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Diversity Assessment of Floral Species and Screening of Potential Nickel Hyperaccumulator in Nickel-Rich Kinalablaban Delta, Cagdianao, Claver, Surigao del Norte, Philippines

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Abstract
The study was conducted to determine the status of floral diversity and screen plant species that has the potential to accumulate nickel in the nickel-rich Kinalablaban Delta located in Claver, Surigao del Norte. Fifty-nine (59) species of plants belonging to thirty-seven (37) families were found. The most represented families were Poaceae, Moraceae, and Fabaceae. These were families generally encountered anywhere and can be found in areas undergoing ecological succession. Notable species observed were *Imperata cylindrica* and *Saccharum spontaneum* in which these species were widely distributed all throughout the delta. Rhizophora sp. was also found since they are intentionally planted by the adjacent mining firm as part their environmental protection and management plan. The diversity, dominance and evenness were low indicating a need to enhance the vegetation in the delta. Out of the 59 species identified, only four species tested positive as potential nickel hyperaccumulator including *Ardisia elliptica, Premna odorata, Phyllantus securinegoides* and *Phyllantus* sp. These plant species however, must be tested further if it is indeed a nickel hyperaccumulator.
Keywords: flora, ultramafic, hyperaccumulator, nickel mining

1. Introduction

Deltas when affected by the periodic presence of water are considered as wetlands. Wetlands are ecologically important ecosystems since it harbor diverse array of floral and faunal species. Plant nourishes the people with the necessities of life such as food, medicine, energy, and other products to enhance and improve the way of living. However, diversity in such ecosystems have declined due to anthropogenic threats and inappropriately utilizing natural resources by over harvesting, logging, burning and clearing wetlands (Rai and Raleng 2011).

Vegetation in a wetland is deemed important since it provides habitat and source of food to many animals including insects, bird, mammals, and several species of decomposers. It also acts in filtering pollutants, sediments, and silts that go downstream. Waterways with emergent vegetation were observed to have a significant effect on the transport of sediment (Leonard and Reed 2002). Sediment rates were also high because of direct trapping ability of the stems and leaves of emergent plants to sediments (Harvey *et al*. 2003). Aside from trapping silts and sediments, vegetation also takes up nutrients since it is a requirement for growth and development. Uptake of nutrients by plants is one management option that will remove excessive nutrients in a wetland. Plants with high rates of net primary productivity and higher nutrient uptake are preferred in wetlands subjected to wastewater inputs (Kansiime *et al*. 2007). However, a downside of the influx of sediments in estuaries is the accumulation of pollutants that adsorbed to sediment grains which gradually deteriorates the benthic environment and water quality (Choi *et al*. 2006). The presence of hyperaccumulator plants may address this problem. Hyperaccumulator plants have the means to take up and accumulate heavy metals in higher concentrations and store it in different parts of the plant. The concentration of heavy metals in the plant is sometimes higher than that found in the soil (Sagiroglu *et al*. 2006).

Kinalablaban Delta, located near a large scale mining firm in Cagdianao, Surigao del Norte, is home to several species of plants. The mined area in the upland is composed mainly of serpentinized ultramafic rocks, and a reddish iron oxide stain predominates these types of rocks (Pantilo 2003). Ultramafic outcrops holding nickel-rich laterites are prime nickel mining targets which are also in direct conflict with biodiversity (Van der Ent *et al*. 2013). Soils derived from ultramafic rocks are called as serpentine soil composed of Mg, Fe, Cu Co, Ni, and Cr elements due to weathering of olivine, pyroxene, and chromite minerals (Shah *et al*. 2010). Serpentine soil hosts a distinctive flora, shallow, well drained and prone to erosion with high concentrations of heavy metals which may account for the stunted growth of plants and patches of the vegetation in this type of soil (Brooks 1988). Hyperaccumulators growing in such soil has 10-20 times higher concentration of nickel compared to normal plants growing in the same nickel-rich environment (Homer *et al*. 1991). Due to the presence of vegetation in the entire delta, it had somehow prevented a more serious sedimentation and siltation that may occur in coastal shore. The diversity assessment in the area was conducted to determine the prevailing species of plants that were present. Since the study site may have been indirectly affected by mining activities, there is a higher chance that nickel deposition also occurs such that determining the potential hyperaccumulator plants was also conducted.

