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# Benthic Macroinvertebrate Community as an Indicator of Stream Health: The Effects of Land Use on Stream Benthic Macroinvertebrates

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## ABSTRACT

Biomonitoring of stream health in the tropics still emphasize on the use of standard water chemistry methods (physicochemical variables), which require expensive and elaborate measuring apparatus. In this study, the reliability of benthic macroinvertebrates as bioindicators of freshwater streams was carried out. The study also attempted to determine the discriminating power of various biotic indices in characterizing sites across land use. Benthic macroinvertebrate samples were obtained from nine streams in Silago, Southern Leyte and were identified to family level. One-way analysis of variance was performed on various biotic indices to assess the water quality of streams based on land use. Average Tolerance Score per Taxon (ATSPT) was the only biotic index that differentiated the nine streams based on land use (P<0.001). Forested sites achieved the lowest ATSPT score, whereas mixed forestedagricultural sites had the highest ATSPT scores. Physicochemical variables (e.g., stream width, conductivity, total dissolved solids, water temperature) and biological metrics (e.g., Simpson's diversity index, total macroinvertebrate density) used in the study supported this assessment. The results show that benthic macroinvertebrates can be used as potential biomonitoring tool to evaluate the ecological integrity of

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waterways in the country. Long-term data sets will be generated from future sampling efforts for the development of the Philippine Biotic Index.

*Keywords*: Average Tolerance Score per Taxon (ATSPT), biotic indices, stream monitoring, physicochemical, Philippines

# INTRODUCTION

Habitat degradation due to rapid population growth and economic development intensifies global decline in biodiversity and ecological functionality of freshwater ecosystems. Because of the increase in human land use pressure, the following threats imperil stream habitats (Karr 1991; Brisbois et al. 2008; Miserendino et al. 2011; McGoff et al. 2013): deforestation, land conversion, contaminant pollution, alteration of stream channels, and excessive nutrient input. Such disturbances have led to the disruption of ecological integrity because of the resulting decrease in primary production (Henley et al. 2000), altered trophic structure (Gregory et al. 1991), modified channel dynamics (Walsh et al. 2001), and reduced bank stability (Findlay et al. 2001).

Several assessment and monitoring strategies have been implemented to assess the biological quality of freshwater habitats and to sustain human and ecological demands for fresh waters. For example, traditional stream assessments are generally performed using water chemistry, wherein physicochemical parameters, namely dissolved oxygen (DO), temperature, conductivity, total dissolved solids, water hardness, and water flow rate are recorded and analyzed in situ (Dinka et al. 2004; Halstead et al. 2014). However, this method was deemed inefficient in providing thorough habitat evaluation due to underlying constraints (Scrimgeour and Wicklum 1996; Heatherly et al. 2007). This then paved the way for the emergence of new approaches (i.e., biological monitoring or biomonitoring) in making comprehensive analysis of the overall condition of freshwater ecosystems.

Biomonitoring utilizes a wide array of organisms as biological indicators (or simply bioindicators) to determine the overall status of stream habitats. Diatoms are used because of their ubiquity, short generation time, broad range of tolerance against contaminants, ease of use, and well-documented taxonomy (Kireta et al. 2012; Mendes et al. 2012). Fishes are also used due to their well-known community structure and recreational value (Carey and Mather 2008; Resh 2008). Macroinvertebrates, which are indispensable components of aquatic ecosystems,

are widely used indicator species in freshwater biomonitoring because of a set of distinct advantages they offer (Reece and Richardson 2000; Barbosa et al. 2001; Clements et al. 2002; Bae et al. 2005): their ubiquity and sedentary nature makes them good representatives for spatial analyses of pollutants; their relatively longer life cycles compared to other freshwater organisms can elucidate temporal changes; their constant exposure to varying water quality conditions allows them to accumulate toxins from the sediments they live in and feed on; and their well-described taxonomy aids in the ease of identification and evaluation of collected samples.

Several studies have considered the use of abundance and species richness among macroinvertebrates to detect environmental responses because of their variable sensitivity towards multiple disturbances (Davis 2003; Ferreira et al. 2011; Friberg et al. 2011). Moreover, this set of organisms does not experience rapid blooms and death in response to nutrient inputs compared to algae. They also do not possess great mobility similar to that of fishes, preventing them to escape pollution by moving towards unaffected tributaries (Morse et al. 2007).

