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Water spinach (*Ipomoea aquatica*) as potential macrophytes to remediate acid mine drainage (AMD)

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Abstract

One of the serious environmental problems in mining industry is Acid Mine Drainage (AMD) after mineral sulfides are exposed for oxidation-reduction. Acid water from AMD has devastating effect on aquatic natural resources and caused toxicity in plants, but here macrophytes were tested for its capability of increasing water pH. In an experiment following Completely Randomized Design involving five treatments replicated four times: T_0 – *Typha latifolia* (Cattail), T_1 – *Centella asiatica (*Gotu kola), T_2 - *Ipomoea aquatica* (Water spinach), T_3- *Eichhornia crassipes* (Water hyacinth), and T_4- *Bacopa monnieri* (Water hyssop), it revealed that *I. aquatica* significantly increased the pH by 81%. It is as effective as *T. latifolia* (Cattail) – the control, in increasing pH of AMD after Analysis of Variance and Duncan Multiple Range Test (DMRT). A macrophyte *Ipomoea aquatica* (water spinach) therefore has the capacity to neutralize acidity in an Acid Mine Drainage (AMD).

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Introduction

Acid Mine Drainage (AMD) is a metalliferous water draining in open surface characterized by elevated acidity (Jennings *et al.*, 2008; Abbasi *et al.,* 2009). This is caused by the oxidation-reduction of exposed mineral sulphides (Ravengai *et al.,* 2004; Abbassi *et al.*, 2009) mostly in mining areas – in the tailings, ponds and abandoned mines. Examples of common mineral sulphides are the pyrite, chalcopyrite, pyrrhotite, sphalerite, covelite, millerite and galena (Abbassi *et al.,* 2009).These mineral sulphides upon exposure to air and water undergo a series of chemical reactions producing acidic protons – the hydrogen. The low pH of the discharge mine water results to the dissolution of minerals, thus create an acidic environment which is harmful to the vegetation, aquatic life and wild life (Fromm, 1980; Abreu and Lavorante, 2003; Kumari *et al.,* 2010; Ochieng *et al.* 2010; Martinez *et al.*, 2013; Dowarah *et al.;* Martinez *et al.*, 2013; Choudhurry *et al.*, 2017;), necessitating immediate solutions, but current solutions are expensive.

There are different treatments at which mining operators can implement to neutralized acid water. Blowes *et al.* (2014) enumerated a number of approaches both active and passive to solve the problem, and one of the Passive solutions is the use of plants. The potential of indigenous South African grasses, *Hyparrhenia hirta* and *Setaria sphacelatab* was tested by Ramla and Sheridan (2015). The work of Javed (2011) focuses on the mechanisms by which *Elodea canadensis* and *Eriophorum angustifolium* change the surrounding water pH in the presence of toxic ions. It was found out in the study that *Elodea canadensis* shoots increased the pH of the surrounding water in the presence of free Cd ions while *Eriophorum angustifolium* roots increased the surrounding water pH, and this effect was enhanced in the presence of arsenic and metals. Currently, there is paucity of information as to what plant species capable of remediating acid mine drainage.

In the current study, we investigated five aquatic macrophytes whether they have the ability to increase pH of water in an Acid Mine Drainage. Specifically, we aimed to determined which macrophytes species significantly increase pH from initial value of 4.0, and how much improvement in pH was made over fifteen days of experiment.

Materials and Methods

The study was an experiment following the Completely Randomized Design (CRD). There were five (5) treatments including the control, each was replicated four (4) times: Treatments are as follows: To – *Typha latifolia* (control), T_1 – *Centella asiatica*, T_2–*Ipomoea aquatica*, T_3–*Eichhornia crassipes* and T_4 – *Bacopa monnieri.*

These aquatic plant species were collected because they are common weeds in the lagoon with dirty water from nearby residential areas, road networks, and commercial areas; multiplying rapidly in the City of Cagayan de Oro, Philippines. The collected plants were washed and cleaned thoroughly with tap water and rinsed with distilled water to remove dirt, soil and extraneous materials. Cuttings were then prepared with up to eight inches long.There were 12 cuttings for each plant treatment for the four replications with three samples each. A total of sixty cuttings were then prepared, and then acclimatized for one month under Peter's nutrient solutions.

