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ORIGINAL RESEARCH

Assessment of root knot nematode incidence as indicator of mangrove biodiversity in Lunao, Gingoog City

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ABSTRACT

This study aimed to determine the biodiversity of mangrove species in two different sites; Site 1 (more disturbed) and Site 2 (less disturbed) and assess the incidence and severity of nematode in three mangrove species, Avicennia officinalis, Rhizophora apiculata and Sonneratia ovata. Inventory within the 600 sq. meters or 0.06 of a hectare in Lunao mangrove area, revealed 6 species in three families consisting of 37 individuals in site 1 and 62 in site 2. The most species rich family was Avicenniceae. Mangrove species located at site 2 had a higher value of diversity index than site 1.This indicates that species richness and density were profoundly observed in less disturbed site. The assessment revealed that the severity and incidence of root galls in three mangrove species were higher in site 1(60 %)-more disturbed area compared to site 2 (46.67 %) –less disturbed area. Out of the three mangrove species examined, Sonneratia ovata, obtained the highest % of root knot incidence and severity for both sites 1 and 2. The assessment revealed that the severity and incidence of root galls in three mangrove species were higher in more disturbed area. Several kinds of wastes and accumulated inorganic materials like plastics, rubber and the like are already observable in site 1 since this area is near the open portion prone for possible human activities and disturbance. This shows that site 1 is likely polluted, the incidence of nematodes increases as indicated by greater number of root galls which in turn revealed low diversity value of mangrove species. Hence, nematode incidence can be a potential indicator of sediment quality that affects mangrove diversity.

Keywords: Mangrove, Root-knot nematode, Biodiversity.

Introduction

Philippines coastal system consists of wide variety of plant species and reported as one of the richest worldwide (Calumpong and Menez 1996). An important component of the coastal ecosystem is the mangrove community along the coastal zone usually found within the intertidal zone.

Mangroves are considered to be unique trees due to their capability in thriving in salt water and from unique intertidal forests at the edge of land and sea. The ecosystems of mangrove has been providing home for many animals and macro-organisms (Cannicciet 2008, Nagelkerken et al. 2008). They also offer essential functions and services to coastal population, like timber and other forest products (Bandoranayake 1998, 2002, Waters et al. 2008) and the coastal zone protection (Badola and Hussian 2005, Dahdouh-Guebasetal 2005-Olwig et al. 2007, Barbier et al.).

Mangrove is one of the most threatened ecosystems (Duke et al. 2008).It has declined by 75% over the past 80 years due to the range of anthropogenic pressures. The decline in the forest structure and diversity of plant species in most of the remaining mangrove stand has been alarming since mangroves play a very important function in the coastal ecosystem. Mangroves create unique ecological ecosystem that host rich assemblages of diverse taxa associated with different habitat. The sandy or muddy sediments are home to a variety of epibenthic or endobenthic, macro invertebrates and meio-invertebrates. An invertebrate roundworm nematodes are living in marine, terrestrial environments and freshwater. They dominate numerically in the mangrove endofauna as they do in others benthic environments.

Nematodes cause to reduce the plants uptake of nutrients and water reducing tolerance to other stresses such as drought. Some transmit other disease that caused agents (eg. viruses) to plants as they feed. Plant–parasitic nematodes are being recognized as major agricultural pathogens and mostly known to attack plants throughout the world that cause crop losses (Sasser and Freckman 1987). Severe infestation can lead to the death of a large number of plants (Hooks et al. 2006).

In addition, symptoms of root knot nematode infestation are reduced vigor and yield, patches of unevenly sized tress, and characteristic galls on roots. The nematode produces terminal galls 0.5-10mm. in diameter and adapted to a marine habitat. If the galls are opened in tap water, the nematodes burst (Siddiqi and Booth 1991). Damaged cause by nematodes can reduce growth by as much as 10-20%. From the study of Severity of Root knot nematode (*Meloidogyne* spp.) on eggplant in Chitwan, results revealed that almost all plants known from nursery stage have been attacked by plant-parasitic nematode (Adikari et al. 2008).

The production of *Cleome gynandra*, *Solanum nigrum*, and *Amaranthus* spp. which are among Indigenous Leafy Vegetables (ILV) grown in Kenya was also constrained by pest and disease and 5% yield losses were caused by Root knot nematodes (RKN) globally to cultivated crops (Nchore 2014).

In the country of Fiji, the incidence of root knot nematode (*Meloidogyne spp.)* was investigated.