Unlike in temperate regions, benthic macroinvertebrates are underutilized, poorly established, and rarely applied to tropical freshwater assessments. This happens due to a great deal of challenges occurring among tropical streams (Clews et al. 2014; Feio et al. 2015), such as the paucity of information on the taxonomy of faunal groups, low efficiency of biotic indices, differences in community structure, variation in functional processes, and seasonal variation. However, there is an increasing interest in studying tropical streams using benthic macroinvertebrates.

The municipality of Silago, Southern Leyte (10° 31'45" N, 125° 9'56" E, total land area = 21,505 ha) provides an excellent study site for macroinvertebrate assemblages. The study attempts to determine the validity of extensively used biotic indices (e.g., Hilsenhoff's Family Biotic Index, Biological Monitoring Working Party (BMWP), Average Score per Taxon (ASPT), SingScore, and Average Tolerance Score per Taxon) in providing preliminary assessment of Silago's current stream condition across land use. The study also tested the efficiency of several physicochemical parameters (e.g., water temperature, conductivity, total dissolved solids) and biological metrics (e.g., Simpson's diversity index, total macroinvertebrate density) in describing the ecological integrity of the selected streams. Since there are no published studies on benthic macroinvertebrates in Silago, baseline data from the results will be useful for the development of the Philippine biotic index for freshwater streams.

### MATERIALS AND METHODS

#### **Physicochemical Parameters**

Previously collected data on various physicochemical parameters were used in this study to assess the water chemistry of the nine selected streams in Silago, Southern Leyte. Using a multiparameter water quality meter, the following variables were measured: (i) dissolved oxygen, (ii) pH, (iii) temperature, (iv) conductivity, and (v) total dissolved solids (TDS). In addition, wetted width, water depth, and water velocity were recorded.

#### **Benthic Macroinvertebrates**

This study used the macroinvertebrate samples previously collected from selected streams in Silago, Southern Leyte in June and July 2014, which were deposited at the Aquatic Biology Research Laboratory of the Institute of Biology, University of the Philippines Diliman. Nine streams were surveyed, with each stream having six macroinvertebrate sample collections per location: upstream, midstream, and downstream. These samples were collected using a Surber sampler, stored in 50 mL centrifuge tubes containing 95% ethanol, and were brought to the laboratory for identification.

Using a fluorescent illuminated magnifier, relatively large benthic macroinvertebrates were initially sorted based on morphology. A stereomicroscope was then used to group relatively small individuals. All morphologically-similar organisms were immediately placed in properly labeled 15-mL centrifuge tubes containing 95% ethanol. After sorting, the taxonomic family level of the macroinvertebrates were identified using the keys of Dudgeon (1999), Yong and Yule (2004), and the Mekong River Commission (2006). Finally, all identified samples were transferred into individual 20-mL scintillation vials, with each vial containing only one family per sampling site. All vials were properly labelled with the name of the site, the date of collection, and the respective taxonomic family.

Using the macroinvertebrate data, the following biological metrics were calculated: (i) total invertebrate density, (ii) taxon richness, (iii) richness of Ephemeroptera, Plecoptera and Trichoptera (EPT) insect orders, and (iv) Simpson's Index of Diversity. Moreover, widely accepted biological scoring systems were calculated to determine the current condition of the streams in Silago Southern Leyte: (i) Hilsenhoff's Family Biotic Index (HBI), a biotic index for assessing organic and nutrient pollution using tolerance values of arthropod families (Hilsenhoff 1988); (ii) Biological Monitoring Working Party (BMWP), a standardized score system based on tolerance scores of macroinvertebrate families to organic pollution (Mustow 2002); (iii) Average Score per Taxa (ASPT), a biotic index which measures river status using the calculated BMWP score divided by number of taxa (Mustow 2002); (iv) Stream Invertebrate Grade Number – Average Level version 2 (Signal 2), a biotic index for Australian river macroinvertebrates (Chessman 1995, 2003); (v) SingScore, a newly developed biotic index for measuring the health of Singapore's streams using benthic macroinvertebrates (Blakely et al. 2014); and (vi) Average Tolerance Score per Taxon (ATSPT), a biotic index for evaluating stream health integrity using site disturbance scores and benthic macroinvertebrate abundance (Chessman and Giap 2010).