The result of this study was important since it can provide firsthand information to nearby large scale mining company of the status of floral diversity and the potential nickel hyperaccumulator plant species that can be used to rehabilitate and enhance the vegetation in the delta. Kinalablaban Delta has patchy vegetation with a lot of open spaces with fewer plants established. Enhancing floral composition in the area is very crucial and plants with a potential to hyperaccumulate nickel could be integrated as part of the environmental management plan of the mining company. Identification of nickel hyperaccumulators is very important since it can also be utilized in removing and cleaning the area with high concentration of nickel.

The general objective of this study was to determine the status of floral diversity in Kinalablaban delta in Cagdianao, Claver, Surigao del Norte. Specifically, this study aimed at determining the plant species found in the area, describing ecosystem through diversity, dominance, and evenness of plant species, and screening of plant leaves that has the potential to accumulate nickel using colorimetric method.

2. Methodology

2.1 Study Area

The study area was located in a tidal flat and a delta at the same time within the premise of a mining firm in Cagdianao, Claver Surigao del Norte (Figure 1). The delta is a catchment of sediments coming from three tributaries of Kinalablaban River which originates from the upland that is now affected by nickel and iron ore mining activities. The study of Apas and Cedron (2016) found high nickel concentration for the entire delta since the mean values ranges from 2281 mg/kg to 11090 mg/kg. Three transect lines were established perpendicular to the coastline of the delta. Establishment of 10 m x 10 m quadrats was employed in a systematic sampling strategy with 50 m interval from one quadrat to another. If after the 50 m interval and a quadrat falls in a water body, the next quadrat established was adjusted so that it was set in an area where there were plants established. Plants species within the quadrat were identified and listed. Plant samples such as leaves, flowers, and fruits (if any) were gathered for species that cannot be determined on site and pictures were also taken. Initial identification of plant species was done in the area with the help of a plant taxonomist and local guides. The plant samples were further identified by foresters and plant taxonomist in Caraga State University and relevant taxonomic books for identification of plants.

