

## EFFECT OF DIFFERENT REMEDIATION TECHNIQUES AND DOSAGES OF PHOSPHORUS FERTILIZER ON SOIL QUALITY AND *KLEKAP* PRODUCTION IN ACID SULFATE SOIL AFFECTED AQUACULTURE PONDS

Akhmad Mustafa<sup>\*)</sup> and Jesmond Sammut<sup>\*\*)</sup>

### ABSTRACT

Acid sulfate soils (ASS) contain sufficient pyrite which, when oxidised following excavation for brackishwater aquaculture ponds, will generate acid and mobilise toxic metals. Production in affected ponds can be low due to poor growth of shrimp and fish, mass mortalities of stock and low plankton blooms. The resultant low soil pH can also cause poor *klekap* production due to the retention of phosphorus associated with elevated concentrations of Fe and Al in the pond soils. A series of experiments was conducted to determine the effects of different soil amelioration techniques and dosage of phosphorus (P) on soil and *klekap* production under laboratory conditions. The treatments consisted of two factors. The first factor tested was different techniques for ASS improvement (non-improvement, improvement through liming and improvement through remediation involving forced oxidation of pyrite, flooding and flushing of oxidation products). The second factor tested was phosphorus dosages, that is, with phosphorus and without phosphorus-based fertilizer. Each treatment had three replications. The experiment showed that liming and remediation had the same effect on several soil variables; they raised the soil pH ( $pH_F$ ,  $pH_{FOX}$ ,  $pH_{KCl}$ ) and decreased  $S_{POS}$ , Fe and Al. Remediation of ASS decreased retention of P and increased available-P of soil, whereas liming did not show a significant effect on retention of P and available-P in the doses used for this experiment. The interaction between the different soil improvement techniques and phosphorus fertilising showed a significant effect on *klekap* production with the highest *klekap* production of 23.21 mg/cm<sup>2</sup> found in remediated soil and with a phosphorus fertiliser dosage of 75 kg/ha.

**KEYWORDS:** remediation, phosphorus, acid sulfate soil, *klekap*

### INTRODUCTION

Acid sulfate soil (ASS) is soil or sediment which contains cubic or framboidal pyrite ( $FeS_2$ ) (Dent, 1986; Schaetzl and Anderson, 2005) usually found in Holocene coastal environments (Sammut *et al.*, 1996). ASS cover an area of around 6.7 million hectares in Indonesia and many brackishwater aquaculture ponds have been severely degraded by soil acidification and the associated increase in toxic metal species. Many ponds constructed in ASS are abandoned because of low

productivity (Sammut and Hanafi, 2000). However, many farmers are unaware of the causes of poor production but identify low plankton blooms, poor growth of fish and shrimp, mass mortalities of stock, dyke cracking and iron precipitation as common and related problems (Sammut, 1999). Although metals may be directly toxic to aquatic organisms, indirect effects of low pH and metals may also occur (Sammut, 1999). Low phosphorus (P) can lead to poor natural food production and impact on post larvae in the early stages of production. Farmers commonly report having to add much

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<sup>\*)</sup> Research Institute for Coastal Aquaculture, Maros, Indonesia

<sup>\*\*)</sup> School of Biological, Earth and Environmental Sciences, The University of New South Wales, Sydney, NSW, Australia

higher amounts of fertilizer to compensate for the loss of P although they do not fully understand the processes that cause the P deficiencies.

Phosphorus builds the pyrophosphate bond in ATP (adenosine triphosphate) which is an important energy source for biochemical activity in living plant cells; it is also an important cell builder (Noggle and Fritz, 1986). Phosphorus is the key nutrient in the production of natural food in fresh water and brackish water ponds (Boyd, 1990). The soil pH plays an important role in P dynamics in aquaculture ponds. Under alkaline conditions  $\text{HPO}_4^{2-}$  is the dissolved form of P. In the normal pH range, two forms of the P ion ( $\text{HPO}_4^{2-}$  and  $\text{H}_2\text{PO}_4^-$ ) are present (Essington, 2004) and are absorbed by plants, macroalgae and phytoplankton (Tisdale and Nelson, 1975).

The lack of the element P in soil can be due to an absence of P in the parent material. The available-P is often absorbed by forms of Fe and Al which are found in most soils (Prasetyo *et al.*, 2001a, b) but prevalent in ASS. Acidic soils, where the availability of the dissolved elements Fe, Al and Mn is greater, tend to absorb P which will produce phosphate hydroxide (Hakim *et al.*, 1986). At high concentrations, Fe and Al in pond soils will form insoluble P compounds (Poernomo, 1983; Tu *et al.*, 1993), such as  $\text{FePO}_4$  or  $\text{AlPO}_4$  (Kselik *et al.*, 1992). Consequently, P fertilizer is essential for the production of natural feed (*klekap*, plankton). *Klekap* (a benthic complex of blue-green algae, protozoa, diatoms, bacteria and detritus) is an essential natural feed for fish and shrimp in Indonesian brackishwater ponds, especially in ponds which are managed in traditional and semi-intensive systems. Phosphorus-based fertilizer can result in the accumulation of P residue because the P nutrient has an efficiency of about 20% (Adiningsih *et al.*, 1989).

In order to improve *klekap* production in ponds, ASS usually require remediation to minimize the effects of Fe and Al on P availability. Remediation of ASS is required at the start of the cycle and preferably as an important component of major pond preparation between production cycles and in ponds that are rehabilitated. Remediation of pond soils in the form of drying to oxidise pyrite, flooding to mobilise oxidation products and flushing away the elements and compounds causing acidity and pondwater pollution, has

been attempted by Brinkman and Singh (1982), Singh (1982a, b) and Poernomo (1983, 1986). The other common form of ASS remediation is liming to reduce the actual and potential acidity of the pond soils. By contrast, when soil pH is above 7.0, various compounds of Ca and P can precipitate and also reduce P availability (Tisdale and Nelson, 1975). The purpose of the present study is to test the effectiveness of various techniques for improving ASS to maximize the available P and consequently increase *klekap* production in severely acidified pond soils.

## MATERIALS AND METHODS

### Experimental Apparatus

The experiment was conducted in a 'plastic house' at the Research Institute for Coastal Aquaculture (RICA) in Maros Regency, South Sulawesi Province, Indonesia for five months. The plastic house acts in the same way as a green house and also helps to minimise external inputs such as rainfall.

The experimental containers consisted of 18 fiberglass aquaria each measuring 0.50 m in length, 0.30 m in width and 0.30 m in height. The aquaria were placed on tables 0.75 m high.