## **Data Analysis**

Data were log10(x) or log10(x + 1) transformed to improve normality and homoscedasticity after exploratory data analysis (Quinn and Keough 2002), where necessary. One-way ANOVA was performed to determine significant difference across land use for the various physicochemical, benthic macroinvertebrate metrics, and biotic indices (Magbanua et al. 2010; Narangarvuu et al. 2014; Aguiar et al. 2015). If land use had a significant effect, pairwise comparisons with Tukey's HSD (or Games-Howell, in cases of persisting heteroscedasticity) post hoc tests were conducted.

## **RESULTS AND DISCUSSION**

## **Physicochemical Variables Across Land Use**

All variables, except stream depth and DO, showed significant differences across land use (*P*<0.05 in all cases; Table 1). Forested land use had the lowest mean values for all parameters other than pH (Figure 1). Water physicochemistry, particularly water temperature, conductivity, TDS, pH, water velocity, and stream width, showed significant results in discriminating selected streams across land use.

	Land Use						
Parameter	Forested	Agricultural	Mixed	P-value	Ranking		
Stream width	4.67 (0.42)	8.66 (1.48)	17.87 (1.49)	<0.001	F <a<m< td=""></a<m<>		
Stream depth	0.13 (0.01)	0.16 (0.01)	0.17 (0.03)	0.341			
Water velocity	0.31 (0.09)	0.42 (0.09)	0.54 (0.20)	0.003	F <a<m< td=""></a<m<>		
Temperature	23.60 (0.09)	25.24 (0.35)	26.59 (0.18)	<0.001	F <a<m< td=""></a<m<>		
Conductivity	6105.09 (912.07)	12216.31 (1201.71)	12965.23 (231.08)	<0.001	F <a<m< td=""></a<m<>		
TDS	4113.73 (611.34)	7931.57 (795.90)	8173.66 (139.15)	<0.001	F <a<m< td=""></a<m<>		
pН	7.79 (0.19)	7.17 (0.34)	9.62 (0.74)	0.012	A <f<m< td=""></f<m<>		
DO	5.09 (0.35)	5.39 (0.61)	5.54 (0.52)	0.930			

Table 1. Mean (± standard error) stream physicochemical param	eter values
of selected streams in Silago, Southern Leyte across different	land uses
F = forested; A = agricultural; M = mixed	

Data analysis revealed that forested areas had the lowest water temperature as opposed to the other land uses. This supports the prediction that forested sites are abundant in diverse sets of trees and vegetation, contributing to the canopy cover which provides shade (Studinski et al. 2012). On the other hand, both agricultural and mixed areas achieved a relatively warmer temperature due to the decrease in the surrounding riparian zone. Moreover, as reflected in its narrow stream width, forested sites had stable banks, which is indicative of the rich vegetation that holds the soil intact and reduces the effects of erosion. However, the case was different among agricultural and mixed sites, which generated higher measurements for their respective stream width due to poor bank stability caused by farming practices and other land development occurring in the area.

The high water conductivities within agricultural and mixed areas suggest excess nutrient inputs in these particular sites. This is expected due to the presence of farming activities, which contribute to increased fertilizer and pesticide loading via terrestrial runoff (Al-Shami et al. 2011; Piggott et al. 2012). Forested sites, in turn, had low measurements for both conductivity and TDS, indicating minimal anthropogenic activity.

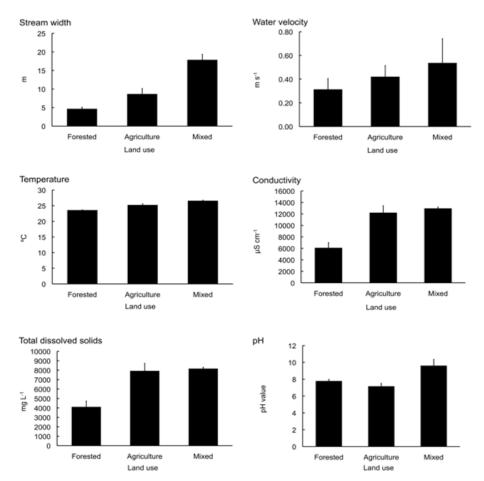


Figure 1. Mean (± standard error) stream physicochemical parameter values of selected streams in Silago, Southern Leyte across different land uses.

### **Biological Response Variables Among Streams**

Two of the biological metrics used, namely total benthic macroinvertebrate density (P=0.001) and Simpson's Diversity Index (P=0.030), markedly differed across land use (Figure 2). Apart from physicochemical values, diversity indices are also extensively used in assessing the health of freshwater streams. Diversity indices measure the degree of diversity of benthic macroinvertebrates in a particular site and provide an evaluation of its condition (Lenat 1988; Death and Winterbourn 1995; Linke et al. 1999). In fact, there is a wide selection of diversity indices that can be used to determine water quality (Carter et al. 2009); however, only the Simpson's Diversity Index was selected for this study. Simpson's Diversity Index does not only give the number of different taxa present across sites, but it also measures the evenness of the distribution of individuals amongst taxa (Bailey et al. 1998).

