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Are the stream macrobenthos impacted by the wastewater from rubber factories?

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Abstract We assessed how the wastewater generated from raw rubber factories affected the water quality parameters and how such changes influenced the stream macrobenthic assemblages in some streams in the wet zone of Sri Lanka. For this assessment, water quality parameters *viz.* DO, COD, BOD_5, conductivity, TDS, T, pH, and OMC in the sediment were measured, and the macrobenthic fauna were sampled during the dry season at six sampling sites established based on judgemental sampling technique *viz.* rubber factory wastewater effluent canal (site A), point of wastewater discharge in the stream (site B), 50 m upstream site from site B (site C), 50 m downstream site from site B (site D), 100 m downstream site from site B (site E), and 150 m downstream site from site B (site F) following standard field sampling techniques. Secondary research data (dry season) from two other streams subjected to rubber factory wastewater effluents, namely Rakwatte Ela (2001) and Gurugoda Oya (2011), were also used for this assessment. Results revealed that the COD, BOD_5, OMC, conductivity, and TDS levels were elevated, and the DO level was reduced significantly in the highly polluted A and B sites in all three streams. Parallel to them, the macrobenthic diversity decreased significantly ($p<0.5$; ANOVA) at these two sites. However, the complete opposite was observed at the furthermost downstream site F, where the water quality parameters and the microbenthic composition became almost the same as that in the upstream control site C. It is evident that the changes made to the stream water quality and the macrobenthic assemblages by the rubber factory wastewater are never permanent and disappear within a relatively short stretch of 150 m along the streams, most probably due to the dilution of wastewater along the stream. The pollution-tolerant tubificids and chironomids were bioindicator candidates to detect such changes where they became highly abundant at the highly polluted wastewater effluent canal (i.e., Site A) and point of wastewater discharge in the stream (i.e. Site B), but their abundance gradually decreased along the downstream sites probably due to wastewater dilution by the fresh water supply from the stream.

Keywords: chironomids, macrobenthos, point-source pollution, rubber factory effluents, tubificids

INTRODUCTION

Wastewater generated from natural rubber processing has been identified as a key polluter of inland waterways (Das, Saha & Bhattacharjee, 2016; Omoigberale et al. 2021) since it contains uncoagulated rubber latex, latex serum substances, and chemicals added to facilitate latex coagulation (Mohammadi *et al.* 2010; Edirisinghe,2013, 2014). This wastewater has a high affinity for changing the water quality and the inhabiting faunal assemblages in the natural waterways that receive them (Arimoro 2009; Idris et al. 2013; Edirisinghe 2014; Pillai & *Girish 2014;* Omoigberale *et al.* 2021*).* Some of these water quality changes include eutrophication and bioaccumulation of heavy metals in the sediment (Arimoro 2009, 2011; Pillai & Girish 2014; Omoigberale *et al.* 2021), while the biological changes include increasing or decreasing the abundance of macrobenthic fauna through physiological and morphological changes. For example, Arimoro (2009, 2011) observed head capsule deformities in the benthic chironomid *Chironomus transvaalensis* in a stream in Niger Delta in southern Nigeria contaminated with wastewater from a raw rubber processing facility. Such individual changes may eventually reflect major changes in the macrobenthic community structure in such water bodies.

Freshwater benthic macroinvertebrates are widely used as bioindicators in assessing ecosystem health (Madhushankha, Asanthi & Maithreepala 2014; Arimoro, Meme & Keke 2021; Wang *et al.*

2021). The pollution-intolerant sensitive species decline as a result of pollution distress, while pollution-tolerant taxa dominate the macrobenthic community (Zhang *et al.* 2014; Belal, El-Sawy & Dar 2017; Wijeyaratne & Liyanage 2021) upon receiving the pollutant containing wastewater.

Sri Lanka is one of the leading natural rubber manufacturing countries in the world. However, it has often been reported that more than half of the countries' raw rubber processing facilities do not adhere to the recommended wastewater treatment practices so that the extent of pollutants exceeds the National Water Pollution Control Standards set by the Central Environmental Authority (CEA) of Sri Lanka (Edirisinghe 2013; Edirisinghe 2014). For example, Edirisinghe (2013) recorded that the BOD_5, COD, and TSS levels of 62 rubber processing plants were 74 mgL^{-1}, 58 mgL^{-1}, and 62 mgL^{-1}, respectively, and are well above the CEA standards. Thus, natural rubber processing has been highlighted as a key surface water pollution source in the country despite the foreign exchange it earns by exporting the processed rubber. Although some information on the water quality changes that occurred in the stream water is available (e.g., Edirisinghe 2013; Edirisinghe 2014), there is a dearth of information to what extent these wastewaters affect the stream macrobenthic assemblages and how the pollution indicator species in these benthic assemblages can be used to assess such changes. Therefore, the current study was carried out to determine how the wastewater generated from raw rubber processing affects the water quality and the macrobenthic assemblage inhabiting three wet zone streams in Sri Lanka, namely Aswathu Oya, Gurugoda Oya, and Rakwatte Ela, all of which are found in association with rubber processing facilities. It also aimed to identify if there are any biological indicators, specifically benthic indicator species, to assess such changes using primary and secondary research data.

