

# Macro-benthos Diversity in a Headwater Stream Affected by Tea and Paddy Agricultural Runoff, Sri Lanka

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## Introduction

It is well documented that soil, organic matter, fertilizer and pesticides from agricultural lands are transported into adjacent streams with surface runoff changing the quality of those water ways (Neuman and Dudgeon, 2002). The eroded soil particles transported over the agricultural lands include nutrients, pesticides and their residuals. Therefore, soil erosion can cause both physical and chemical impacts in adjacent aquatic systems (Merrigton et al. 2002). Even a thin film of fine sediments in aquatic systems can eliminate or reduce the sensitive taxa population of macro invertebrates, such as Ephemeroptera and Plecoptera (Evans, 1996; Leeks, 1995). However, pesticides transportation is one of the severe impacts on aquatic ecosystem compared to other chemicals such as artificial fertilizers (Liess and Schulz, 1999; Merrigton et al. 2002) altering the dynamics of macro-benthos communities in streams.

The effects of agricultural runoff have been monitored by several authors through the assessments of stream water quality and its biota (Liess and Schulz, 1999; Neuman and Dudgeon, 2002; Song et al., 2009). The changes of physical and chemical nature of water can create diverse biological effects ranging from severe to subtle effect levels (Bartram and Ballance 1996). Ecological methods which are based on the community structure and the presence or absence of species are more prominent in literature than the physical and chemical methods (Bartram and Ballance 1996).

Particularly, macro-benthos are suitable in ecological approach of monitoring an ecosystem (Bartram and Ballance 1996). The abundance and the distribution of macro-benthos are affected by various physical and chemical conditions of water

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such as water depth, water velocity, and organic matter content in sediment and toxicant levels in water and sediments. According to Mandawille (2002) macro-benthos diversity is used to predict the health of water bodies in many studies as they are species rich, sedentary and long lived and therefore can indicate long-term effects on their habitat. Bengtsson (1998) has documented that the long-term goal of managing and developing sustainable ecosystems mostly depend on the understanding of the linkages between key species or functional groups rather than focusing only on the species diversity.

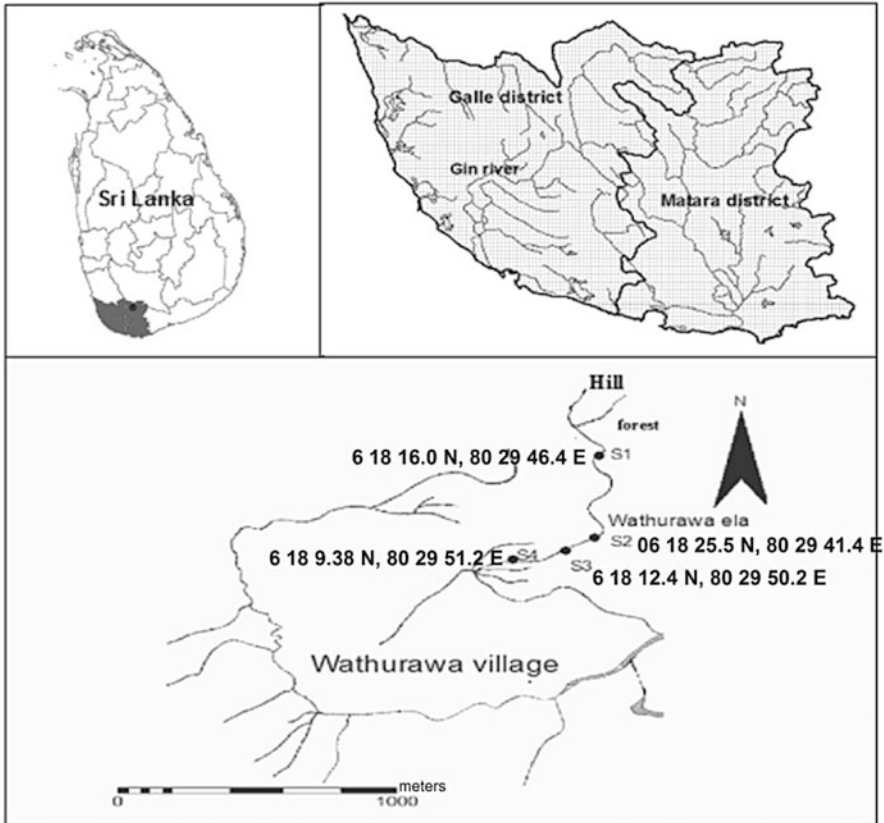
The macro-benthos are secondary producers which form important links in the food webs of aquatic ecosystems (Sharma et al., 2009). Furthermore, they play a major role in transferring energy from the first trophic level to second trophic level in fresh water ecosystems. Therefore, secondary production can be used to evaluate the trophic potential within a particular ecosystem (Tumbolo and Downing, 1994). The quality of a particular ecosystem can be predicted by estimating the trophic potential (Wilber and Clarke, 1998). The higher diversity together with the higher secondary production indicates better environmental quality and vice versa (Munari and Mistri, 2007). Also, the secondary production of macro-benthos can be used as an important tool for monitoring the changes in an ecosystem.