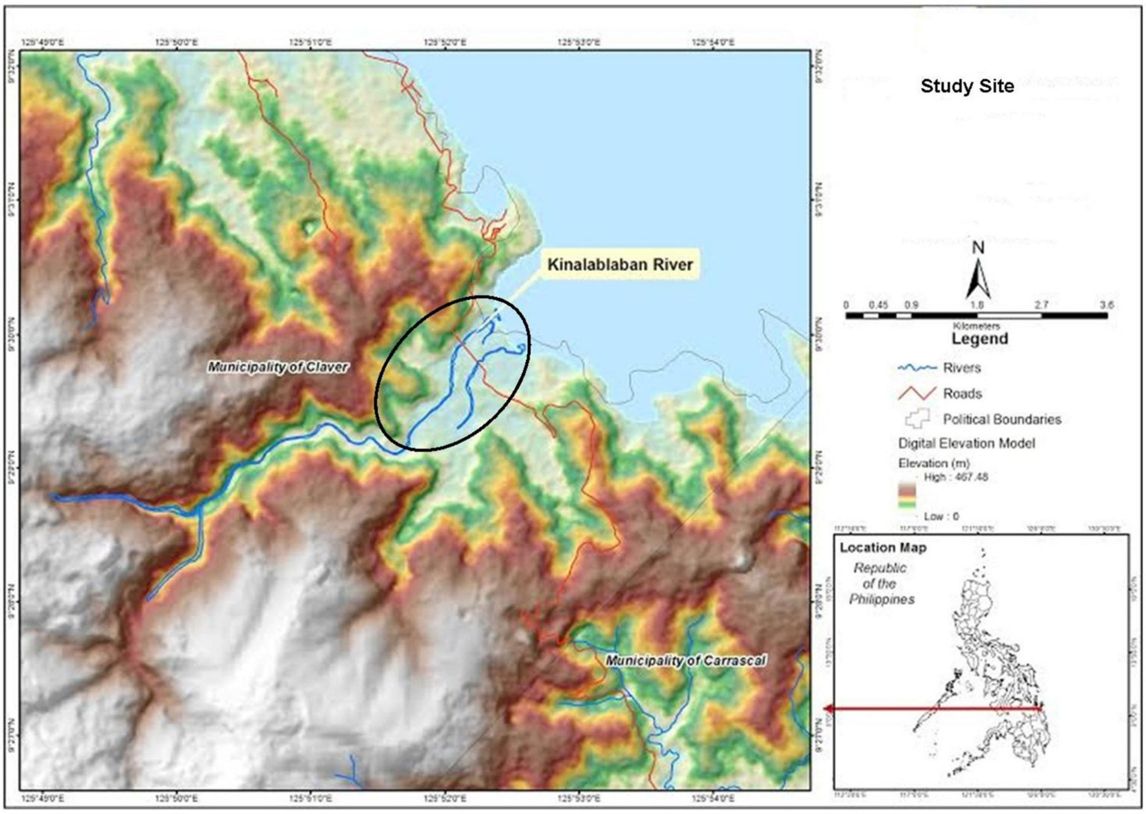

Figure 1. Map showing the study site

2.2 Data Analysis

The data obtained from the study was analyzed using the different indices such as diversity index, evenness index, and index of dominance.

Species diversity was computed using Shannon Wiener Diversity Index which determines the average uncertainty per species in an infinite community made up of species with known proportional abundance (Odum 1971).

$$H' = -\sum_{i=1}^{s} Pi \ln Pi$$

where,

H' = Shannon-Wiener Index
s = the number of species
pi = the proportion of individuals or the probability of the each species
ln = natural logarithm

Evenness refers to how species abundance is distributed among species and was calculated using the formula

$$J' = \frac{H'}{\log S}$$

Where

J' = Evenness Index
H' = Shannon-Wiener Index
S= Number of species

Index of dominance was determined using the formula

$$D = 1 - E$$

Where

D= Index of Dominance
E= Evenness Index

2.3 Detection of Nickel Concentration in Plant Tissue Using Semi Quantitative Screening

Filter papers (Whatman No. 41) were impregnated with nickel specific colorimetric 1% dimethylglyoxime. One gram of dimethylglyoxime (DMG) was dissolved in 70% Ethanol (EtOH) solution. Filter papers were cut with a diameter approximately two centimeters and were soaked in the prepared solution for five minutes, and air dried at room temperature for two days. All plants species found inside the plot were screened for nickel accumulation. Plant samples subjected to screening were brought to Science and Technology Laboratory in Caraga State University. The collected fresh leaves were crushed using mortar and pestle to take out the leaf extract. The extract was then placed and pressed against the impregnated filter paper. If there is a change in color on the filter paper, it indicates that the plant is a potential hyperaccumulator (Ahemad 2014). Positive results usually have an appearance of purple (Bayramoglu *et al*. 2012), rose pink color (Ghasemi *et al*. 2014), or red color (Roccotiello *et al*. 2015) upon contact with nickel.