### Soil Materials

The soil materials for the experiment were collected from the bottom of an ASS-affected pond located in Lampuara Subdistrict, Ponrang District, Luwu Regency, South Sulawesi Province. The soil material was collected at a depth of 0.10-0.40 m from the soil surface and as quickly as possible was placed in a plastic bag to minimise soil contact with air and to maintain soil saturation during transportation. The samples were kept at 4°C to prevent bacterially mediated oxidation of pyrite. Shells and mangrove roots were removed from the soil samples and the bulk soil sample was then mixed and placed in a closed fiberglass container in the Wet Laboratory of Research Institute for Coastal Aquaculture (RICA) in Maros Regency.

### Experimental Procedure

Each aquarium was filled with soil with a wet weight of 12.0 kg (water content of 27.03%) or a thickness of 7.5 cm. The first factor was the technique used for improving the soil (treatments included: without improvement, soil improvement through liming, soil

improvement through remediation (drying, flooding and flushing) and the second factor was the different dosage for P fertilizer (without phosphorus fertilizer, with phosphorus fertilizer). For each treatment there were three replications resulting in 18 experimental units. The experiment was set up based on Randomized Completely Factorial Design (Sokal and Rohlf, 2003).

Lime ( $\text{CaCO}_3$ ) was applied with a dosage based on the soil  $S_{\text{POS}}$  (Ahern *et al.*, 1998a), and the remediation treatment consisted of: drying for 14 days, soil flooding with water of salinity 25 ppt for ten days, and this was followed by flushing. The procedure was repeated three times (Mustafa and Sammut, unpublished data). For the treatment without soil improvement, the soil was flooded with water and was placed in a roofed enclosure to reduce evaporation during the processes of remediation. For the treatment involving P fertilizer, the dosage of triple superphosphate (TSP,  $\text{Ca}(\text{H}_2\text{PO}_4)_2$ ) fertilizer was 75 kg/ha (1.125 g/aquarium) in accordance with the findings of Mustafa (2007). After the application of each soil treatment, the soil was dried for one week and urea ( $\text{H}_2\text{NCONH}_2$ ) fertilizer was applied to the wet soil with a dosage of 25 kg/ha (0.375 g/aquarium) along with TSP fertilizer. Water with a salinity of 25 ppt (Arifin, 1984) was brought into the aquarium until the water was 5 cm high. The water in the aquaria was allowed to evaporate until the soil became dry again and the fertilizer had been absorbed into the soil. The water was brought in again in stages until it was 15 cm high in accordance with the shallow water method for growing *klekap* (Ilyas *et al.*, 1987). The experiment was continued for six weeks, beginning with adding water to grow the natural food. During the process all the aquaria were closed with a transparent plastic sheet from 17:00 hours till 7:00 hours to prevent the entry of foreign objects during the night hours.

### Sample Analyses

There was an initial soil sample, that is before the application of treatments for soil improvement techniques; after the application of treatments for soil improvement techniques; and every two weeks after filling with water to grow natural food for all soil variables except total-P, retention of P and available-P. The forms of P were analysed in the initial soil sample; after the application of treatments for soil improvement techniques; and every three

weeks after filling aquaria with water to grow natural food. The soil quality variables which were analysed directly in the 'plastic house' included: field pH ( $\text{pH}_f$ ) and field pH after oxidation with 30% hydrogen peroxide ( $\text{pH}_{\text{FOX}}$ ) (Ahern and Rayment, 1998). For laboratory analyses, soil samples weighing around 50 g wet from each aquarium was dried in an oven at 80–85°C for a minimum of 48 hours (Ahern dan Blunden, 1998). The dried soil was ground until it could be passed through screen (30 mesh). The soil quality variables covered pH measured from an extract of KCl 1 M ( $\text{pH}_{\text{KCl}}$ ), pH measured from KCl which was totally oxidised ( $\text{pH}_{\text{OX}}$ ), sulfur analysed from an extract KCl 1 M ( $S_{\text{KCl}}$ ), sulfur analysed after total oxidation with 30% hydrogen peroxide ( $S_p$ ), peroxide oxidisable sulfur ( $S_{\text{POS}} = S_p - S_{\text{KCl}}$ ), total actual acidity (TAA, in 1 M KCl which is titrated to a pH of 5.5), total potential acidity (TPA, in 1 M KCl which is totally oxidized and titrated to a pH of 5.5), total sulfidic acidity (TSA = TPA - TAA), pyrite (Ahern and Rayment, 1998; Ahern *et al.*, 1998b, 1998c), Fe and Al (Menon, 1973; Melville, 1993). Available-P was determined by method of Bray and Kurtz (1945). Total-P was determined by digesting soil with 25% of HCl. Retention of P was determined using the method outlined by Sukanta *et al.* (1989).

*Klekap* production was quantified by measuring the biomass of *klekap* each week. The *klekap* samples were collected using a 1 inch (2.54 cm) diameter pipe at five points in each experimental unit. The *klekap* which had been separated from the bottom soil were placed into bottles. The *klekap* biomass was calculated by weighing the *klekap* sample in a porcelain dish. The sample was dried in an oven at a temperature of 105°C for 24 hours. After it reached room temperature, it was weighed again to calculate the dry weight. The sample was then ashed in a small furnace for six hours at a temperature of 600°C. Beforehand the *klekap* sample was treated with 0.5 mL nitrate acid and cooled down in a dessicator and weighed again. The *klekap* production was obtained using the following formula (Arifin, 1984):

$$\text{Klekap production} = \frac{\frac{n}{s} \frac{W}{A_{T-Q}} \times T}{A} \times B$$

Where:

$n$  = weight lost after ashing

$s$  = sub sample taken from ashing

$W$  = dry weight after oven-drying to constant weight at 105°C

$T$  = original wet-weight of *klekap*

$Q$  = weight of qualitative analysis

$A$  = area of the sampler.

### Data Analyses

Before the calculation of mean or differences of pH values, the pH values were converted to activity of hydrogen ions. Normal distribution and variance homogeneity of each dataset were tested by Kolmogorov-Smirnov and Levene Test. The data were transformed to satisfy the assumptions of homogeneity of variance. The effects of different techniques to improve soil, different phosphorus fertilizers, and their interaction were analysed using two-way analysis of variance (ANOVA) performed separately during each sampling period for all data. When the ANOVA showed no interaction was occurring, different techniques to improve soil treatments were combined within different P fertilizers treatments, and different P fertilizers treatments were combined within different techniques to improve soil treatments for further analysis. Where appropriate, the Duncan Multiple Comparisons Test was applied for multiple comparison among means. Standard error in each variable and treatment was calculated and expressed as mean ± SE. The coefficient correlation ( $r$ ) and significance of the model ( $P$ ) were used to express the relationship between the available P and other soil variables. The level for statistical significance was set at 0.05%. All statistical analyses were performed with the computerised statistical package, Statistical Product and Service Solution (SPSS) version 9.0, with the exception of the Duncan Multiple Comparisons Test which was used when there was an interaction between different techniques to improve soil and different phosphorus fertilizers following the method of Gomez and Gomez (1984).