This study assessed the impact of agricultural runoff from both tea and paddy cultivations to a small headwater stream at Wathurawa, Sri Lanka. Headwater streams are defined as all the first and second order streams which make 2/3 of the total length of a river network, and large rivers can be fed by hundreds of thousands of headwater streams (Leopold et al., 1964). These stream ecosystems are critical sites for organic matter processing and nutrient cycling (Bilby and Likens, 1980; Wallace et al., 1997). Sri Lanka is blessed with 103 perennial rivers which are fed with numerous headwater streams radiating from hill country region towards Indian Ocean. As for the use of considerable amounts of agrochemicals for tea and paddy cultivations, and more obvious soil erosion in these hill slopes, it is essential to assess the impacts on macro-benthos diversity and their productivity as the second trophic level for explaining the quality of these headwater streams.

## Materials and Methods

### *Study Site*

Wathurawa is a mountainous remote village surrounded by a forest reserve which is located in between the margin of Matara and Galle administrative districts (Fig. 1). Nearly 300 families living in this area are engaged in paddy and tea cultivation as their major livelihood. These cultivations are extended upto the reserved forest margin towards the crest. Wathurawa stream, which originates from this hill top and later connects with the Gin River, was selected for this study. The stream valley is used for paddy cultivation due to the ease of getting water and draining back to the



**Fig. 1** The sampling sites in Wathurawa stream (S1, S2, S3 and S4)

downstream. Tea plantations are extended throughout the hill slopes facing the stream valley. Four Sampling sites along the stream reach were selected in respect to the ease of access and the points affected by agricultural runoff as follows.

The site 1 (S1) was located at the natural forest margin and it was considered as the reference site. The site 2 (S2) and site 3 (S3) were located at the point of connection of run-off from tea land to the stream while site 4 (S4) was selected as a place where the run-off from both tea and paddy lands are connected to the stream. There was no more variation of the substrate characteristics at all four sampling sites and slightly shaded with stream bank vegetations. Total length of the selected steam reach was about 600 m with sites apart from each other approximately by 100 to 200 m. Samples were taken during the rainy season (average rainfall 579.3 mm) from November 2010 to February 2011 twice a month as both the fungicides application and paddy cultivation are done concurrently throughout this season.

## ***Measurement of Physico-chemical Parameters***

Bottom water temperature (YSI 85, Japan), pH (pH meter: AD 111, Hungary), flow rate (Digital flow meter: Kenek VP 1000, Japan), and depth (Digital depth gauge: Hondex, PS-50C, Japan) were determined using relevant instruments and total suspended solids, total dissolved solids,  $\text{NO}_3^-$ -N, and  $\text{PO}_4^{3-}$  concentrations were determined by standard methods described in APHA (1985).

## ***Macro-benthos Sampling and Analysis***

Macro-benthos were collected using a Surber sampler of 0.5 mm mesh size and with a quadrat area of 25 cm<sup>2</sup>. The Surber sampler was positioned against the stream flow and vigorously stirred the substrate area at nine random points per each site during each sampling occasion. Altogether 54 replicates were collected during six sampling efforts. The benthos collected at the cod end of the Surber sampler were preserved in 4 % formalin and identified to the Family level following the keys of Benjamin et al. (2003), Bouchard (2004), Fernando (2002), Hartmann (2006), and Pescador et al. (1995). They were quantified as individuals/m<sup>2</sup> under each taxa and Alpha diversity of macro-benthos was estimated by counting the number of individuals in each taxonomic group and calculating Shannon-Weiner diversity index for four sampling sites separately. EPT-Index and taxa richness were also calculated for the total number of families including Orders Ephemeroptera, Plecoptera and Tricoptera at each site (Mandawille, 2002).