3.Results And Discussion

3.1 Floral Diversity in Kinalablaban Delta

Fifty-nine (59) species of plants belonging to thirty-seven (37) families were found in the study site (Table 1). Poaceae, Moraceae, and Fabaceae were the most represented families. These families were encountered anywhere and are usually found in areas undergoing continuous ecological succession. Table 2 shows the diversity, dominance, and evenness for plants in the three (3) transects that were established in Kinalablaban Delta. Results indicated that transect 2 has slightly high diversity as compared with other transects. However, the values for all transect falls under a low diversity category. This further indicates that revegetation and rehabilitation in the delta should not only be focused on one species but should also include several species of plant as well. The evenness was low, which also supports the fact that the area was low in diversity. Dwarfed plants in areas with soils derived from ultramafic parent material resulted from a shortage of one or more major nutrients or from very high nickel concentration in soil (Brooks *et al.* 1995). This is because ultramafic soils have naturally high concentrations of metals and are often low in major plant nutrients which may have also accounted for the low diversity of vegetation in the area (Proctor 2003). Several ecological factors may also play a role in the diversity of a certain area. This could be affected by the presence of microorganisms like bacteria and some species of fungi (Brearley 1999). The absence of a fungus in association with plants roots somewhat contributes to the low diversity in areas with high concentrations of heavy metal such as nickel. The abundance of species endemic or preferential to ultramafic soil can be interpreted as a loss of ecological quality of the community (Leung *et al.* 2012).

Several species under Poaceae were observed in the site. Species such as *Imperata cylindrica* (L.) P. Beauv, *Paspalum conjugatum* P.J. Bergius, *Muhlenbergia lindheimeri* Hitchc., *Bambusa albifolia*, *Saccharum spontaneum* T.H.Wen & J.J.Hua, and *Stipa capillata* L.were found *S. spontaneum* T.H.Wen & J.J.Hua was one of the dominant grass species encountered in the entire delta. The extensive rooting system of *S. spontaneum* may be effective in trapping transported sediments coming from the upland.

Pandanus tectorius Parkinson, locally known as a "pandan" is a perennial species which has leaves utilized in making indigenous products such as mats and hats. The presence of prickles along its leaf margins hampers the intrusion of humans in a pandan-dominated area though cutting off its trunk was observed all throughout the delta.

C. equisitofolia J.R.& G.Forst was more prominent in areas closer to the coast. This plant is commonly used for rehabilitation of mined out areas since it is prolific and grows very fast even in very disturbed sites. Other typical beach forest plants found on the site were *T. catappa* L., *C. inophyllum* L, *I. pes-caprae* (L.)R.Br. and some species of mangroves.

X. verdugonianus Naves commonly known as "magkono" was one of the plant species found in the entire delta with a height less than a meter. This plant is native to the Philippines and considered as a vulnerable plant species. It is commonly found in an ultramafic area which is the soil characteristic of the study site and because of the inherent hardness of its wood it has been harvested at a faster rate.

Mangrove species *Rhizophora sp*. were present but it is planted as part of the mitigating measures and rehabilitation efforts of the nearby mining company. Roots of *Rhizophora* sp are effective in controlling the influx of silts and sediments towards the coastal area.

Table 1. Floral species composition and growth habits of plants encountered in Kinalablaban Delta.

