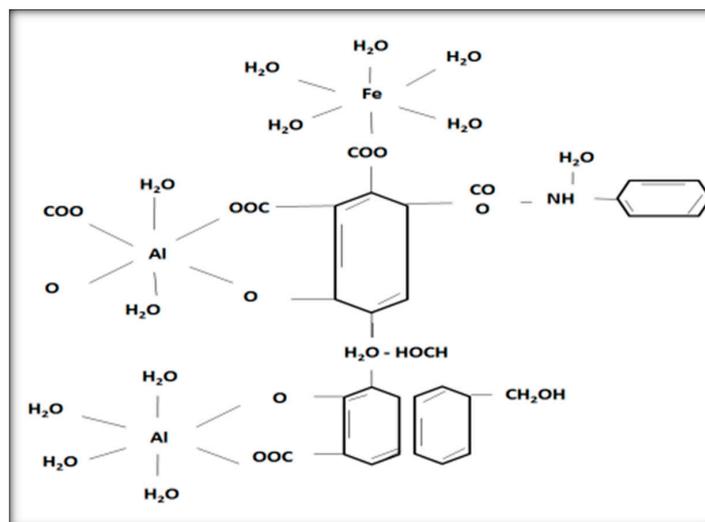


Figure 6. The mechanism of Al and Fe chelation shown by an organic material in soil.

4.4. Residual Effects of GML and Basalt Application on Nutrients

Applying the amendments at the appropriate rates and time would result in the significant increase of exchangeable Ca^{2+} . This extra Ca would help reduce the effect of Al toxicity [34]. Dissolution of GML or basalt increased the concentration of Zn and Cu in the soil (Table 2). GML in Malaysia contained Zn and Cu at 29.5 and $16.6 \text{ mg} \cdot \text{kg}^{-1}$, respectively [35]. Sahibin et al. [36] found that basalt in Segamat, Malaysia, contained $41\text{--}196 \text{ mg} \cdot \text{kg}^{-1}$ Zn, while its Cu content was $54\text{--}279 \text{ mg} \cdot \text{kg}^{-1}$. Applying this basalt into the soil would increase the availability of Zn and Cu for rice consumption that would eventually improve its growth (Table 2). Panhwar et al. [37] found that rice grown on riverine soils in the Kelantan Plains, Malaysia, responded positively to Zn and Cu application that consequently enhanced the quality of rice grain.

This study showed that after three seasons of growing rice on the same plots, there were still sufficient amounts of Ca, Mg, Zn, and Cu contents in the soil for rice requirement. That means applying GML or basalt once at the rate proposed in this study can last at least up to three seasons. However, we tend to believe that the rice production can even be sustained for a longer period of time.

4.5. Residual Effects of GML, Basalt with or without Bio-Fertilizer Application on Rice Growth

The enhanced rice growth and the eventual grain yield increase (Figure 3) were partly attributed to the increase of Zn and Cu content in the soil (Table 3). Chlorophyll content in the leaf was higher in the GML with bio-fertilizer treatment compared to that of basalt with bio-fertilizer treatment probably because the ameliorative effects of the latter was not fully realized yet due the slow rate of basalt dissolution [7,29]. The pattern of changes in the plant height followed that of the chlorophyll content (Table 5). Regarding the tiller number, GML and basalt in combination with bio-fertilizer treatment gave the best results (Table 5). The treatment that gave the highest harvest index was the GML with bio-fertilizer treatment. It looks as though the best agronomic practice to alleviate the infertility of acid sulfate soils for sustainable rice cultivation is the application of GML with bio-fertilizer. However, we are still of the estimation that the best option would be to apply basalt in combination with bio-fertilizer, considering the latter long-term ameliorative effects on rice yield (Figure 3a).

The grain yield was about $3 \text{ t}^{-1} \cdot \text{ha}^{-1} \cdot \text{season}^{-1}$ in the control treatment (Figure 3a). When GML was applied, it increased to about $5 \text{ t}^{-1} \cdot \text{ha}^{-1} \cdot \text{season}^{-1}$. The reason for this is that the fertility of the soil had been improved somewhat by GML application, which increased the water pH, while the concentration of Al^{3+} and/or Fe^{2+} was concomitantly reduced. The growth of rice was further enhanced by the increase in Ca, Mg, Zn, and Cu contents [35,38]. Basalt treatment gave low grain yield

in the first season, but with time, it increased to that of the GML treatment. GML with bio-fertilizer treatment gave a grain yield of about $6 \text{ t}^{-1} \cdot \text{ha}^{-1}$ season, the highest value recorded in this study; however, it tended to decrease with time. By contrast, basalt with bio-fertilizer treatment, which gave comparable grain yield to that of the former, tended to increase with time (Figure 3a). This is consistent with our study that basalt with bio-fertilizer treatment would produce long-term ameliorative effects that sustained rice production in the long run.

In Malaysia, GML costs USD 70 per ton while basalt only costs USD 10 per ton. However, basalt dissolved slowly; hence, its ameliorative effects could only be felt significantly from the second season onwards. However, the problem of acidity and toxicity due to Al^{3+} and/or Fe^{2+} was alleviated partly by the PSB present in the bio-fertilizer. These PSB could also produce plant growth phytohormones, such as indole-3-acetic acid, that further enhanced rice growth and eventually contributed to increased rice yield [39,40]. Protein content in the rice grain increased due to the application of GML or basalt that supplied Zn and Cu (Tables 2 and 6), which was in line with the findings of Panhwar et al. [37].

5. Conclusions

The productivity of acid sulfate soils in Malaysia can be enhanced by applying GML and basalt with or without bio-fertilizer fortified with N_2 -fixing and phosphate-solubilizing bacteria. GML and basalt contained some Zn and Cu; thus, their application would increase Zn and Cu reserves in the soil. Applying basalt in combination with bio-fertilizer appeared to be the best agronomic option to improve the fertility of acid sulfate soils for sustainable rice production. This is because, besides increasing Ca and Mg, as well as raising the pH to precipitate Al^{3+} and/or Fe^{2+} , it produced silicic acid, which could be taken up by rice plants, preventing the outbreak of rice blast. Ground basalt is cheaper compared to GML; however, basalt dissolved slowly even though under acidic conditions. Hence, its ameliorative effects could only be felt significantly from the second season onwards. Acidity had been partly alleviated by PSB, which increased water pH. PSB in the bio-fertilizer not only increased the available P, but also helped release organic acids which inactivated Al^{3+} and/or Fe^{2+} via the process of chelation. It was proven by earlier studies that the bio-fertilizer could also produce phytohormones that further enhanced rice growth. The use of bio-fertilizer fortified with N_2 -fixing bacteria is a green technology that would reduce NO_3^- and/or NO_2^- pollution and also cut down cost of rice production. The findings of this study encourage the assumption that that the infertility of acid sulfate soils occurring in the ASEAN countries can be alleviated for sustainable rice production.

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Conflicts of Interest: The authors declare no conflict of interest.

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