The dry weight and the Ash Free Dry Weight (AFDW) of taxonomic Orders were determined by drying (110 °C for 24 h) and incineration (550 °C for 1 h) for benthos of each taxonomic group. Secondary production was estimated by using the empirical formula described by Brey (1990), which has been followed by Dara and Duoglas (1998) for miscellaneous macro-benthos groups.

Secondary productivity was estimated for each site using following formula (Brey, 1990):

$$\log P = -0473 + 1.007 \log B - 0274 \log W$$

where  $P$  is annual production (g AFDW m<sup>2</sup>yr<sup>-1</sup>),  $B$  = annual biomass (g AFDW m<sup>-2</sup>); and  $W$  = individual weight (g AFDW).

## ***Data Analysis***

Karl Pearson's correlation coefficient was used to quantify the relationship between hydrological attributes and biological attributes. Mean values of hydrological

attributes in each study site was used to calculate the inter relationships ( $r$ -values) according to Sharma et al. (2008). One way ANOVA was performed to analyze the variation of hydrological and biological attributes between sampling sites using SPSS 16.0 package. Cluster analysis and correspondent analysis (PCA) were done for the physico-chemical parameters at sampling sites with the help of Primer-5 package. Multi-Dimensional Scaling (MDS) was performed to observe the variation of diversity of macro-benthos at each sampling site during the study period.

## Results

### *Physico-chemical Parameters*

The mean values of physico-chemical attributes at four sites are given in Table 1. Wathurawa stream is a shallow stream and the mean depth ranges from 0.15 m to 0.18 m. Water velocity varied within a small range from 0.227 to 0.274 m/s and the highest mean velocity was  $0.274 \pm 0.017$  m/s at the site 2 and the lowest velocity was  $0.227 \pm 0.009$  m/s at the site 3 (Table 1).

There were no significant spatial differences of pH, TSS, TDS,  $\text{NO}_3^-$  and  $\text{PO}_4^{3-}$  at the sampling sites. However, the highest TSS (174 mg/L) and  $\text{PO}_4^{3-}$  (0.033 mg/L) were observed at the downstream site (S4). The highest  $\text{NO}_3^-$  concentration (1.21 mg/L) was observed at S3 and the value was much lower than the EU standards for drinking water (50 mg/L) (Merrigton et al. 2002).

The cluster analysis based on all physico-chemical attributes shows that the similarity between S1 and S2 is about 22 % and the similarity between S3 and S4 is 10 %. Therefore, Fig. 2 shows that the group separation and the similarity between the two groups was 12 % (Fig. 2).

### *Macro-benthos Diversity and Other Indices*

Immature stages of several insect orders such as Plecoptera, Odonata, Diptera, Ephemeroptera, Tricoptera, Hemiptera, Coleoptera and Lepidoptera and some other taxa of Amphipoda, Class Turballaria, Class Gastropoda and Gordea were observed in the stream.

The orders and families found in the stream are given in Table 2. All together 43 taxa were found from the benthos in Wathurawa stream and the highest taxa richness was 36 at site 1.

Mean EPT index values were  $8.67 \pm 1.05$ ,  $6 \pm 0.63$ ,  $6.67 \pm 0.67$  and  $5.17 \pm 0.85$  at the sites of S<sub>1</sub>, S<sub>2</sub>, S<sub>3</sub>, and S<sub>4</sub> respectively. According to paired test of Tukey HSD the EPT index was significantly different between the site 1 and the site 4 ( $P < 0.05$ ). The calculated values of mean Shannon-Wiener diversity index