## RESULTS AND DISCUSSION

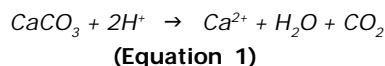
### Soil Quality

The properties of soil used in this experiment are summarized in Tables 1, 2 and 3. The chemical analysis of the soil indicates that the ASS had an initial  $pH_F$  of 5.22,  $pH_{FOX}$  of 0.56,  $pH_{KCl}$  of 4.88 (Table 1),  $S_{POS}$  1.5643%, TPA 376.2 mol H<sup>+</sup>/tonne, pyrite 1.8764%, Fe 8,674 mg/L, Al 354 mg/L (Table 2), total-P 198 mg/L, retention of P 47% and available-P 4.50 mg/L

(Table 3). The soil  $pH_F$  was moderately acidic at pH 5.22 and then became more acidic as the soil was dried, presumably due to the oxidation of some pyrite still present in the soil ( $pH_{KCl}$  4.88).  $SO_4$  is produced when pyrite undergoes oxidation or exposures to the atmosphere. The changes in some chemical properties of the soil with time and different treatments are discussed below in detail.

The different techniques for soil improvement had a significant effect on all acidity variables for ASS, that is, the variables being analysed in this experiment. Liming and remediation increased the pH ( $pH_F$ ,  $pH_{FOX}$ ,  $pH_{KCl}$ ) of ASS significantly compared to soil without improvement. Liming increased the soil  $pH_F$  up to 1.28 units whereas remediation by forced oxidation, flooding and flushing only increased it to 0.97 units. Although the soil was submerged for the treatment without soil improvement, however for the treatment without soil improvement there an increase in soil  $pH_F$  of 0.53 units also occurred. This increase in soil pH was a result of flushing removing the oxidation products.

The agricultural lime ( $CaCO_3$ ) which was applied to the ASS reacted with the source of acidity as show by the reaction below:



The Ca which originated from the agricultural lime was also able to replace the Fe and Al ions in the soil absorption complex (Tisdale and Nelson, 1975; Tan, 1991; Baruvka and Rechcigl, 2003). The increase of Ca in the soil will reduce the concentration of elements causing soil acidity. High concentrations of  $Fe^{2+}$ ,  $Al^{3+}$  and  $H^+$  cause soil acidity (Gosavi, 2004; Gosavi *et al.*, 2004) through mineral and proton acidity. The effect of  $H^+$  on acidity generation is immediate whereas Al can release acidity through hydrolyses. The oxidation of  $Fe^{2+}$  to  $Fe^{3+}$  produces acid and consume dissolved oxygen in water (Cook *et al.*, 2000). Al is a major cation causing soil acidity in the ASS (Sammut *et al.*, 1996). Large increases in soil pH through liming may rapidly precipitate  $Al^{3+}$ . Removal of Al via precipitation to gibbsite (Friesen *et al.*, 1980) most likely occurs when the solution pH increases to a value above 5.0. In limed ASS, the decrease in Al is also due to the effect of lime and increase in soil pH at which the solubility of Al is thought to decrease and cease beyond pH 4.5 (Singh, 1982a). Generally,

Table 1. The pH and  $S_{\text{pos}}$  (mean  $\pm$  SE) of soil at different techniques to improve acid sulfate soil and phosphorus fertilizer in different times

Variables	Treatments	Before treated	Time			
			0 week	2 weeks	4 weeks	6 weeks
pH <sub>F</sub>	Without improving	5.22	5.52 $\pm$ 0.17 <sup>a</sup>	5.75 $\pm$ 0.10 <sup>a</sup>	5.69 $\pm$ 0.14 <sup>a</sup>	
	Liming	5.22	6.21 $\pm$ 0.04 <sup>b</sup>	6.45 $\pm$ 0.07 <sup>b</sup>	6.33 $\pm$ 0.04 <sup>b</sup>	
	Remediation	5.22	6.16 $\pm$ 0.06 <sup>b</sup>	6.11 $\pm$ 0.14 <sup>b</sup>	6.19 $\pm$ 0.15 <sup>b</sup>	
pH <sub>rox</sub>	Without phosphorus	5.22	6.02 $\pm$ 0.08 <sup>a</sup>	6.09 $\pm$ 0.12 <sup>a</sup>	6.05 $\pm$ 0.11 <sup>a</sup>	
	With phosphorus	5.22	5.90 $\pm$ 0.17 <sup>a</sup>	6.12 $\pm$ 0.13 <sup>a</sup>	6.14 $\pm$ 0.11 <sup>a</sup>	
	Without improving	0.56	0.68 $\pm$ 0.07 <sup>a</sup>	0.88 $\pm$ 0.03 <sup>a</sup>	0.72 $\pm$ 0.04 <sup>a</sup>	
pH <sub>KCl</sub>	Liming	0.56	1.41 $\pm$ 0.13 <sup>b</sup>	1.40 $\pm$ 0.15 <sup>b</sup>	0.98 $\pm$ 0.15 <sup>a</sup>	
	Remediation	0.56	1.14 $\pm$ 0.07 <sup>b</sup>	1.27 $\pm$ 0.05 <sup>b</sup>	0.97 $\pm$ 0.07 <sup>a</sup>	
	Without phosphorus	0.56	1.14 $\pm$ 0.14 <sup>a</sup>	1.31 $\pm$ 0.11 <sup>a</sup>	0.84 $\pm$ 0.04 <sup>a</sup>	
pH <sub>KCl</sub>	With phosphorus	0.56	1.01 $\pm$ 0.12 <sup>a</sup>	1.13 $\pm$ 0.11 <sup>a</sup>	0.93 $\pm$ 0.11 <sup>a</sup>	
	Without improving	4.88	4.96 $\pm$ 0.24 <sup>a</sup>	4.65 $\pm$ 0.15 <sup>a</sup>	4.64 $\pm$ 0.15 <sup>a</sup>	
	Liming	4.88	7.57 $\pm$ 0.05 <sup>c</sup>	6.66 $\pm$ 0.22 <sup>b</sup>	6.11 $\pm$ 0.36 <sup>b</sup>	
S <sub>pos</sub> (%)	Remediation	4.88	6.57 $\pm$ 0.12 <sup>b</sup>	6.62 $\pm$ 0.11 <sup>b</sup>	5.46 $\pm$ 0.10 <sup>b</sup>	
	Without phosphorus	4.88	6.40 $\pm$ 0.37 <sup>a</sup>	6.07 $\pm$ 0.33 <sup>a</sup>	5.34 $\pm$ 0.26 <sup>a</sup>	
	With phosphorus	4.88	6.35 $\pm$ 0.43 <sup>a</sup>	6.15 $\pm$ 0.50 <sup>a</sup>	5.47 $\pm$ 0.30 <sup>a</sup>	
S <sub>pos</sub> (%)	Without improving	1.5643	1.4207 $\pm$ 0.0855 <sup>b</sup>	1.3054 $\pm$ 0.1006 <sup>b</sup>	1.6237 $\pm$ 0.0874 <sup>c</sup>	
	Liming	1.5643	1.1704 $\pm$ 0.1062 <sup>ab</sup>	1.0692 $\pm$ 0.1625 <sup>ab</sup>	1.1061 $\pm$ 0.0799 <sup>a</sup>	
	Remediation	1.5643	1.0099 $\pm$ 0.1239 <sup>a</sup>	0.5868 $\pm$ 0.2458 <sup>a</sup>	1.2689 $\pm$ 0.0418 <sup>b</sup>	
S <sub>pos</sub> (%)	Without phosphorus	1.5643	1.1796 $\pm$ 0.0794 <sup>a</sup>	1.1075 $\pm$ 0.1717 <sup>a</sup>	1.2535 $\pm$ 0.0898 <sup>a</sup>	
	With phosphorus	1.5643	1.2211 $\pm$ 0.1220 <sup>a</sup>	0.8667 $\pm$ 0.1721 <sup>a</sup>	1.4123 $\pm$ 0.0928 <sup>b</sup>	