It has been reported that the severe ginger damage by root knot nematode with infected rhizomes rendered unacceptable for export (Graham 1971). Based on a study on the host status to RKN concluded that 9 of 16 graminaceous forage crops and 13 of 18 legume species in Fiji were hosts of one or more RKN and can adversely affect crop production. By observing the extent of galling on the root system the severity of RKN infection was determined while the incidence of RKN was detected through visual examination of plant roots for characteristic root galls. This study was made to determine the current distribution and occurrence of RKN in agricultural areas on Viti Level Island, Fiji.

Furthermore, in Pakistan and other countries, the presence of root knot nematode with different medicinal plants and fruit trees and vegetables ornamental have been reported (Khan et al. 2006). Two species of RKN; *M. javanica* and *M. incognita* have been found to parasitize potato in almost all potato growing areas of Punjab (Anwar et al. 1991).Deduction of yield on vegetables especially to potato crops grown in the sandy loam soil of Punjab was caused by *M. incognitae*.

A new root knot nematode species, *Meloidogyne (Hypsoperine)* mersa found parasitising roots of mangrove, *Sonneratia alba* trees, in Pulau Bedukang, Brunei Bay, Brunei Darussalam, was described and illustrated (Siddiqi and Booth 1991). The impact of nematode disease and the browning intensity were appreciably higher in the infected plants at the polluted sites such as coal-smoke pollutants (Khan 1993).

Among other benthic taxa, root knot nematodes seem to be most successful in colonizing the organically enriched oxygen poor environments (Alongi 1987, Olafson et al. 2000). Some nematodes can survive in harsh, polluted, disturbed environments better than others, and some have short life cycles and respond to the environmental changes rapidly (Wang and McSorley 2005). Nematodes turned out to be a suitable organism group for monitoring sediments quality (Heininger et al. 2006) and some nematode genera are indicator for polluted sediments (Ababio 1999) since they are found to be tolerant to pollution.

In the province of Misamis Oriental Philippines, Barangay Lunao, Gingoog City has a mangrove area. However, this mangrove area is located along the river bank near the residential community. Thus, this is not exempted to future deterioration due to growing population and various human activities in the community.

Hence, this study was undertaken to compare the mangrove species diversity of each site and determine the incidence and severity of nematode in selected mangrove species in more disturbed and less disturbed area. This assessment provides additional information regarding the incidence of nematode in mangroves species at two sites. It will promote environmental awareness on the effect of nematode infestation to mangrove trees in coastal ecosystem.

Materials and methods

Protocol Entry

The Local Government Unit through the Barangay Council was formally informed to get permit and approval to conduct the study.

Survey of the place and establishment of sampling site

An ocular survey was carried out in Barangay Lunao, Gingoog City, to determine the different sites to be studied. The specific locations (longitude and latitude) were determined using a GPS gadget. Two sites were selected. Site 1 (N $08^0$50'51.741", E $125^0$03'56.234)- near the open area and accessible to people entry; hence, classified as more disturbed while Site 2 (N $08^0$50'549.980", E $125^0$03'55.547) - far from the open area; hence, classified as less disturbed.

In each site, three subplots of 10 x 10 meters were established. All mangrove trees = or > 10 cm in diameter were counted. Field collection of branches, flowers and other morphological features were done until all species in the three subplots were represented in the collection. The specimens were placed in a plastic bag and properly labeled for further identification.

Identification and collection of specimens

The specimens were identified and classified using some taxonomic keys. Specimens were also brought to DENR for further identifications.

Determination of biodiveristy index

Biodiversity of mangrove species was determined using Shannon - Wiener Diversity using the PAST software.

Nematode bioassays (Taylor *et al*. 1978)

Three mangrove species were selected for the study of nematode incidence namely *Avicennia officinalis*, *Rhizophora apiculata* (Bakauan), and *Sonneratia ovata (*Pagatpat). Five samples of roots (about 100 grams) per mangrove species of were collected within the established sites.

a. Root Samples (Gall Ratings)

The roots were carefully washed with tap water and blot dried. Each root samples was checked through visual examination of plant roots for characteristic root galls.

Nematode incidence was determined using the formula;

$$\text{Incidence} = \frac{\text{Numberofplant galled}}{\text{Total numberofplants sample}} X\ 100$$

b. Severity of Root Galls

RGS 1 - no galls

RGS 2 -0-4 galls

RGS 3 - 5-12 galls

RGS 4 - 13-40 galls

R GS 5 - > 40 galls

Physico-chemical measurement

The physico-chemical (temperature, dissolved oxygen, conductivity, pH, total dissolved solids, and salinity) parameters of the surrounding water were measured.