Family	Scientific name	Growth Habits
Adiantaceae	*Adiantum capillus-veneris* L.	Herb
Anacardiaceae	*Buchanania arborescens* (Blume)	Tree
Araliaceae	*Polycios nodosa* (Blume) Seem	Tree
Araceae	*Pothos longipes* Schoot	Vine
Asparagaceae	*Liriope muscari* (Decne.) L.H.Bailey	
Asparagaceae	*Liriope spicata* (Thunb.) Lour	
Aspleniaceae	*Asplenium scolopendium L.*	Herb
Boranginaceae	*Cordia dichotoma* G.Forst.	Tree
Casuarinaceae	*Casuarina equisetifolia* J.R.& G.Forst.	Tree
Cannabaceae	*Tremna orientalis* L. Blume	Tree
Clusiaceae	*Calophyllum inophyllum*	
Convolvulaceae	*Ipomea pes-caprae* (L.)R.Br.	Vine
Combretaceae	*Lumnitzera racemosa* Willd.	
Combretaceae	*Terminalia catappa* L.	Tree
Combretaceae	*Lumnitzera littorea* (Jack) Voigt	
Dipterocarpaceae	*Hopea acuminata* Merr.	Tree
Ebenaceae	*Diospyrus pilosanthera* Blanco	Tree
Euphorbiaceae	*Homalanthus populifolius* Graham	Shrub
Fabaceae	*Acacia auriculiformis* A.Cunn ex Benth.	Tree
Fabaceae	*Acacia mangium* Willd.	Tree
Fabaceae	*Paraserianthes falcataria* (L.) Nielsen	Tree
Fabaceae	*Colophospermum mopane* (J. Kirk ex Benth.) J. Léonar	Tree/Vine
Fabaceae	*Leucena leucocephala* (Lam.) de Wit	Tree
Fabaceae	*Mimosa pudica* L.	
Gleichnaceae	*Dicranopteris linearis* (Burm.f.) Underw	
Lamiacea	*Gmelina arborea* Roxb.	Tree
Lythraceae	*Sonneratia alba* Sm.	Tree
Meliaceae	*Sandoricum koetjape* (Burm.f.) Merr.	Tree
Moraceae	*Artocarpus altilis* (Parkinson) Fosberg	Tree
Moraceae	*Ficus heteropleura* Blume	
Moraceae	*Ficus benjamina* L.	
Moraceae	*Ficus pseudopalma* Blanco	Tree
Mrytaceae	*Xanthostemon verdugonianus* Naves	Tree
Mrytaceae	*Psidium guajava* L.	Tree
Oleaceae	*Ligustrum vulgare* L.	Shrub
Pandanaceae	*Pandanus tectorius* Parkinson	Shrub
Poaceae	*Imperata cylindrica* (L.) P. Beauv.	Grass
Poaceae	*Paspalum conjugatum* P. J. Bergius	Grass
Poaceae	*Muhlenbergia lindheimeri* Hitchc.	Grass
Poaceae	*Bambusa albifolia* T.H.Wen & J.J.Hua	Grass
Poaceae	*Saccharum spontaneum* L.	Grass

Poaceae	*Stipa capillata* L.	Grass
Podocarpaceae	*Podocarpus imbricatus* Blume	
Primulaceae	*Ardisia elliptica* Thunb.	Shrubs
Phyllanthaceae	*Phyllanthus sp.*	
Phyllanthaceae	*Phyllanthus securinegoides* Merr.	
Rhizophoraceae	*Rhizophora apiculata* Blume	Tree
Rhizophoraceae	*Rhizophora mucronata* Lam	Tree
Rubiaceae	*Morinda citrifolia* L.	Tree
Sapotaceae	*Palaquium luzoniense* (Fern.-Vill.) Vidal	
Sapotaceae	*Pouteria sapota* (Jacq.) H. E. Moore & Stearn	Tree
Smilacaceae	*Smilax aspera* L.	Vine
Thymelaceae	*Wikstroemia indica* (L.) C. A. Mey	Shrub
Ulmaceae	*Sponia amboinensis* (Willd.) Decne.	Tree
Urticaceae	*Leucosyke capitellata* (Poir.)Wedd.	Tree
Verbenaceae	*Premna odorata* Blanco	Tree
Verbenaceae	*Lantana camara* L.	Shrub
Verbenaceae	*Stachytarpheta jamaicensis* (L.) Vahl	Herb
Zingerberaceae	*Alipinia rufa* (C.Presl) Náves	Shrub

Table 2. Diversity indices of plants species found in Kinalablaban delta in Claver, Surigao del Norte.