Figures followed by the same superscript in the same column and treatment factor show no significant effect using Duncan Multiple Comparisons Test at 95% level of significance

Table 2. The TPA, pyrite, Fe and Al (mean ± SE) of soil at different techniques to improve acid sulfate soil and phosphorus fertilizer in different times

Variables	Treatments	Before treated	Time			
			0 week	2 weeks	4 weeks	6 weeks
TPA (mol H <sup>+</sup> /tonne)	Without improving	376.2	498.8±15.4 <sup>c</sup>	432.6±28.6 <sup>c</sup>	417.1±14.0 <sup>b</sup>	
	Liming	376.2	23.5±9.4 <sup>a</sup>	55.9±38.0 <sup>a</sup>	169.8±57.7 <sup>a</sup>	
	Remediation	376.2	144.8±20.2 <sup>b</sup>	213.9±8.1 <sup>b</sup>	220.9±6.4 <sup>a</sup>	
	Without phosphorus	376.2	216.0±71.4 <sup>a</sup>	212.2±55.6 <sup>a</sup>	253.0±46.1 <sup>a</sup>	
	With phosphorus	376.2	199.9±46.0 <sup>a</sup>	228.7±73.2 <sup>a</sup>	285.5±46.2 <sup>a</sup>	
	Without improving	1.8764	1.4896±0.1156 <sup>c</sup>	1.9264±0.1271 <sup>c</sup>	1.8571±0.0621 <sup>b</sup>	
Pyrite (%)	Liming	1.8764	0.2452±0.0454 <sup>a</sup>	0.1049±0.0422 <sup>a</sup>	0.7582±0.2578 <sup>ab</sup>	
	Remediation	1.8764	0.8441±0.0621 <sup>b</sup>	0.6395±0.0906 <sup>b</sup>	0.9550±0.2057 <sup>a</sup>	
	Without phosphorus	1.8764	0.8266±0.1734 <sup>a</sup>	0.9593±0.3179 <sup>a</sup>	1.1262±0.2057 <sup>a</sup>	
	With phosphorus	1.8764	0.8926±0.2054 <sup>a</sup>	1.0161±0.3258 <sup>a</sup>	1.2540±0.2091 <sup>a</sup>	
	Without improving	8.674	8.527±493 <sup>c</sup>	5.467±803 <sup>b</sup>	8.133±248 <sup>a</sup>	
	Liming	8.674	1.474±428 <sup>a</sup>	1.222±565 <sup>a</sup>	5.632±971 <sup>b</sup>	
Fe (mg/L)	Remediation	8.674	4.093±328 <sup>b</sup>	3.483±183 <sup>ab</sup>	6.049±291 <sup>b</sup>	
	Without phosphorus	8.674	4.266±997 <sup>a</sup>	4.511±1.284 <sup>a</sup>	6.682±686 <sup>a</sup>	
	With phosphorus	8.674	5.130±1.141 <sup>a</sup>	4.662±933 <sup>a</sup>	6.528±530 <sup>a</sup>	
	Without improving	354	320±37 <sup>b</sup>	442±24 <sup>b</sup>	323±15 <sup>c</sup>	
	Liming	354	170±23 <sup>a</sup>	139±45 <sup>a</sup>	164±25 <sup>a</sup>	
	Remediation	354	187±8 <sup>a</sup>	382±6 <sup>b</sup>	227±6 <sup>b</sup>	
Al (mg/L)	Without phosphorus	354	249±35 <sup>a</sup>	332±50 <sup>a</sup>	210±27 <sup>a</sup>	
	With phosphorus	354	202±29 <sup>a</sup>	310±53 <sup>a</sup>	266±23 <sup>b</sup>	

Figures followed by the same superscript in the same column and treatment factor show no significant effect using Duncan Multiple Comparisons Test at 95% level of significance

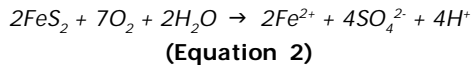
Table 3. The phosphorus (mean  $\pm$  SE) of soil at different techniques to improve acid sulfate soil and phosphorus fertilizer in different times