**Table 1** Mean values of physico-chemical attributes at the four sampling sites in Wathurawa stream

Site	pH	Bottom water Temperature (°C)	Flow rate (m/s)	TSS (mg/L)	TDS (mg/L)	NO <sub>3</sub> <sup>-</sup> (mg/L)	PO <sub>4</sub> <sup>3-</sup> (mg/L)	Depth (m)	DO (mg/L)
1	6.95 ± 0.18	23 ± 0.2	0.241 ± 0.02	2.42 ± 0.55	68.25 ± 18.95	0.56 ± 0.15	0.013 ± 0.003	0.15 ± 0.01	10.84 ± 0.26
2	6.95 ± 0.14	24.1 ± 0.4	0.274 ± 0.02	4.58 ± 1.11	104.12 ± 9.61	0.70 ± 0.24	0.01 ± 0.002	0.17 ± 0.01	11.1 ± 0.15
3	6.81 ± 0.22	24.6 ± 0.5	0.227 ± 0.01	8.12 ± 3.34	90.12 ± 21.24	0.92 ± 0.15	0.01 ± 0.003	0.17 ± 0.01	10.86 ± 0.20
4	6.56 ± 0.27	24.9 ± 0.6	0.267 ± 0.01	36.42 ± 27.60	81.42 ± 19.15	0.85 ± 0.05	0.03 ± 0.017	0.18 ± 0.01	10.97 ± 0.17

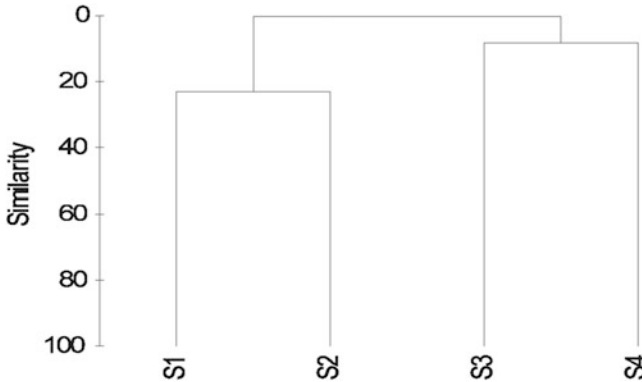


Fig. 2 Cluster analysis for physico-chemical parameters

(S-W index) of the benthic macro-invertebrates are shown in Fig. 3. According to that, the highest index value ( $3.54 \pm 0.14$ ) was at the S1 and the lowest value ( $2.91 \pm 0.33$ ) was at S4. However, there was no significant difference among mean Shannon-Wiener index values at the four stations.

Taxa richness (TR) values were  $20.17 \pm 1.54$ ,  $17 \pm 1.13$ ,  $16.67 \pm 0.88$  and  $12.33 \pm 2.16$  at the sites of S1, S2, S3 and S4 respectively (Fig. 4). According to one way ANOVA there was a significant difference of taxa richness at the four sampling sites ( $P < 0.05$ ). Tukeys HSD multiple comparisons test shows a significant difference of TR between S1 and S4 ( $P = 0.008$ ). It was observed as a clear decline of the sensitive taxa (Ephemeroptera, Plecoptera, Odonata, Tricoptera) towards the downstream (Fig. 5).

The highest density of Tricoptera ( $36.3 \% \text{ ind/m}^2$ ) was found at S1 and the lowest density ( $22.9 \% \text{ ind/m}^2$ ) was found at S2. However, the highest density of Class Gastropoda was recorded ( $11.1 \% \text{ ind/m}^2$ ) at S4 and the lowest density ( $2.66 \% \text{ ind/m}^2$ ) was recorded at S1. The highest density of Odonata was  $28.1 \% \text{ ind/m}^2$  at the S1 and the density of Plecoptera, which is one of the most sensitive groups, was zero at S4.

The highest value of Shannon-Wiener diversity index of the macro-benthos was  $3.54 \pm 0.14$  at the S1 and the lowest value was  $2.91 \pm 0.33$  at S4. However, there was no significant difference of Shannon-Wiener index at the four sampling sites.

Also, there were some significant correlations between the physico-chemical parameters and density of the macro-benthos. Table 3 gives the Pearson's correlation and the related parameters. The  $\text{NO}_3^-$  shows significant negative correlations with the abundance of orders of Ephemeroptera, Plecoptera and Odonata.

MDS (Multi-Dimensional Scaling) shows the variations of the diversity at S1, S2, S3 and S4 (Fig. 6). Arrows are indicating the degree of variations of Shannon-Wiener diversity at S<sub>2</sub>, S<sub>3</sub> and S<sub>4</sub> sites compared to the reference site (S1). The highest variation can be observed between S1 and S4.