METHODOLOGY

Field sampling for primary data was conducted at the Aswathu Oya, a stream that runs through the Seethawaka Divisional Secretariat Division (DSD) in the Colombo district of Sri Lanka. The area including the Seethawaka DSD experiences a distinct dry season (December-March) and a wet season (May-September) with heavy spells of rain annually. The stream water is relatively clean and clear, so villagers use it for their daily needs. This stream flows bordering a large rubber plantation with an onsite large-scale raw rubber processing factory. The factory wastewater is released into a narrow-excavated canal (approximately 1 km) that eventually opens to the Aswathu Oya. Although the effluent water is treated onsite in a wastewater treatment facility before being released into the canal, closer observations revealed that the treated water in the effluent canal is still quite grey and emits an obnoxious pungent odour.

Altogether, six sites were established for sampling, with the first site (site A) being in the wastewater canal and the other five sites (sites B-F) along the Aswathu Oya stream. The location, site specifics, and general appearance of these six sites are given in Figure 1, Table 1, and Plate 1, respectively.

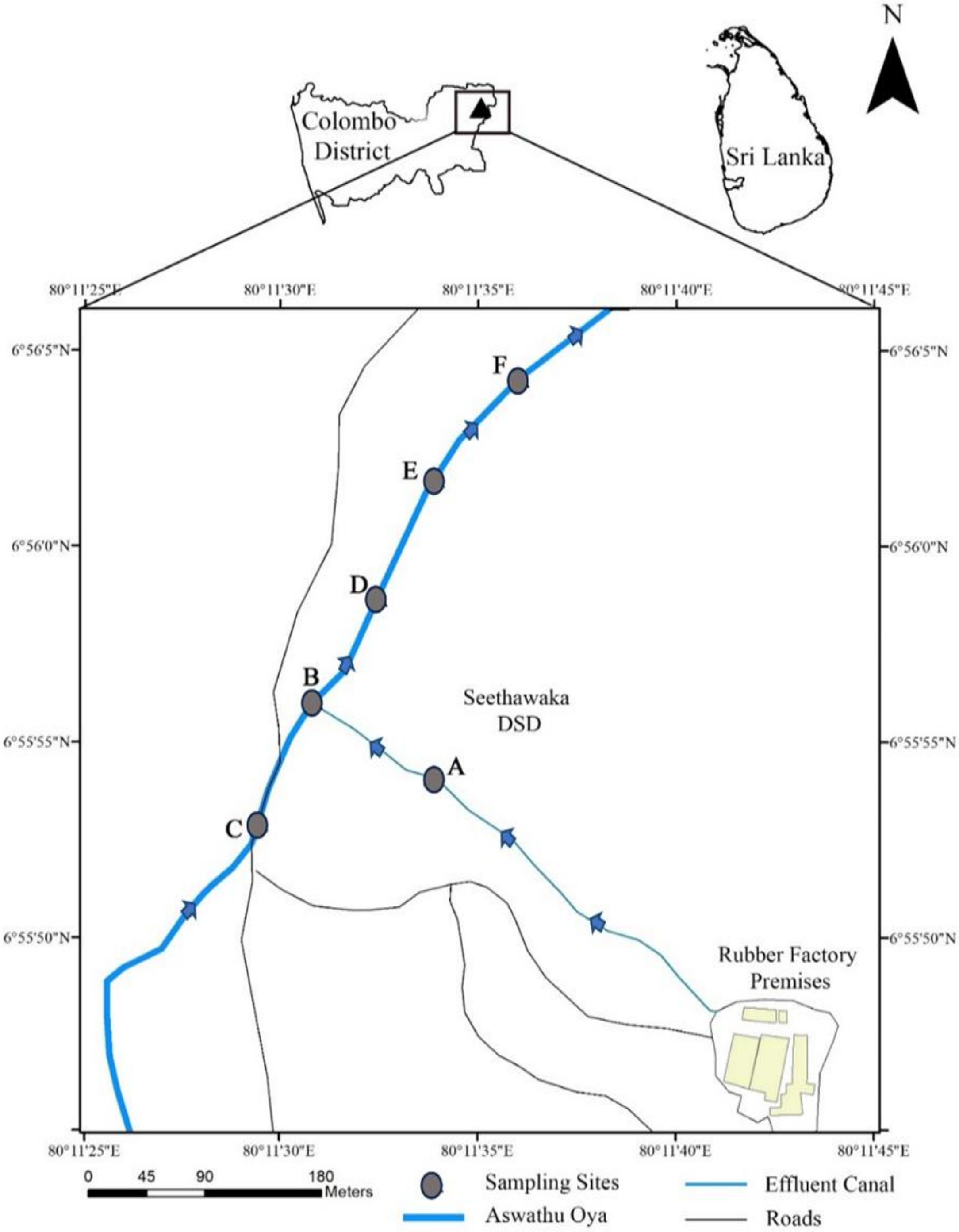

Fig 1 The map showing the location of the raw rubber factory, the effluent canal and the six sampling sites *viz*. A-F at Aswathu Oya stream. The sampling site A was established in the wastewater canal, while sites B-F were established 50 m away from each other along the Aswathu Oya stream. The upstream site C was considered as the control reference site since it does not receive any rubber factory wastewater. The blue arrow represents the flow direction. Site specifications and the general appearance of each sampling location are given respectively in Table 1 and Plate 1.

Table 1 Site-specific descriptions of the six sampling sites in Aswathu Oya.

Sampling site	Description
A	The depth and width are about 0.5 m and 1 m, respectively. The bottom is mainly clayish with fine gravel and sand and contains plant debris. The canal banks are margined with a dense growth of *Panicum* spp. and *Colocasia* spp.; its water emits an obnoxious pungent odour.
B	Shallower but broader than the effluent canal, where the depth and width are about 0.35 m and 5 m, respectively. The bottom is mostly sandy, and the banks are margined with *Panicum* spp. and *Alocasia* spp.