Indices	Transect 1	Transect 2	Transect 3
Diversity	1.02	1.17	0.77
Evenness	0.55	0.52	0.44
Dominance	0.50	0.43	0.58

3.2 Growth Habits of Floral Species Found in Kinalablaban Delta

Most of the plants encountered were species of trees, shrubs, and grasses. Trees were frequent which could indicate that the area is continuously undergoing succession where regeneration of some tree species occurred. Saplings were also observed which accounts for the high occurrence of tree species in the area. Saplings of mangroves encountered were all *Rhizophora sp.* but most of it were planted as part of its reforestation and rehabilitation management system in the delta. Mangroves have specialized rooting systems and are effective in trapping sediments such that its presence in the delta is significant. It provides a protection mechanism from coastline erosion and act as sediment "traps" catching terrestrial sediments and inputs, thus preventing smothering to interconnected coastal ecosystems (Chiarucci *et al.* 2004).

However, roots of *S. alba* were observed to be crooked which could indicate that roots of this species are undergoing stress from root burial due to sediments and high concentration of heavy metal. When sediment burial occurs, mangroves can still be adversely affected by becoming unhealthy, prone to pest and disease, or die. Since the area is always affected by sedimentation coming from the upland, a more suitable species of mangrove could be planted to tolerate rapid sediment accretion. Though roots of *Rhizophora sp* are strongest binders of sediment (Ellison 1998), the monoculture of this plant had high mortality (Scoffin 1990).

A high percentage of shrubs and grasses were also observed in the entire area. Shrubs and grasses were plant communities considered as a pioneer and shade-intolerant species (Rollon and Samson 2008). Hence, the presence of this group may be more dependent on canopy gaps to allow its growth and colonization.

There were only a few vines, ferns, and herbaceous plants encountered. The lesser percentage of vines could be due to its dependency on large plants for support where its maturation takes place (Denslow *et al.* 1990). The growth of some understory plants like ferns and herbs may have been attributed to the availability of light since it was observed in the area that canopy was not totally closed. The presence of light can influence the growth of ferns and herbaceous plants especially when other environmental variables are viable for the growth and establishment of plants.

3.3 Potential Nickel Accumulator

Out of the 59 species that were identified, only four species tested positive as potential nickel hyperaccumulator indicating that presence of such plants in the area were scanty. Plant species that have the potential to be nickel hyperaccumulator were *A. elliptica, P. odorata, P. securinegoides* and *Phyllanthus* sp commonly known as "Haiti". This finding also validates that some plants belonging to Phyllanthaceae are nickel hyperaccumulators found mostly in tropical regions (Reeves 2006). Among the four (4) species that has the potential to accumulate nickel, species belonging to Phyllanthaceae gave the most intense coloration of pink implying a high possibility of nickel accumulation. *P. securinegoides* was recently discovered and confirmed by Van der Ent *et al.* (2015) as nickel hyperaccumulator from Sabah, Malaysia, which also contained the highest accumulated nickel in its leaves. The growth forms of plants that have a potential to be nickel hyperaccumulator were shrubs and herbaceous. Most nickel hyperaccumulators are herbaceous and shrubs which can also be a dual-metal (Cd and Zn) accumulator and tolerant of high levels of lead, zinc and copper (Brooks *et al.* 1991). Most of the plants species that naturally grows in the delta have no reaction with DMG during the semi-quantitative test.

4. Conclusion

Out of the 59 species were identified, only four (4) species of plants tested positive in potential nickel accumulator. These plants species were *A.elliptica, P.odorata, Phyllanthus sp*, and *P. securinegoides* which were shrubs and herbaceous. This finding supports that notion that only few nickel hyperaccumulators thrive in a nickel-rich environment and some of the plants are shrubs and herbaceous. Quantifying the level of nickel in the different part of the plants using Atomic Adsorption Spectrophotometer is necessary to validate if the plant that tested positive was indeed a nickel hyperaccumulator. Nickel hyperaccumulators can be planted in the area as a means to take out the nickel from the soil. Diversity, dominance, and evenness were low which could be due to the inherent characteristic of the soil and other ecological factors like symbiotic relationship of plants to the microorganism, the influence of physical factors such as light and nutrient availability. Floral species in the delta can then be monitored and establishment of a long-term monitoring plot is necessary to determine changes and succession of plants across time.

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