Variables	Treatments	Before treated	Time		
			0 week	3 weeks	6 weeks
Total P (mg/L)	Without improving	198.00	261.82 $\pm$ 19.13 <sup>a</sup>	293.78 $\pm$ 43.62 <sup>a</sup>	255.02 $\pm$ 15.21 <sup>a</sup>
	Liming	198.00	270.67 $\pm$ 22.14 <sup>a</sup>	306.07 $\pm$ 35.41 <sup>a</sup>	294.63 $\pm$ 25.99 <sup>a</sup>
	Remediation	198.00	267.48 $\pm$ 28.24 <sup>a</sup>	263.43 $\pm$ 20.30 <sup>a</sup>	242.97 $\pm$ 5.85 <sup>a</sup>
	Without phosphorus	198.00	232.00 $\pm$ 7.25 <sup>a</sup>	223.57 $\pm$ 11.36 <sup>a</sup>	240.30 $\pm$ 10.31 <sup>a</sup>
Retention of P (%)	With phosphorus	198.00	301.31 $\pm$ 18.43 <sup>b</sup>	351.96 $\pm$ 20.19 <sup>b</sup>	287.11 $\pm$ 16.62 <sup>b</sup>
	Without improving	47.00	46.33 $\pm$ 0.72 <sup>ab</sup>	48.33 $\pm$ 0.80 <sup>b</sup>	49.00 $\pm$ 1.37 <sup>b</sup>
	Liming	47.00	47.67 $\pm$ 0.84 <sup>b</sup>	49.33 $\pm$ 0.33 <sup>b</sup>	44.83 $\pm$ 0.48 <sup>ab</sup>
	Remediation	47.00	45.33 $\pm$ 0.72 <sup>a</sup>	44.83 $\pm$ 1.47 <sup>a</sup>	43.33 $\pm$ 2.11 <sup>a</sup>
Available P (mg/L)	Without phosphorus	47.00	47.56 $\pm$ 0.63 <sup>b</sup>	49.00 $\pm$ 0.3179 <sup>b</sup>	46.22 $\pm$ 0.62 <sup>a</sup>
	With phosphorus	47.00	45.33 $\pm$ 0.50 <sup>a</sup>	46.00 $\pm$ 0.3258 <sup>a</sup>	45.22 $\pm$ 1.93 <sup>a</sup>
	Without improving	4.50	7.98 $\pm$ 1.75 <sup>a</sup>	3.61 $\pm$ 0.59 <sup>a</sup>	3.83 $\pm$ 0.66 <sup>a</sup>
	Liming	4.50	13.60 $\pm$ 0.84 <sup>a</sup>	6.78 $\pm$ 1.02 <sup>b</sup>	5.16 $\pm$ 0.84 <sup>ab</sup>
Remediation	Without phosphorus	4.50	25.42 $\pm$ 4.29 <sup>b</sup>	6.88 $\pm$ 0.83 <sup>b</sup>	6.22 $\pm$ 0.86 <sup>b</sup>
	With phosphorus	4.50	13.16 $\pm$ 2.48 <sup>a</sup>	4.08 $\pm$ 0.50 <sup>a</sup>	3.67 $\pm$ 0.45 <sup>a</sup>
	With phosphorus	4.50	18.17 $\pm$ 3.84 <sup>b</sup>	7.43 $\pm$ 0.69 <sup>b</sup>	6.47 $\pm$ 0.58 <sup>b</sup>

Figures followed by the same superscript in the same column and treatment factor show no significant effect using Duncan Multiple Comparisons Test at 95% level of significance

a pH of 5.5 is considered the threshold for Al solubility before most Al begins to polymerise and precipitate. Sammut (1998) showed that Al was nonetheless still toxic at around pH 5.5 due to the polymerization of Al which cause severe gill damage in aquatic organisms.

For the remediation of ASS, soil drying triggered pyrite oxidation reaction as follows:



The elements and compounds resulting from pyrite oxidation became suspended in the water when the ASS was flooded and subsequently were flushed away with the waste water. The flushing away of this acidic material led to an increase in soil pH in the soil remediation treatment. From Table 1, it can be seen that the increases in the soil pH was better in the liming rather than the remediation treatment. Liming of ASS based on soil  $S_{\text{POS}}$  enabled a more quantitative approach to raising soil pH although under field conditions not all of the available pyrite will oxidise. The effect of the liming and remediation on ASS was relatively stable until six weeks after the treatments were finished.

The addition of TSP did not show a significant effect on the pH for the ASS. TSP is largely a monocalcium phosphate that is manufactured by adding phosphoric acid to rock phosphate (Tisdale and Nelson, 1975). Phosphoric acid contains up to 2% sulfur, which is neutralized by the inherent neutralising capacity of compounds contained in the TSP, and therefore did not cause a decrease in soil pH.

The soil improvement, both through liming and through remediation caused a significant decrease in the elements or compounds that cause soil acidity as analysed in this experiment i.e.  $S_{\text{POS}}$ , TPA, Fe and Al. The Ca contained in lime can replace Al in the soil absorption complex and the Al precipitates as  $\text{Al}(\text{OH})_3$ . The decrease of Fe and Al in the remediated ASS can be attributed to the removal of Fe and Al from the soil in the leachate through the exchange process as well as precipitation when increases in pH above 4 and 5.5 respectively. Pyrite concentration was reduced through the oxidation process as seen in the Equation 2.

Total-P refers to the sum of available and citrate-soluble phosphorus representing the

total amount present (Tisdale and Nelson, 1975). The improvement of ASS through liming and remediation did not show a significant effect on soil total-P. Phosphorus fertilization with a dosage of TSP 75 kg/ha was able to significantly increase the total-P of ASS. This was not surprising because the TSP fertilizer contained 19% P or 44%  $\text{P}_2\text{O}_5$ . The interaction between soil improvement and phosphorus fertilization also showed a significant effect on total-P in the third week after the application of the treatments was finished. The highest total-P was obtained for the treatment DOP1, but there is no significant difference with D2P1 (Table 4). The soil which was left under flooded conditions and the addition of TSP fertilizer caused the high total-P for treatment DOP1. Under flooded conditions no P was flushed away. On the contrary for the treatment involving remediation, P was most likely flushed away when the flooded water was discharged and replaced three times. Golez (1995) noted that remediation causes a reduction in available P because of the flushing process which can occur in ponds during water exchanges. This can present a problem in ASS because the soil can absorb most of the P and the remaining available-P can be lost from the system by flushing and not easily replenished by P released from the soil.

In general, the total-P rose when the aquaria were first filled with water (0 week) because of the TSP fertilizer application. However, total-P in soil decreased at the end of the experiment (6 weeks) due to the release of some P to the overlying water and its consumption by organisms.

Retention of P refers to that component of the phosphorus which is loosely held by the soil and can generally be extracted with dilute acids (Tisdale and Nelson, 1975; Barůvka and Rechcigl, 2003). This form of phosphorus is considered to be largely available to plants. However Tan (1991) states that  $\text{PO}_4$  can be adsorbed and retained (fixed) in soil in an insoluble form that is not readily available for plants. The improvement of ASS shows a significant effect on decreasing the soil retention of P, especially in ASS which is remediated. The low soil retention of P can be explained by a commensurate decrease in Fe and Al concentration in the remediated soil. According to Tan (1991),  $\text{PO}_4$  can be absorbed on the surface of colloids because Fe, Al and Mn ions act as chemical bridges. When soil pH



Table 4. Total-P and retention of P (mean  $\pm$  SE) in different combinations of techniques to improve soil with phosphorus fertilizer in the third week in acid sulfate soils