Acclimatized cuttings were then transferred to sixty 1Liter bottle containing 600 mL Peter's nutrient solution (a general purpose fertilizer). The pH of this solution was adjusted initially to 4.0 using either a stock solution of hydrochloric acid (1.0 g/molHCl) or sodium hydroxide solution (1M NaOH) to create an artificial Acid Mine Drainage condition. This acid nutrient solution was changed every week to replenish the lost nutrient. In each occasion, the pH of artificial AMD was recorded prior to changing the nutrient solution, and then brought back to the last recorded pH after replacement of nutrient solution. Data collection was done twice weekly for fifteen days. F-test was implemented to analyzed the data at 5% level of significance, coupled with Duncan

Multiple Range Test (α=0.05) using Paleontological Statistics (PAST) version 3.0.

Results and discussion

Results revealed that from initial pH (4.0) of the artificial acid mine drainage condition, all plants tested in the study *viz. Typha latifolia, Ipomoea aquatica*, *Centelia aquatica, Eichornia crassipes* and *Bacopa monieri* were able to increase the pH up to 7.7 (Fig. 2) after fifteen days of growing similar to *Elodea Canadensis* (Nyquist and Greger, 2009).This capacity to increase pH however varies significantly among each other as shown in Table 1 after F-test. Duncan Multiple Range Test (DMRT) in Table 2 shows that *Ipomoea aquatic* is as effective as *Typha latifolia* in increasing pH of the initially acidic water (pH 4.0). Although *Bacopa monieri* obtained the lowest pH value among other plants, but its pH of 6.25 is twenty two times higher than the initial 4.0.

Table 1. Results of the Analysis of Variance among five treatment plant species at 5% level of significance.

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	6.046644	4	1.511661	4.74653114667621 **	0.007410289	2.8660814
Within Groups	6.36954	20	0.318477			
Total	12.416184	24				

**Highly significant.

Typha latifolia (Cattail) exhibits the highest pH value of 7.70; this is 92.5% increase from initial pH of 4.0, and then followed by *I. aquatic* which increased by 81%. *E. crassipes, C. asiatica* and *B. monnieri* obtained 6.81 (70.25%), 6.72 (68%) and 6.25 (56.25%) pH, respectively. Cattail which served as control is known as an acid tolerant and can thrive under a variety of environmental conditions (Kumar *et al.,* 2008; Udayabhanu and Prasad, 2010).

Table 2. Results of Duncan Mutiple Range Test for the five plant treatments at 5% level of significance.

Treatment	Mean of the Treatment	DMRT (B)
Typhy latifolia	7.704	A
Ipomea aquatica	7.238	ab
Eichhornia crassipe	6.814	bc
Centella asiatica	6.722	bc
Bacopa monnieri	6.252	c

The case of *I. aquatica* is consistent with that of Herniwante *et al.* (2013) where they found a 53% increase in pH. However, such study did not account the buffering capacity of the soil it added into the passive treatment model and the evapotranspiration in the experiment for 29 days. The current study is much more controlled where it eliminates these confounding factors, and obtained and improvement of results.

Stoltz and Greger (2002) found similar results in which pH of acid water from mine tailings (pH 2.6-3.2) was increased by 69% and 100% by Tall Cottongrass (*Eriophorum angustifolium* Honckeny) and White Cottongrass (*Eriophorum scheuchzeri* Hoppe) respectively after one month.

Tall Cottongrass was tested again in the work of Javed *et al.* (2013) corroborating results of Stoltz and Greger (2002) in its capacity to increase pH similar to the current study.

Rice plant (*Oryza sativa* L.) was also found to elevate pH, however under cadmium stressed environment (Zeng *et al.,* 2008). Under environmental stresses, plant metabolites and a number of unknown substances are released as roots exudates with consequent increase in pH (Lopez-Bucio *et al.*, 2000).

Fig. 1. Photos of macrophytes collected and tested in nutrient solution at initial pH of 4.0. To–*Typha latifolia* (control), T_1–*Centella asiatica*, T_2–*Ipomoea aquatica*, T_3–*Eichhornia crassipes* and T_4–*Bacopa monnieri.*

These metabolites are organic acids with charged ends that attract protons, thereby acting as chelating compounds in the surrounding medium (Henry, 2003). The number of protons attracted depends on the number of carboxylic groups (Javed *et al.*, 2013).