Results

As shown in table 1, there were 6 species in three families found in the established sites. There were five species observed in site 1 (more disturbed) while six species were noted in site 2 (less disturbed).The family Avicenniaceae was represented by many individuals. Among 6 species, *Avicennia officinalis* had the highest number of individuals having 45 followed by *Sonneratia ovata* and *Rhizophora apiculata* with 19 and 13 individuals respectively.

Table 1. List of species and Number of individuals occurring in two sites.

FAMILIES	SPECIES	Site 1 (More Disturbed)				Site 2 (Less Disturbed)			
		Plot 1	Plot 2	Plot 3	Total	Plot 1	Plot 2	Plot 3	Total
Avicenniaceae	*Avicennia officinalis*	11	4	4	19	21	3	2	26
	Avicennia marina	2	2	1	5	2		2	4
	Avicennia lanata		2		2		4	2	6
Rhizophoraceae	*Rhizophora apiculata*	1		5	6	1	3	3	7
Sonneratiaceae	*Sonneratia ovata*		4	1	5	6	5	3	14
	Sonneratia alba				0	3		2	5
	Total	14	12	11	37	33	15	14	62

Table 2 revealed the total number of individuals in each site. There were 37 individuals in site 1 (more disturbed) while 62 individuals in site 2 (less disturbed).Furthermore, Tables 2 also showed the biodiversity index of site 1 and site 2 which had a value of 1.336 and 1.553 respectively.

Incidence of nematode in three mangrove species was presented in Table 3. There were five samples tested for each mangrove species namely *Avicennia officinalis, Rhizophora apiculata and Sonneratia ovata,* at two different sites. In Site 1- (more disturbed) *Sonneratia ovata,* had the highest percentage of nematode incidence with the value of 80% and followed by *Rhizophora apiculata* with 60 %. While *Avicennia officinalis* had the least incidence of 40%. The same result was observed for Site 2- (less disturbed). *Sonneratia ovata still* had the highest percentage of nematode incidence with the value of 80% and followed by *Rhizophora apiculata* (40%) and *Avicennia officinalis* (20%).Mangrove species located in Site 1 (60 %) had a higher incidence of nematode infestation compared to Site 2 (46.67 %).

Table 2. Shannon-Wiener Diversity Index.

SITE	Number of Families	*Number of Species*	Number of Individuals	Shannon-Wiener Diversity Index H'
1 (More Disturbed)	3	*5*	37	1.336
2 (Less Disturbed)	3	*6*	62	1.553

Table 4 showed the severity of root galls in three mangrove species at two different sites. For site 1-more disturbed, *Rhizophora apiculata* and *Sonneratia ovata* had a mean of 15 and 23 galls respectively which were group in RGS 4 (13-40 galls) while *Avicennia officinalis* was in RGS 2 for having a mean galls of 7. In site 2- (less disturbed) *Avicennia officinalis* had RGS of 4, while *Rhizophora apiculata* and *Sonneratia ovata* had 2 and 3 respectively.Of the three mangrove species*; Sonneratia ovata* was frequently and severely infested with root knot compared to *Rhizophora apiculata* and *Avicennia officinalis.*

Table 3. Incidence of Root Knot in three Mangrove Species.

Samples	Incidence (%)					
	Site 1(More disturbed)			Site 2 (Less disturbed)		
	Avicennia officinalis	*Rhizophora apiculata*	*Sonneratia ovata*	*Avicennia officinalis*	*Rhizophora apiculata*	*Sonneratia ovata*
R_1	✔	✔	✔	X	✔	x
R_2	X	x	✔	✔	x	✔
R_3	X	✔	✔	X	x	✔
R_4	X	✔	x	X	✔	✔
R_5	✔	x	✔	X	x	✔
%	**40%**	**60%**	**80%**	**20%**	**40%**	**80%**
Mean			**60%**			**46.67 %**

Table 5 showed the selected physicochemical parameters of the two sampling sites. All the values of the selected parameters were within the acceptable limits. There is a significant difference in the DO, temperature, conductivity, TDS and salinity in the two sampling sites but the pH value of the two sites did not differ significantly.

Table 4. Severity of Root Knot Incidence in three Mangrove Species.

	Severity					
	Site 1(More disturbed)			Site 2 (Less disturbed)		
	Avicennia officinalis	*Rhizophora apiculata*	*Sonneratia ovata*	*Avicennia officinalis*	*Rhizophora apiculata*	*Sonneratia ovata*
R_1	5	11	28	0	3	0
R_2	0	0	22	18	0	4
R_3	0	25	8	0	0	17
R_4	0	8	0	0	5	12
R_5	9	0	35	0	0	9
Mean	7	15	23	18	4	10
RGS	**3**	**4**	**4**	**4**	**2**	**3**

RGS 1 - (no galls) RGS 2 - (0-4galls) RGS 3 - (5-12 galls)

RGS 4- (13-40 galls) RGS 5 - (> 40 galls).