Treatments combination	Total P (mg/L)	Retention of P (%)
D0P0	200 $\pm$ 14 <sup>a</sup>	49.33 $\pm$ 0.33 <sup>b</sup>
D0P1	387 $\pm$ 24 <sup>b</sup>	47.33 $\pm$ 1.45 <sup>b</sup>
D1P0	242 $\pm$ 29 <sup>a</sup>	49.67 $\pm$ 0.33 <sup>b</sup>
D1P1	284 $\pm$ 28 <sup>a</sup>	49.00 $\pm$ 0.58 <sup>b</sup>
D2P0	228 $\pm$ 8 <sup>a</sup>	48.00 $\pm$ 0.58 <sup>b</sup>
D2P1	384 $\pm$ 10 <sup>b</sup>	41.67 $\pm$ 0.67 <sup>a</sup>

D0 : Without improving; D1 : Liming; D2 : Remediation;

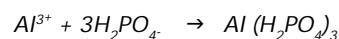
P0 : Without phosphorus; P1 : With phosphorus

Figures followed by the same superscript show no significant effect using the Duncan Multiple Comparisons Test at 95% level of significance

decreases, the subsequent increase in dissolved Fe and Al fixes more of the available P (Tisdale and Nelson, 1975). This process, although common in severely acidified ASS, also occurs in yellow reddish soil, brown reddish soil up to red soils (Oxisol, Ultisol, Lateritic) (Prasetyo *et al.*, 2001a). In acidic grassland soils, particularly peat and peaty gleys, organic matter with Fe and Al also rapidly fix P by organic matter and combinations of organic matter with Fe and Al, also (Pizer, 1965). It was a different matter with the liming of ASS, although the Fe and Al content was relatively similar to remediated soil. The retention of P was higher in the liming treatment compared to the remediation treatment. This is probably the result of PO<sub>4</sub> which was not only absorbed on the surface of colloids with Fe and Al ions, but also co-precipitated with Ca (Tan, 1991). Prasetyo *et al.* (2001a) who showed that the P applied to soil in the pH range from 4.4 to 6.5 has minor influence on Ca-phosphate content of soil, but can increase the sesquioxide-bound phosphate content. Table 1 shows that liming of ASS raised the soil pH<sub>KCl</sub> to greater than 6.5 which potentially increased the formation of Ca-phosphate. At a soil pH of 4.1, the Ca-phosphate concentration is only 60 mg/L, and the increase in the the soil pH to 7.5 also raises the Ca-phosphate to 108 mg/L (Prasetyo *et al.*, 2001a). The formation of Ca-phosphate compounds is partly controlled by the concentration of Mg (Martens and Harris, 1970), carbonate ions (Stumm and Morgan, 1981), organic acids (Sinha, 1971), humic

substances (Inskeep and Silvertooth, 1988), redox potential (Moore and Reddy, 1994) and pH (Olila and Reddy, 1993).

For the treatment without soil improvement, there was an increase in retention of P. The highest retention of P for the treatment without soil improvement occurred in the sixth week. This is consistent with the findings of Tan (1991) who showed that the Al-phosphate precipitation is increasingly less available for plants over time. The large Al-phosphate concentration encountered in the treatment without soil improvement was due to the high concentration of soil Al. Phosphate can also react with metal ions like Fe, Al and Mn through the following reaction (Tan, 1991):



(Equation 3)

Longer time periods of contact between soil and phosphorus fertilizer increases the soil retention of P. Tisdale and Nelson (1975) relate this reaction to the subsequent alteration of the fixation product such as dehydration and crystal reorientation. This finding suggests that contact of fertiliser with the soil in the pond preparation stage will affect the release of P into the water column. Additionally delaying fertiliser application after liming will help reduce precipitation of P in the presence of the high initial Ca concentration.

Soil improvement showed a significant effect on soil available-P. In general, a high

available-P was measured for remediated soil treatment. Soil pH was a primary factor in the control of available-P because the soil pH for the remediation treatment ranged from 6.11 to 6.19 and was in the soil pH range where the maximum available-P occurs. In peat soil, Mustafa (1996) obtained a maximum available-P for a pH range from 6.06 to 6.19. In most soils the available-P is maximal in the pH range 5.5 to 7.0, (Tisdale and Nelson, 1975; Brady, 1974). The correlation between available-P and the soil reaction at a pH range from 6.0 to 7.0 needs to be considered in aquaculture management to improve fertilizer efficiency.

A low available-P was found in ASS which were not improved. This is caused by the high concentration of Fe and Al in this treatment. The low available-P in ASS is due to the rapid fixation of P with Fe and Al (Singh, 1982a) and with Mn if its concentrations are high in pond soil (Poernomo, 1983). Golez and Kyuma (1997) said also demonstrated that low soil pH increased Fe and Al but also decreased available-P caused by precipitation of phosphate ions by the elevated metals.

Increasing the pH of an acidic pond bottom soil will increase the soil available-P added in fertilizers (Boyd and Massaut, 1999). After the application of all treatments and at the time of the water filling (0 week), it was clear that the available-P as a percentage of total-P was about 3.05% in unimproved soil, 5.02% in liming in soil and 9.50% in remediation of ASS. These results showed that 96.95, 94.98, and 90.50% of total-P in the soil was in various fixed forms

for the treatment without soil improvement, the treatment involving improvement through liming, and the treatment involving improvement through remediation, respectively. These results indicate that for effective use of P in ASS, soil acidity must be remediated well before fertilizer application. Liming can also help the build-up of available-P can cause P fixation with Ca if pH increases above 7.

Soil available-P was found to be highly and significantly correlated with soil  $pH_{KCl}$  and soil Fe, and significantly correlated with soil  $pH_{FOX}$  and soil Al, but weakly correlated with soil retention of P, soil total-P, soil  $pH_F$ , soil  $S_{POS}$ , soil TPA and soil pyrite (Table 5). The release of Fe and Al following oxidation was offset by a loss of these metals through flushing. As the amount of Fe and Al decreased, it showed a corresponding increase in the amount of available-P.

The relationship between available-P and soil pH ( $pH_{FOX}$ ,  $pH_{KCl}$ ) can be seen in Figure 1 and for available-P and soil Fe and Al in Figure 2. The increase in soil pH caused a significant increase in available-P in ASS. Mokawunye (1975), showed that raising soil pH will decrease soil positive charges which decreases soil absorption capability decreasing releases P.