Forested sites had the greatest number of taxa, whereas agricultural sites exhibited the least number of taxa. This alone immediately suggests that the water quality among forested sites is indeed excellent as opposed to the other land uses. However, care should be taken on solely using diversity indices because of the possibility of obtaining a false assessment (Wilsey et al. 2005; Heino et al. 2008).

Accurate stream health assessments could be constructed from considering the type and abundance of the present taxa (e.g., pollution-tolerant, pollution-sensitive). For example, macroinvertebrates belonging to the Ephemeroptera-Plecoptera-Trichoptera (EPT) orders are highly sensitive to organic pollution (Henriques-de-Oliveira et al. 2007; Narangarvuu et al. 2014; Zaiha et al. 2015), which makes them good water quality indicators. Pollution tolerant species, on the other hand, are insensitive to various environmental stressors, allowing them to thrive even in heavily degraded habitats (Guimaraes et al. 2009; Frizzera and Alves 2012; Rosa et al. 2014).

The taxa compositions across the three land uses were different (Figure 3). Agricultural and mixed land uses exhibited a greater number of EPT insect orders as opposed to those of forested sites. This observed pattern was most likely due to increased input of organic nutrients from farmlands, which could have possibly given the macroinvertebrates additional opportunity to increase in number (Niyogi et al. 2007; Wagenhoff et al. 2011). Interestingly, the preponderance of Baetidae and Hydropsychidae among agricultural and mixed areas could also be attributed to the introduction of nutrients from the surrounding land uses. Several studies have shown that both taxa are commonly abundant in mildly polluted, nutrient enriched areas (Czerniawska-Kusza 2005; Ratia et al. 2012; Xu et al. 2014).

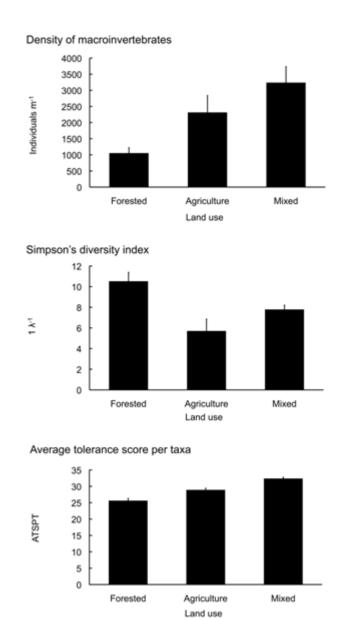


Figure 2. Mean (± standard error) biological metric values and indices of selected streams in Silago, Southern Leyte across different land uses.

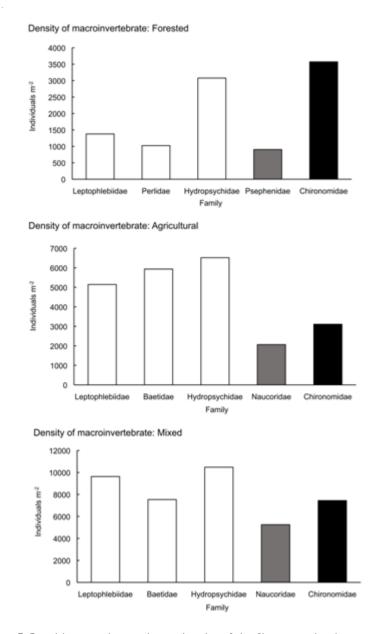


Figure 3. Benthic macroinvertebrate density of the five most dominant taxa across different land uses. White = pollution-sensitive taxa; gray = moderately pollution-tolerant taxa; black = pollution tolerant taxa. Macroinvertebrate classification followed Chang et al. (2014).

Hydropsychidae larvae (Trichoptera) are reported to be sedentary filter feeders that commonly inhabit fast flowing rivers (Andersen and Klubnes 1983). Due to their mode of feeding, this taxon greatly prefers streams with high concentration of suspended organic matter, which may originate from various sources (e.g., fish farm effluents, waste treatment plants) (Guilpart et al. 2012). The observed high density of Hydropschidae in agricultural sites was consistent with other studies that showed increase in Hydropsychidae density in areas influenced by farm-related activities (Strand and Merritt 1997; Kyriakeas and Watzin 2006; Dahal et al. 2007).