Discussions

In this assessment, five mangrove species were found to be located in site 1(more disturbed) while site 2 (less disturbed) has 6 species with the number of individual of 37 and 62 respectively.

On the other hand, the Shannon index is one of several diversity indices used to measure diversity in categorical data. Typically the value of the index ranges from 1.5 (low species richness and evenness) to 3.5 (high species richness and evenness) (Shannon, 1949). Table 2 showed that the mangrove species located at site 2 had a higher value of diversity richness of 1.553 than site 1 with a value of 1.336. This indicates that species richness and density were profoundly observed in less disturbed site. However, this diversity value (1.553) for site 2-less disturbed only lie on the border line (1.5).

This denotes that the biodiversity of the said area is not stable and is at risk if conservation measures and actions are not done.

Assessment of root galls severity and incidence of nematodes in the mangrove communities in selected mangrove species was investigated at two different sites. It has been observed that *Sonneratia ovata,* obtained the highest % of root knot incidence and severity for both sites 1 and 2. The result is in conformity to the report cited by Khan (2015) that mangrove ecosystem has been found to support numerous nematodes. And also to the study of Siddiqi and Booth (1991) that some nematode species were found parasitizing the roots of mangrove *Sonneratia alba* in Brunei bay, Brunei Darussalam.

Table 5. Mean Value of the Physico-chemical features of the two sampling sites.

Parameters	Standard Values	Sampling Sites	
		Site 1 (More disturbed)	Site 2 (Less disturbed)
1.DO	> 5 mg/L	7.37*	7.50*
2.Temperature	^{o}C	29.1*	27.2*
3.pH	6.5-8.5	7.86	7.89
4.Conductivity	<1,500 uS/cm	22.5*	25.4*
5.TDS	<1000 mg/L	13.10*	15.31*
6.Salinity	< 0.5 mg/L	13.52*	15.87*

* The mean difference is significant at the .05 level.

Furthermore, incidence of nematode was found to be higher in more disturbed area than in less disturbed area. This idea was supported by Wang and McSorley (2005) where nematodes are found to survive in harsh, polluted area and disturbed environment. Certain study also revealed that the impact of nematode disease was appreciably higher in the infected plants at the polluted sites, (Khan 1994). Ababio (2005) also reported that some genera of nematode are good indicator for polluted sediments.

Nematode incidence and abundance were reported to be correlated with sediment quality and anthropogenic contaminations (Heininger 2006), thus there is a possibility that the diversity of mangrove species in Lunao area will continue to decline in the near future because of the high nematode incidence noted in the sites studied.

Root knot nematodes are reported to be one of the most successful taxa in colonizing the organically enriched oxygen poor environments (Olafson et al. 2000). If this incidence is not addressed, this will cause losses in the production and growth of mangrove species and may also lead to death in the standing mangrove trees in the area.

Conclusions

Inventory of mangrove species in Lunao area, Gingoog City within the 600 sq. meters or 0.06 of a hectare revealed 6 species in three families consisting of 37 individuals in site 1 (more disturbed) and 62 in site 2 (less disturbed). The results suggest that there is more number of standing mangrove trees in less disturbed site.

The most species rich family was Avicenniceae.

Mangrove species located at site 2 had a higher value of diversity index than site 1.This indicates that species richness and density were profoundly observed in less disturbed site.

The assessment revealed that the severity and incidence of root galls in three mangrove species were higher in site 1-more disturbed area compared to site 2 –less disturbed area. Out of the three mangrove species examined, *Sonneratia ovata,* obtained the highest % of root knot incidence and severity for both sites 1 and 2.

Several kinds of wastes and accumulated inorganic materials like plastics, rubber and the like are already observable in site 1 since this area is near the open portion for possible human activities and disturbance. This shows that in site 1 which is likely polluted, the incidence of nematodes increases as indicated by greater number of root galls which in turn revealed low diversity value of mangrove species. Hence, nematode incidence can be a potential indicator of sediment quality that affects mangrove diversity.

It is recommended that anthropogenic pressure and other factors in lined with pollution should be controlled to lessen plant-parasitic nematode which causes great damage to plants.

Therefore, it is further recommended that the mangrove forest in this area be continuously monitored and conserved due to its important function in the ecosystem. Since the area is located near the residential community, intensive protection shall be made.

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