There was a close relationship between available P and  $pH_{FOX}$  and  $pH_{KCl}$ . The available-P possibly had a close relationship with the actual acidity of soil. To measure  $pH_{FOX}$ , 30% hydrogen peroxide was added to the soil and

Table 5. Correlation coefficient (*r*) and probability (*P*) among available-P and other soil variables of acid sulfate soils with different techniques to improve soil and different phosphorus fertilizing

Variables	<i>r</i>	<i>P</i>
Retention of P	-0.149	0.385
Total-P	-0.126	0.463
$pH_F$	0.134	0.436
$pH_{FOX}$	0.344	0.04
$pH_{KCl}$	0.468	0.004
$S_{POS}$	-0.278	0.09
TPA	-0.305	0.07
Pyrite	-0.303	0.073
Fe	-0.479	0.007
Al	-0.343	0.04

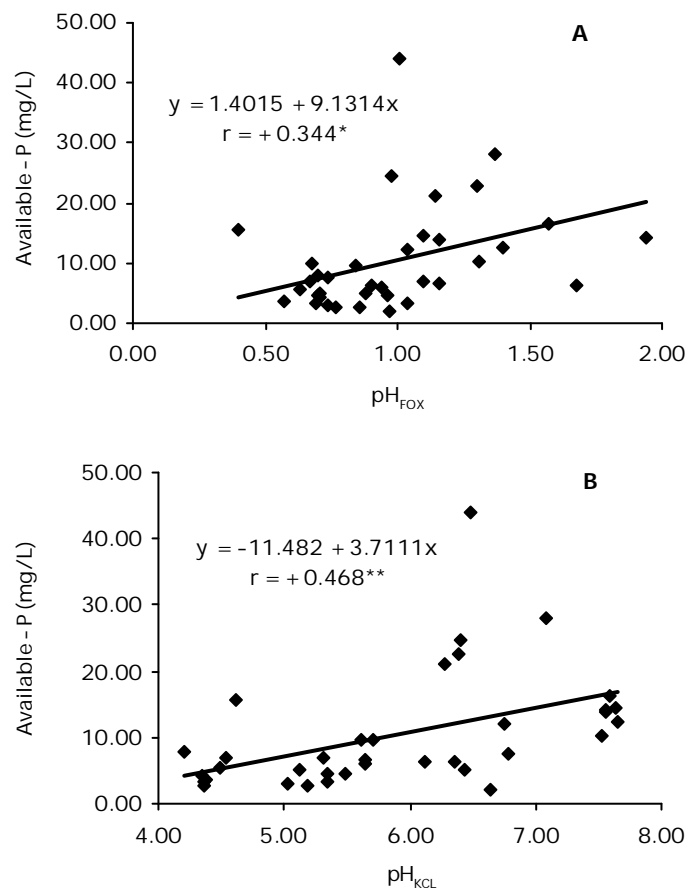


Figure 1. Relationship between soil available-P and pH<sub>FOX</sub> (a) and pH<sub>KCL</sub> (b) in acid sulfate soils

forced oxidation. To determine the pH<sub>KCL</sub> of the soil, samples were dried beforehand and then KCl 1 M was added which was considered to be indifferent as an electrolyte.

Available-P in the soil had very significant negative correlation ( $P = 0.007$ ) with the soil Fe and a significant negative correlation ( $P = 0.040$ ) with soil Al (Figure 2). This showed that available-P would increase very significantly with a decrease in the Fe concentration, and available-P would increase significantly with the decrease in soil Al. The stronger relationship of Fe compared to Al with soil available-P was assumed to be caused by the pH of the soil which was not too acidic (pH around 5.0), with a result that the reactivity of Al was not too strong. Al-Jabri *et al.* (1987) reported that the transformation of Fe strongly influences the retention of P and available-P.

### Klekap Production

Soil quality is a major control on the growth of *klekap*. The *klekap* production during the experiment is shown in Figure 3. The different techniques for soil improvement and phosphorus fertilizing showed a significant effect on *klekap* production during the experiment. The interaction of different techniques for soil improvement and phosphorus fertilizing showed only a significant effect on *klekap* production at the end of the experiment (the sixth week) as shown in the Table 6.

The highest *klekap* production during the research was obtained in the soil remediation treatment (concerning the soil improvement factor), and this was followed by the soil improvement through liming and the non-

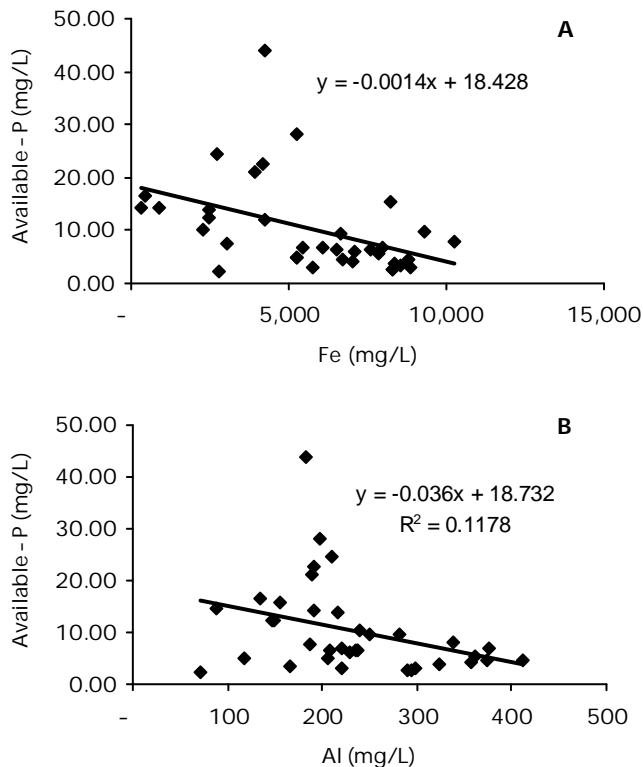
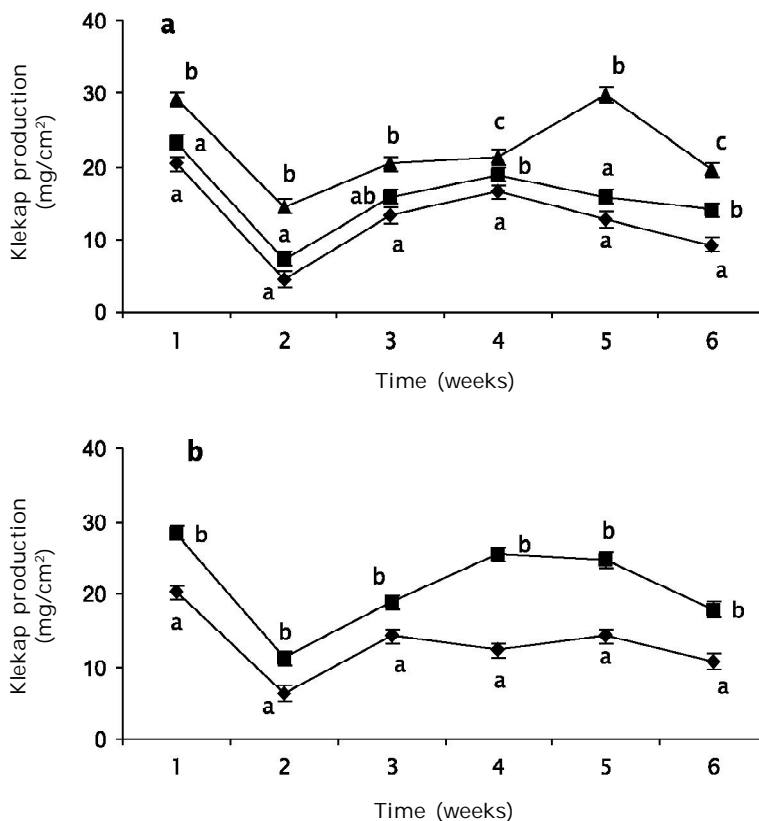