The work of Zeng *et al.* (2008) shows significant positive correlation between pH and organic acids. Oxalic acid, malic acid and citric acid were the organic acid exudates from roots which were suggested to play an important role in the variation of rhizosphere pH (Zeng *et al.*, 2008; Zhang *et al.*, 2010; Favas *et al.*, 2014). The same could be the case of macrophytes suspending in water because these organic acids are involved in processes such as plant metabolism, metal detoxification and nutrient solubilization (Jorge and Arruda, 1997).As such, these are constantly secreted by plant roots (Lopez-Bucio *et al.*, 2000); however, its quantity is genetically defined.

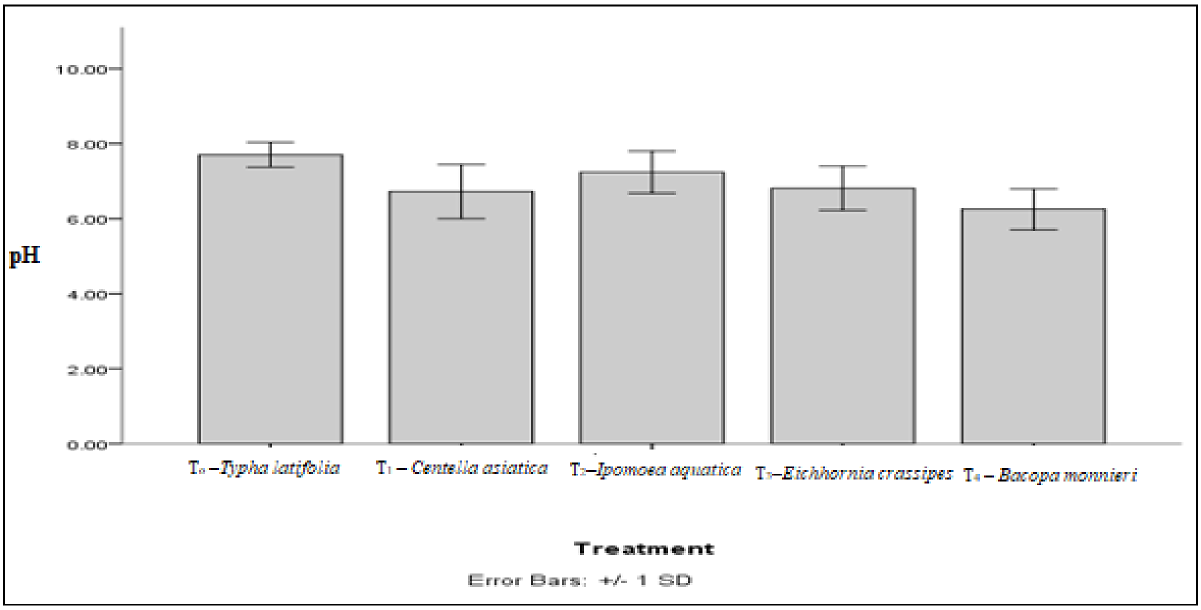

Fig. 2. Potential of the hydrogen after 15 days of growing of the five treatment plant species.

Although there was no stress introduced in the current study, e.g. from over concentrations of heavy metals, such as the case observed in rice (Zeng *et al.,* 2008), cottongrass (Stoltz and Greger, 2002; Javed *et al.* 2013), and *Elodea canadensis (Nyquist and Greger, 2009),* the observed increase in pH may have been due to releases of hydroxyls, bicarbonates and carbonates to maintain charge balance in the medium (Hinsinger *et al.*, 2003 and Javed *et al.* 2013).

From among the plant treatments, except the control, *I. aquatica* could be an alternative plant to *T. latifolia* in terms of remediating acid mine water. The ideal characteristic is the presence of enormous amount of root hairs which was the same as *T. latifolia*. Its highly intensive root system denotes more contact and more likelihood in nutrient uptake and absorption, and release of root exudates.

The *I. aquatica* and *T. latifolia* showed the same ability in improving pH of the artificial Acid Mine Drainage. This study suggests that *I. aquatica* could be a good plant in remediating AMD, since it can tolerate the acidic medium. Its roots were able to regrow and significantly increased the pH by 81% from its initial pH during the entire duration of the study.

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