C	The depth and width are about 0.3 m and 3 m, respectively. The bottom is mostly sand and gravel with generally clear water. *Panicum* spp. are seen along the banks, while aquatic macrophytes, such as *Imperata* spp., are seen in the water column. Various freshwater fish species, including *Glossogobius giuris* and *Pethia nigrofasciata*, were abundant in the water column.
D	The depth and width are about 0.5 m and 5 m, respectively. The bottom contained a thick layer of plant litter. Both banks are covered with *Panicum* spp., *Alocasia* spp., large *Caryota urens*, and *Artocarpus nobilis* trees.
E	The depth and width were about 1.5 m and 4.5 m, respectively. The bottom is clayish with sand and gravel. Both banks are covered with *Bambusa* spp., *Panicum* spp., and *Eichhornia* spp. etc.
F	The depth and width are about 1.4 m and 4 m, respectively. The bottom is muddy with a thick layer of plant litter. The water appeared to be clean. Both banks are densely margined with *Panicum* spp., *Alocasia* spp., and *Eichhornia* spp. giving shade to the site. Various freshwater fish species, including *Glossogobius giuris* and *Pethia nigrofasciata*, were abundant in the water column.

(a). Site A

(b). Site B

(c). Site C

(d). Site D

(e). Site E

(f). Site F

Plate 1 General appearance of the six sampling sites A-F. Site A is in the wastewater canal, while the remaining five sites, B-F, are along the Aswathu Oya stream.

Field sampling was conducted during the dry season from December 2021 to January 2022. The temperature, total dissolved solids (TDS), pH, conductivity, and dissolved oxygen (DO) of the overlying water at random locations in each sampling site were measured in *in-situ* (n=4 each) using a multi-probe water quality checker (HACH/Model: Hq40d). Four water samples each for the determination of chemical oxygen demand (COD) and 5-day biological oxygen demand (BOD_5) were collected from near the bottom into amber-coloured glass stoppered bottles (250 ml each) ensuring no air bubbles were trapped. The water in the four COD bottles was preserved *in-situ* by adding concentrated H_2SO_4 (0.1 mL each), but the other four were left intact. Four sediment core samples (6 cm diameter × 10 cm deep each) were collected from random locations using a soil core sampler, wet sieved *in situ* through a 0.5 mm sieve separately, stored the residues retained on the sieve in labelled polythene bags, and preserved using a solution of Rose Bengal containing 5% formaldehyde to determine the macrobenthic fauna. Four other sediment samples were collected from the bottom surface to determine the soil organic matter content (OMC). This procedure was repeated for all the six sampling sites.