Figure 2. Relationship between soil available-P and iron (a) and aluminium (b) in acid sulfate soils

improvement of the soil. The higher *klekap* production for the soil improvement through remediation and liming created a more suitable growing environment than in the treatment without soil improvement. The higher soil pH and available P in the improved soils were considered more favourable for *Klekap*. Additionally, Fe and Al were lower for the improved soils. For good *klekap* growth a soil pH between 7.0 and 8.0 is required (Ballesteros and Mendoza, 1976; Davide, 1976). Padlan (1976) showed that a soil pH of 6.8 up to 7.5 enhances *klekap* growth because soil nutrient availability and microbial activities are maximal in this pH range. The higher *klekap* production for the treatment involving phosphorus fertilizing compared to the treatment without phosphorus fertilizing was due to the higher concentration of available-P in the treatment involving P fertilizing. Olsen and Dean (1965) classed available-P in soil into four categories, that is: very low when smaller than 3 mg/L; low when it is between 3 and 7 mg/L; medium when it is between 7 and 20 mg/L and high when it

is greater than 20 mg/L. The available-P concentration for the treatment without phosphorus fertilizing was classed as average at the beginning of the research and then was classed as low, whereas, for the treatment involving phosphorus fertilizing, it was classed as medium during the experiment.  $PO_4$  is present in algae cells in the form of organic P (such as ribonucleic acid (RNA), deoxyribonucleic acid (DNA), fat) and inorganic-P (Nalewajko and Lean, 1980). In the presence of sunlight, DNA and inorganic-P is converted into  $PO_4$  bonds such as adenosine triphosphate (ATP). The ATP is important to synthesis amino acids and proteins.  $PO_4$  compounds such as ATP, adenosine diphosphate (ADP) and nicotinamida adenine dinucleotide phosphate (NADP) critical in the photosynthesis process and metabolism of amino acids (Tisdale and Nelson, 1975).

Figure 2 shows that the *klekap* production fluctuates from time to time in accordance with the environmental conditions. In general, the highest *klekap* production was obtained in the



The lines followed by the same letter at the same time show a non-significant effect using Duncan Multiple Comparisons Test at 95% level of significance

Figure 3. The *klekap* production (mean± SE) for different techniques to improve soil (a) and for the application of phosphorus fertilizer (b) in acid sulfate soils

first week. This is assumed to be the result of the availability of nutrients, especially P which is relatively high, along with the low amount of acidic material, such as Fe and Al in the first week.

In the second week, the *klekap* production decreased for all treatments. This was assumed to be because the algae forming the *klekap*, which had already experienced rapid development in the first week, then decayed. In a pond situation, consumption of *klekap* would occur and the pond environment, therefore, could produce different results than in a laboratory experiment. In addition, the *klekap* in all containers was raised up and floated as a result of the higher water temperature in the afternoon. The floating *klekap* covered a part of the surface of the container and this hindered the penetration of

sunlight thereby hindering photosynthesis and slowing *klekap* growth. The sunlight greatly influenced the *klekap* such that the *klekap* would grow well if the sunlight penetrated through to the bottom soil. This effect would need to be considered in pond management so that sunlight is not attenuated.

From the third to the fifth week, *klekap* production increased for all treatments except the treatment involving no soil improvement. The increase in *klekap* production was assumed to be due to the available nutrients which were still sufficient to support *klekap* growth. After the fifth week, *klekap* production experienced a decline. This was assumed to be because the availability of nutrient released by the soil had more and more decreased because it was used by the algae. According to Fogg *et al.* (1973) there nutrient content

Table 6. *Klekap* production (mean±SE) in different combinations of techniques to improve soil with phosphorus fertilizer in the sixth week in acid sulfate soils

Treatments combination	<i>Klekap</i> production (mg/cm <sup>2</sup> )
DOPO	7.88±0.76 <sup>a</sup>
D1PO	8.32±0.44 <sup>a</sup>
DOP1	10.51±0.76 <sup>a</sup>
D2PO	15.77±0.76 <sup>b</sup>
D1P1	19.71±2.01 <sup>c</sup>
D2P1	23.21±1.58 <sup>cd</sup>

DO : Without improving; D1 : Liming; D2 : Remediation;  
PO : Without phosphorus; P1 : With phosphorus

Figures followed by the same superscript show no significant effect using the Duncan Multiple Comparisons Test at 95% level of significance

which more and more decreased from time to time constituted another cause for decrease in the growth of the algae that formed the *klekap*.

The interaction between soil improvement and phosphorus fertilizing gave a significant effect for *klekap* production in the sixth week (Table 6). From the Table 6 it can be seen that the highest *klekap* production, that is 23.21 mg/cm<sup>2</sup> was obtained for treatment D2P1 that is soil improvement by remediation accompanied with phosphorus fertilizing amount TSP 75 kg/ha. However this was not significantly different to the D1P1 treatment, that is soil improvement by liming accompanied with P fertilizing amount TSP 75 kg/ha. Soil improvement in ASS can create better conditions such that available-P is increased and results in higher *klekap* production.

## CONCLUSIONS

The liming and remediation treatments had the same effect on soil quality, that is, the liming and remediation were able to increase the pH (pH<sub>F</sub>, pH<sub>FOX</sub>, pH<sub>KCl</sub>) and decrease the values of soil S<sub>POS</sub> and TPA and concentrations of Fe and Al. The liming and remediation of ASS decreased retention of P and increased available-P of soil. However, remediation had a more significant effect on decreasing the retention of P and increasing available-P compared to liming most probably because Fe and Al were flushed away. Remediation by oxidation, flooding and flushing of the soil in general resulted in a higher level of *klekap*

production compared to soil improvement by liming as well as no soil improvement. The phosphorus fertilizing using a dosage of 75 kg/ha caused a higher production of *klekap* compared to the treatment involving no phosphorus fertilizing. The interaction between the different techniques of soil improvement and phosphorus fertilizing showed significant effects on *klekap* production such that the highest *klekap* production, that is 23.21 mg/cm<sup>2</sup>, was obtained for soil remediation and phosphorus fertilizing with a TSP dosage of 75 kg/ha.

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