Baetidae larvae (Ephemeroptera) are regarded as tolerant species against organic pollution (Zamora-Munoz and Alba-Tercedor 1996; Buss et al. 2002), which explains their occurrence in sites experiencing intermediate levels of degradation. However, the results failed to support the findings of Timm et al. (2001) and Buluta et al.

Table 2. Mean (± standard error) biological metric values and indices of selected streams in Silago, Southern Leyte across different land uses. EPT = Ephemeroptera-Trichoptera-Plecoptera insect orders; HBI = Hilsenhoff's Family Biotic Index; BMWP = Biological Monitoring Working Party; ASPT = Average Score per Taxa; SIGNAL 2 = Stream Invertebrate Grade Number – Average Level version 2; SingScore = Singapore's macroinvertebrate biotic index Score; ATSPT = Average Tolerance Score per Taxon. F = forested; A = agricultural; M = mixed

	Land Use						
Parameter	Forested	Agricultural	Mixed	<i>P</i> -value	Ranking		
Taxon richness	19.83 (1.54)	16.33 (1.68)	18.17 (0.70)	0.117			
Invertebrate density	1054.11 (165.06)	1894.11 (374.49)	3239.44 (492.78)	<0.001	F <a<m< td=""></a<m<>		
Simpson's diversity	10.52 (0.86)	6.53 (0.81)	7.79 (0.41)	0.002	A <m<f< td=""></m<f<>		
EPT richness	8.56 (0.64)	8.00 (0.72)	9.56 (0.34)	0.116			
HBI	3.35 (0.12)	3.19 (0.10)	3.34 (0.07)	0.462			
BMWP	80.78 (6.03)	73.89 (7.06)	85.94 (3.35)	0.157			
ASPT	6.34 (0.08)	6.16 (0.20)	6.40 (0.06)	0.287			
SIGNAL 2	5.04 (0.09)	5.15 (0.11)	5.06 (0.04)	0.672			
SingScore	131.61 (1.45)	131.33 (3.01)	134.17 (1.71)	0.567			
ATSPT	25.65 (0.63)	29.74 (0.45)	32.39 (0.36)	<0.001	F <a<m< td=""></a<m<>		

(2010) that consider Baetidae as an excellent indicator of pristine freshwater habitats. This then implies that the stream health among agricultural and mixed sites is considered moderately poor as reflected by the gathered macroinvertebrate data.

Chironomidae, on the other hand, was observed in all types of land uses, which was expected, on account of the ability of this specific taxon to thrive in all habitat types: both in highly polluted and minimally disturbed habitats (De Haas et al. 2005; Loayza-Muro et al. 2012). These taxa are described as pollution tolerant species due to their capability to withstand a wide range of environmental conditions (i.e., temperature, pH, dissolved oxygen). It is also important to note that Chironomidae tend to occur in higher densities when oxygen levels are low and organic pollution is high (Buss et al. 2002; Bacey and Spurlock 2007). However, the direct or indirect effects of nutrient enrichment to Chironomidae density could not be assessed as no manipulative test was performed in our study (Miracle et al. 2006; Wagenhoff et al. 2012).

Despite the low number of EPT orders, only forested areas supported a significant number of taxa belonging to Plecoptera (i.e., Perlidae), which has long been regarded as the most pollution intolerant of the aquatic insect orders. This general claim is supported by various studies concerning the evolution of this particular taxa in the cold mountain streams, where oxygen stress was scarce (Zwick 2000; Chang et al. 2014). Based on this observation, it is reasonable to conclude that the water quality among forested sites is in good condition, especially since these areas were minimally disturbed by anthropogenic activities.

Traditional measures, such as total taxa richness and total EPT richness, were considered helpful in providing stream health evaluation according to several studies (Lammert and Allan 1999; Roy et al. 2003; Moya et al. 2011). However, the results obtained from the data analysis show that the values for both parameters were not significant across sites, suggesting that the two are not reliable tools for biomonitoring.

#### Stream Health Assessment Using Biotic Scoring Systems

Average Tolerance Score per Taxa (ATSPT) significantly differed across land use (*P*<0.001). In contrast, HBI, SIGNAL 2, SingScore, BMWP, and ASPT did not show any significant changes across land use (*P*>0.05 in all cases; Table 2).