The unprocessed and processed water and sediment samples were transported to the laboratory at the University of Kelaniya. At the laboratory, the BOD_5 and the COD in water samples were determined (Rice *et al.* 2012). The OMC content was determined by the dry combustion method. The macrobenthos in the sieved samples were carefully sorted out from the residues and identified under the Binocular Stereo Zoom Microscope (×40) (Model: MEIJIEMZ-5) to the nearest possible taxonomic category using the standard taxonomic keys by Fauchald (1977) and, Fernando & Weerawardhena (2002) and, enumerated separately. The whole sampling procedure was repeated during the same dry season.

Unpublished research data by Hewawasam (2001) and Ranathunge (2011) were used as secondary data sources. Hewawasam (2001) studied the effects of industrial wastewater from the Kelani Tyre Factory on the water quality and macrobenthic community in Rakwatte Ela in Kelaniya, while Ranathunge (2011) studied the effects of raw rubber factory wastewater on the water quality and macrobenthic community in Gurugoda Oya in Ruwanwella, Both sites are in the wet zone of the country, and the research was carried out during the dry season, and followed similar field sampling protocols and laboratory procedures for collecting macrobenthic abundance data and measuring water quality parameters as described under the Aswathu Oya stream.

DATA ANALYSIS

The percentage abundance of each recorded taxa, total abundance (N), species richness (SR), Shannon-Weiner heterogeneity index (H'), and the Pielous's species evenness index (J') of the macrobenthic community were determined separately for the six sampling sites in each stream. Spatial variation of the water quality parameters and the most abundant macrobenthic taxa (% abundance>5%) between the six sampling sites in the three streams were analysed using two-way ANOVA after testing for data normality by the Anderson-Darling test. When a significant variation was noted for sites or streams, further analysis was carried out using one-way ANOVA followed by the Tukey post-hoc tests. The similarity of water quality parameters and the abundance of macrobenthic taxa between the six sites in the three streams were analysed separately using cluster analysis, and significant differences between the clusters were tested using one-way ANOSIM. The data were analysed at $\alpha = 0.05$ in MINITAB (Version 17) and Primer (Version 05) software for Windows as appropriate.

RESULTS

Spatial variation of water quality parameters

The water quality parameters varied significantly among the three streams (Table 2) and the six sampling sites ($p<0.05$; two-way ANOVA) (Table 3). Aswathu Oya recorded significantly lower BOD_5, OMC, TDS, conductivity, and significantly higher DO values than the other two streams, while Gurugoda Oya recorded significantly higher COD, BOD_5, pH, TDS, conductivity, and significantly lower DO than the other two streams ($p<0.05$; Tukey post-hoc pairwise tests after one-way ANOVA).

In all three streams, both A and B sites recorded significantly higher levels of COD, BOD_5, OMC, TDS, conductivity, and significantly lower levels of DO than site C (the upstream control site) and site F (the furthest downstream sampling site from the point of wastewater discharge) ($p<0.05$; Tukey post-hoc pairwise tests after one-way ANOVA). However, the aforesaid water quality parameters did not vary significantly among the other three downstream sites D, E and F ($p>0.05$, Tukey post-hoc pairwise tests after one-way ANOVA) except BOD_5, TDS and conductivity. Further, the pH did not vary significantly among the six sampling sites ($p>0.05$; Tukey post-hoc pairwise tests after one-way ANOVA).

Table 2 Summary of the variation and analysis of water quality parameters among the three streams (n=24 for each stream).