**Table 2** Orders and Families of macro-invertebrates found in Wathurawa stream

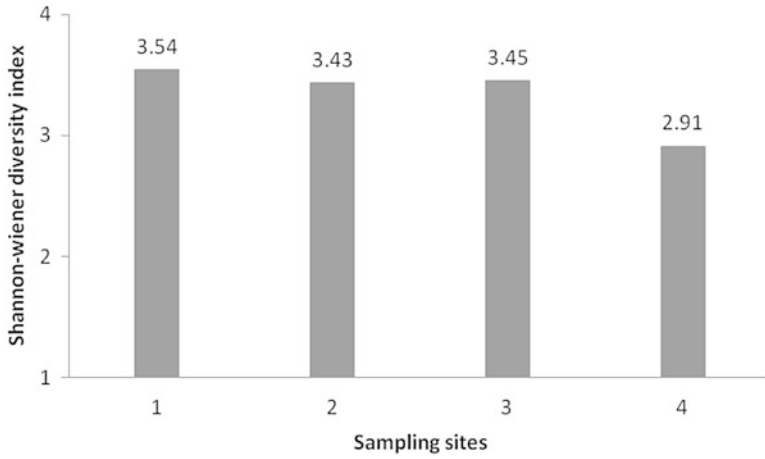
<i>Order</i>	<i>Family</i>	<i>Number of individuals at sampling sites</i>			
			<i>S1</i>	<i>S2</i>	<i>S3</i>
Plecoptera	Nemouridae	11	0	0	0
	Perlidae	80	47	10	0
Odonata	Clorocyphidae	2	2	0	0
	Calopterygidae	2	2	2	0
	Euphaeidae	46	43	15	0
	Lestida	2	0	2	22
	Libellulidae	5	5	0	0
	Platistictidae	311	162	91	0
	Gomphidae	33	29	22	79
	Cordulidae	0	2	0	12
	Tricoptera	Helicopsychidae	87	32	140
	Hydropsychidae	329	159	64	22
	Polycentropodidae	21	0	2	88
	Leptoceridae	50	8	61	4
	Limnephilidae	8	0	2	95
	Lepidostomatidae	14	2	0	11
	Glossosomatidae	0	9	0	0
	Odontoceridae	9	2	0	2
Ephemeroptera	Leptophlebiidae	117	109	67	12
	Ephemeridae	9	7	14	56
	Caenidae	15	9	4	14
	Baetidae	8	11	15	2
Diptera	Tipulidae	52	40	24	10
	Simuliidae	46	4	8	22
	Athericidae	4	0	0	0
	Chironomidae	4	0	0	0
Amphipoda	Atyidae	4	51	38	0
	Potamonidae	0	4	15	4
Coleoptera	Scirtidae	23	20	20	0
	Hydrophilidae	2	11	0	15
	Helophoridae	7	19	0	5
	Psephenidae	20	20	17	20
	Elmidae	34	58	41	45
Class Oligocheata		20	4	8	7
Gordea	Gordiidae	0	0	0	2
Class Gastropoda	Thiaridae	34	18	44	7
	Pilidae	4	15	19	38
	Planorbidae	0	0	2	26
Lepidoptera	Pyralidae	4	0	4	0
	Tortricidae	2	0	2	4
Class Turballaria		0	2	0	0

(continued)