Modern stream monitoring and habitat assessments are being conducted using a relatively new technique that uses different biotic indices (Armitage et al. 1983; Hilsenhoff 1988; Chessman 1995; Mustow 2002; Blakely et al. 2014). The method is an example of a numerical estimation, wherein specific taxa are given corresponding tolerance scores depending on their sensitivity towards organic pollution. A final score that indicates the current state of the freshwater system is then obtained. Originally, it was developed for monitoring temperate freshwater system, but it is now being used in tropical countries, including in Southeast Asia.

In this study, only the Average Tolarance Score Per Taxa (ATSPT) of Chessman and Giap (2010) generated highly significant values across sites. The remaining five biotic indices, namely BMWP, ASPT, HBI, SIGNAL 2, and SingScore, failed to discriminate the three land uses in terms of stream health conditions, as evidenced by their corresponding *P*-values.

Based on the results from SingScore, all sites within different land uses achieved excellent water quality, since it has been suggested that SingScore values (>120) indicate optimal stream conditions (Blakely et al. 2014). Similarly, the data obtained from HBI exhibited the same pattern in line with the proposed values (0.00 – 3.75) for excellent water conditions (Hilsenhoff 1988). On another note, it is interesting to mention that, despite the presence of farming activities and human impairment among agricultural and mixed sites, their corresponding water qualities remain excellent. This observation could be due to the lack of large-scale industries (factories and manufacturing plants) in the municipality of Silago, Southern Leyte, which explains why the ongoing anthropogenic activities are not sufficient to heavily impact the waterways. Furthermore, this indicates that the discriminatory powers of SingScore and HBI were not sensitive enough to be used for freshwater habitat assessment.

SIGNAL 2, BMWP, and ASPT failed to discriminate the streams across the three types of land use, proving to be consistent with the works of Wyzga et al. (2013) and Mohmad et al. (2015). This was because the development of these three biotic indices only accounted for organic pollution, which could potentially underestimate/ overestimate the extent of disturbance occurring among impacted sites. It should also be emphasized that the response of benthic macroinvertebrates to different stressors (i.e., organic enrichment, heavy metal contamination) varies across taxa and is greatly influenced by its geographical setting (Chutter 1972).

In contrast, ATSPT characterized the water quality of the streams across the three land uses, which ranged from moderately poor to excellent. Sites within mixed areas were observed to have the highest ATSPT scores, implying their ability to support a great number of pollution tolerant taxa. These moderately poor quality reference sites augment the occurrence of high-surrounding impervious surfaces within these areas, ultimately leading to increased sediment deposition as observed in other studies (Allan 2004; Walsh et al. 2005; Mantyka-Pringle et al. 2014). In turn, forested sites possessed excellent water quality, as evidenced by their low ATSPT scores. From these findings, ATSPT is a potential bioindicator of water quality that can be used in the Philippines.

Accordingly, several key points about this biotic index should be re-assessed and re-evaluated. First, ATSPT is advantageous over the other biotic scoring systems due to the fact that all of the identified taxonomic families across sites were provided with respective tolerance values, which were obtained from the calculated Site Disturbance Score (SDS) from the time of sampling (Chessman and Giap 2010). This essentially removes the idea of excluding all identified taxa not having preassigned tolerance values, as employed by other biotic indices. Second, the habitat assessment performed by assigning values of 1 to 3 (1 = best possible condition; 3 = worst possible condition) for the computation of SDS remains subjective, bringing about changes depending on the person performing the field sampling. Finally, the tolerance value for each taxa remains dependent to the condition of its immediate habitat at the time of collection.

#### CONCLUSIONS

The results show that benthic macroinvertebrates can be used as a bioassessment tool, as it was able to successfully evaluate and determine the conditions of the stream ecosystems under varying land use in Silago, Southern Leyte. Out of the six biotic indices tested, ATSPT shows potential in distinguishing polluted sites from unpolluted ones. This result was also supported by the data reflected in Simpson's Diversity Index, benthic macroinvertebrate composition, and the physicochemical variables. The ATSPT approach is considered advantageous over the widely used physicochemical method for stream bioassessment and biomonitoring, as ATSPT provides a rapid and cost-effective stream health evaluation without requiring expensive sets of elaborate equipment for data collection. Finally, the findings indicate that a long-term data set generated from future sampling efforts will significantly contribute in the protection, conservation, and restoration of the country's freshwater through the development of the Philippine Biotic Index.

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