Water quality parameter	Gurugoda Oya	Rakwatte Ela	Aswathu Oya
Water temperature (°C)	28.40 ± 0.18^{b} (26.90-29.50)	29.66 ± 0.45^{a} (27.70-34.60)	27.29 ± 0.12^{c} (26.50-28.30)
COD (mgL^{-1})	13.79 ± 0.83^{a} (10.01-20.00)	5.77 ± 0.28^{b} (1.91-6.91)	6.03 ± 0.93^{b} (1.53-16.20)
BOD_5 (mgL^{-1})	10.07 ± 1.01^{a} (4.20-20.40)	0.83 ± 0.09^{b} (0.50-1.80)	0.60 ± 0.08^{c} (0.20-1.30)
pH (25°C)	7.13 ± 0.09^{a} (6.04-7.88)	6.36 ± 0.03^{b} (6.11-6.75)	6.20 ± 0.12^{b} (5.27-7.05)
DO (mgL^{-1})	3.84 ± 0.56^{c} (0.90-7.00)	5.78 ± 0.15^{b} (2.70-7.20)	8.78 ± 0.14^{a} (5.40-10.40)
TDS (mgL^{-1})	64.63 ± 7.96^{a} (19.00-140.00)	43.51 ± 2.13^{b} (38.74-55.00)	21.54 ± 1.12^{c} (14.74-32.00)
Conductivity (μS cm^{-1})	130 ± 11.95^{a} (40.00-250.02)	88.05 ± 3.52^{b} (79.52-110.23)	44.32 ± 1.87^{c} (30.32-60.21)
Sediment OMC (%)	2.56 ± 0.22^{a} (1.01-4.73)	2.83 ± 0.09^{a} (2.20-3.60)	1.62 ± 0.15^{c} (1.14-3.99)

Note: Values are mean ± SE, range in parenthesis. Different superscript letters in a row show significant differences ($p<0.05$) as analysed firstly by two-way ANOVA and by Tukey post hoc pairwise tests after one-way ANOVA.

Table 3 Summary of the variation and analysis of water quality parameters between the six study sites in the three streams (n=12 for each site).

Water quality Parameter	**Site A**	**Site B**	**Site C**	**Site D**	**Site E**	**Site F**
Water temperature (°C)	30.03 ± 0.92^{a} (26.90-34.60)	28.33 ± 0.30^{b} (27.00-30.20)	27.74 ± 0.23^{b} (26.50-29.00)	28.19 ± 0.31^{b} (26.80-29.70)	28.18 ± 0.28^{b} (26.70-29.50)	28.26 ± 0.10^{b} (26.60-29.50)
COD (mgL^{-1})	11.61 ± 1.82^{a} (4.91-20.00)	11.71 ± 1.71^{a} (5.42-19.79)	5.77 ± 0.99^{b} (1.80-10.06)	$7.51 \pm 1.02^{a, b}$ (3.09-12.91)	$6.86 \pm 1.02^{a, b}$ (2.95-12.01)	5.84 ± 1.06^{b} (1.53-11.01)
BOD$_5$ (mgL-^{1})	6.85 ± 2.32^{a} (1.20-20.40)	5.08 ± 1.86^{b} (0.60-15.80)	2.20 ± 0.80^{d} (0.20-7.30)	$4.02 \pm 1.53^{b, c}$ (0.50-15.30)	$2.48 \pm 0.87^{c, d}$ (0.40-7.90)	2.40 ± 0.86^{d} (0.20-7.80)
pH (25°C)	6.57 ± 0.21^{a} (5.27-7.66)	6.52 ± 0.18^{a} (5.45-7.55)	6.59 ± 0.17^{a} (6.00-7.65)	6.58 ± 0.22^{a} (5.40-7.88)	6.51 ± 0.15^{a} (5.72-7.30)	6.62 ± 0.12^{a} (6.00-7.23)
DO (mgL^{-1})	3.02 ± 0.52^{b} (1.00-5.60)	3.16 ± 0.70^{b} (0.90-6.60)	5.44 ± 0.73^{a} (1.80-7.60)	4.93 ± 0.72^{a} (1.80-8.00)	5.28 ± 0.77^{a} (1.70-8.00)	5.47 ± 0.98^{a} (1.70-10.40)
TDS (mgL^{-1})	55.5 ± 13.02^{a} (22.00-140)	$48.9 \pm 7.50^{a,b}$ (25.0-120.0)	16.82 ± 2.13^{d} (14.74-40.00)	$32.67 \pm 7.13^{b,c}$ (19.00-90.00)	$26.00 \pm 4.57^{b,c}$ (17.00-70.00)	$19.54 \pm 1.61^{c,d}$ (15.00-32.00)
Conductivity (µS cm^{-1})	110.42 ± 15.35^{a} (55.32-250.02)	$98.37 \pm 12.9^{a,b}$ (55.05-220.00)	33.45 ± 4.83^{d} (28.03-78.35)	$65.75 \pm 12.9^{b,c}$ (40.34-175.6)	$55.45 \pm 7.58^{b,c}$ (36.35-135.0)	$38.9 \pm 1.89^{c,d}$ (30.32-64.98)
Sediment OMC (%)	3.61 ± 0.19^{a} (2.54-4.73)	3.48 ± 0.22^{a} (2.38-4.30)	1.95 ± 0.25^{b} (1.01-3.10)	2.05 ± 0.22^{b} (1.28-3.12)	1.87 ± 0.19^{b} (1.20-2.75)	2.06 ± 0.25^{b} (1.16-3.84)

Note: Values are mean ± SE, range in parenthesis. Different superscript letters in a row show significant differences ($p<0.05$) as analysed firstly by two-way ANOVA and by Tukey post hoc pairwise tests after one-way ANOVA.