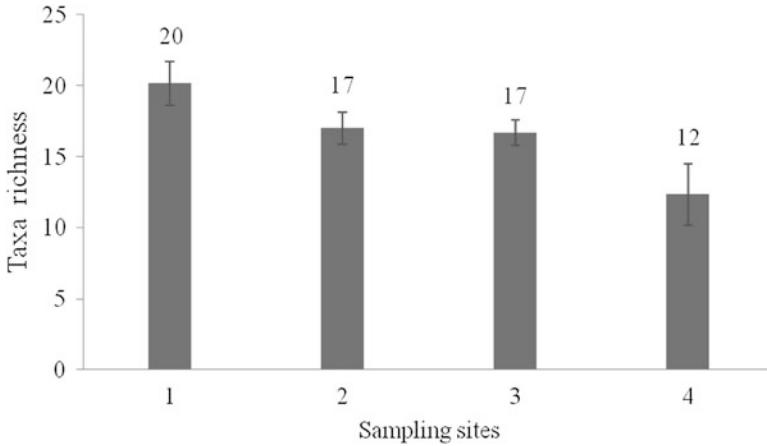


**Table 2** (continued)

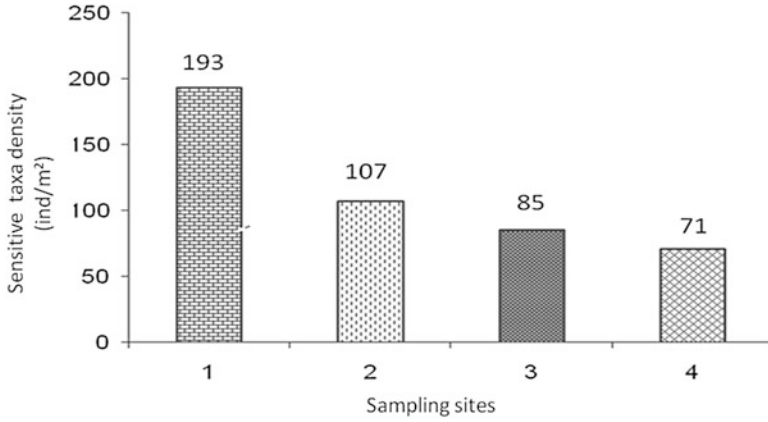
Order	Family	Number of individuals at sampling sites			
		S1	S2	S3	S4
Hemiptera	Naucaridae	7	17	17	2
	Veliidae	0	2	0	6
Total		1426	925	770	639



**Fig. 3** Mean Shannon-Wiener diversity index values at the four sampling sites in Wathurawa stream



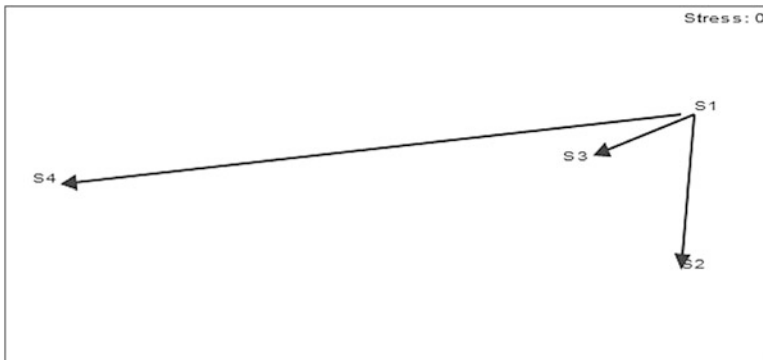
**Fig. 4** Taxa richness at the four sampling sites in Wathurawa stream



**Fig. 5** Sensitive taxa density at the four sampling sites in Wathurawa stream

**Table 3** Pearson’s correlation values between the physico-chemical parameters and the density of macro-benthos

Order	TSS	PO <sub>4</sub> <sup>3-</sup>	TDS	NO <sub>3</sub> <sup>-</sup>
Gordea	0.989	0.989	0.989	
Hemiptera	–	–	0.918	–
Ephemeroptera	–	–	–	–0.96
Plecoptera	–	–	–	–0.96
Odonata	–	–	–	–0.96
Class Gastropoda	–	–	–	0.934



**Fig. 6** The variations of Shannon-Wiener diversity index at the sampling sites in Wathurawa stream (MDS stress = 0.00)

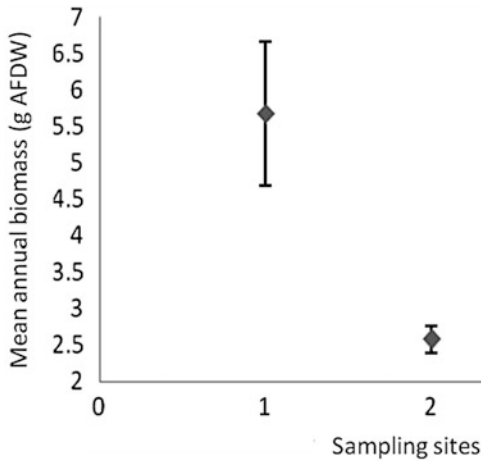


Fig. 7 Mean annual biomass at the four sampling sites

### ***Macro-benthos Secondary Productivity***

The highest secondary productivity ( $8.43 \text{ g AFDW m}^{-2} \text{ yr}^{-1}$ ) was obtained at S1 while the lowest secondary productivity ( $3.95 \text{ g AFDW m}^{-2} \text{ yr}^{-1}$ ) was found at S4. The mean annual biomass of macro-benthos at the four sites is shown in Fig. 7.

## **Discussion and Conclusions**

The cluster separation of site 3 and site 4 according to all physico-chemical characteristics explains the effects of agricultural runoff over the tea and paddy cultivations. Even though, the spatial variation of water quality parameters are not significant, the highest values were observed at the site 3 and site 4 with the effects of agricultural runoff. However, long-term environmental changes can be predicted by assessing the community structure of macro-benthos (Sharma et al., 2009). There were well published evidences for correlations between water quality and abundance of macro-benthos such as Thani and Phalaraksh (2008) who observed that the Plecoptera families except Nemouridae and Perlidae were low in tropical countries due to comparatively higher temperature. Further, Sharma et al. (2009) have observed a negative correlation between Plecoptera and mean water temperature in Tons river, Doon valley, India. Similarly, significant negative correlations were observed for Plecoptera, Odonata and Diptera with the mean water temperature in the present study.

Total Suspended Solids (TSS) is another considerable factor for determining the community structure of the aquatic ecosystem. Merrigton et al. (2002) have revealed that the erosion events are short lived but the impact can be very persistent.

The highest TSS value was recorded at the site 4 ( $36.42 \pm 27.59$  mg/L) where the runoff from both paddy and tea cultivations join the stream. Also, the significant negative correlation between TSS and Shannon-Wiener diversity index shows the importance of TSS on macro-benthos community structure. According to Leek (1995) and Evans (1996), the Ephemeroptera and Plecoptera are severely affected by thin films of fine sediments when there is higher TSS in water. These evidences are supported by the absence of Plecoptera at the site 4 due to higher TSS in the present study.

Particularly,  $\text{NO}_3^-$  is the most prevalent soil mineral in well drained and well aerated soil and it has an ability to accumulate in soil with higher concentrations (Merrigton et al. 2002). However, nitrate concentration in Wathurawa Ela stream was lower than the EU standard limits for drinking (50 mg/L). The strong negative correlations between nitrate concentration and the abundance of sensitive orders explain the complete absence of the most sensitive order of Plecoptera (Stonefly) at the site 4.

Mandawille (2002) has revealed that the Gastropods are highly pollutant tolerance species. In fact a significant positive correlation between Gastropod density and nitrate concentration has been observed by Sharma et al. (2009) in Tons river. Meantime, the present study shows a positive correlation between Gastropod density and nitrate concentration. Phosphate also can be identified as a major component that can cause environmental problems even at a low concentration (20  $\mu\text{g/L}$ ) (Merrigton et al. 2002). The phosphate concentration ranged from 10  $\mu\text{g/L}$  to 30  $\mu\text{g/L}$  and there was a significant negative correlation between phosphate concentration and the Shannon-Wiener diversity index in the present study.

The highest mean diversity index value ( $3.54 \pm 0.14$ ) was found at the site 1 which is located near the forest margin considered as the reference site and it is the lowest disturbed site by anthropogenic activities. The lowest diversity was recorded at the site 4, due to the runoff from both paddy and tea cultivations. According to Sharma et al. (2009), the higher diversity index indicates good habitat quality while the less diversity indicates degraded habitat quality. However, Shannon-Wiener diversity index does not imply any information about the sensitivity of the organisms to the pollution level. Therefore, no significant differences of S-W index among the four sampling sites were observed in this study. Particularly the Stoneflies (Plecoptera), which is the most sensitive taxonomic group among macro-benthos, were absent at the site 4 and the site has been represented by higher tolerant and medium tolerant group of Ephemeroptera (May fly) Families. EPT index was significantly different between site 1 and site 4 which explains the variations of habitat quality with the effects of agricultural runoff.

Munari and Mistri (2007) have stated that higher annual secondary productivity is a better indicator for good quality environmental condition. Therefore, it has been confirmed the effects of agricultural runoff on the reduction of secondary production at the site 4 in present study. According to Clarke (2008), there is still an argument, whether the headwater streams are taxonomically rich or not. Even though, there were effects of agricultural runoff on degradation of the habitat quality and productivity, the present study on the headwater stream has been able

to identify 41 families of macro-benthos at the four sampling sites during the rainy period. However, there should be a systematic approach to mitigate the effects of agricultural runoff not only in headwater streams but also for the entire environment. An interesting finding of Song (2009) was the remaining of a grass or woody vegetation border width of 10–20 m to mitigate effectively the effects of agricultural runoff on sensitive and important habitats.

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