The cluster analysis further attested the above results, generating three unique clusters, namely 'site A & site B cluster', 'site C & site F cluster', and 'site D & site E cluster' for the three study streams (Figure 2). These three clusters were highly significantly different from each other ($p<0.05$; one-way ANOSIM), indicating similar water quality characteristics between the two sites within each cluster.

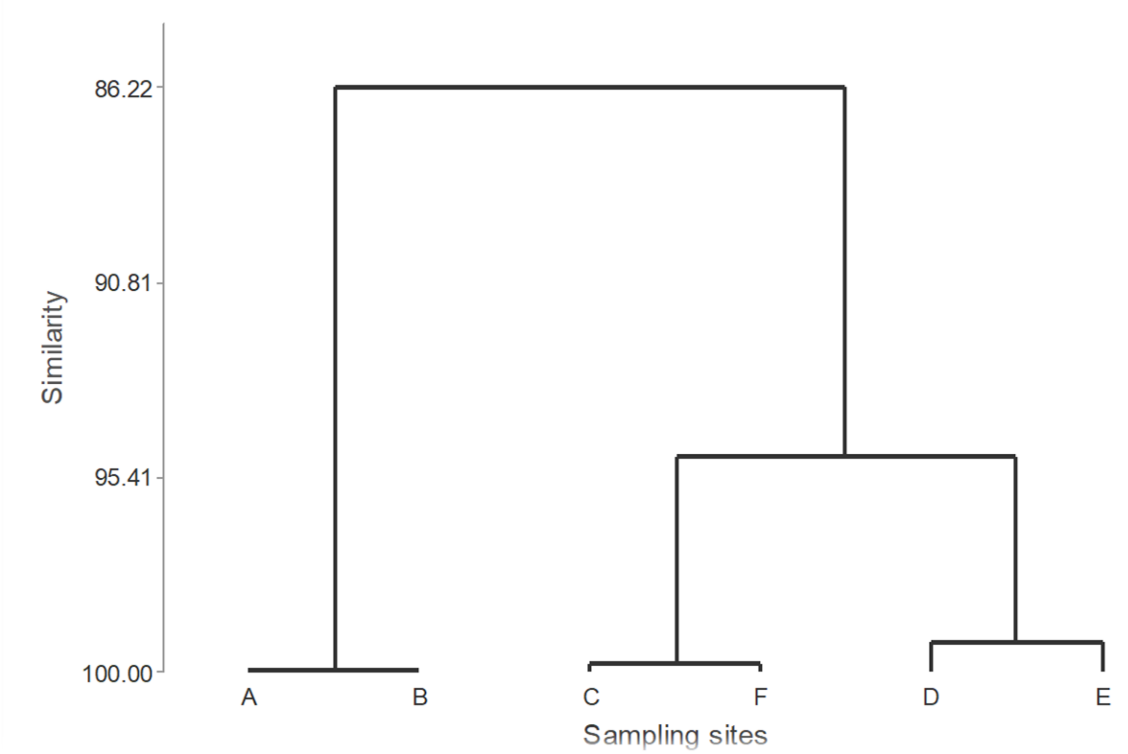

Fig 2 Dendrogram showing the clustering of water quality parameters among the six study sites (n=12 for each site) in the three streams: Aswathu Oya, Rakwatte Ela, and Gurugoda Oya. In all three streams, the water quality parameters in the effluent canal (site A) and the point of discharge (site B), the control site (site C) and the furthest downstream site (site F), and the D and E sites formed three separate clusters at 99.93%, 99.80% and 99.29% similarity levels respectively.

Spatial variation of macrobenthic community diversity

The variation of the macrobenthic community diversity parameters *viz.* SR, H', J' and N among the six sampling sites in the three streams are shown in Table 4. As a general pattern, the highest SR, H' and J' were observed in sites C (the upstream control site) and F (the furthermost downstream site), while the values were the lowest in site A (the wastewater canal). The wastewater discharge point in the stream (site B) recorded intermediate SR, H' and J' values compared to sites C and A.

Table 4 Variation of the macrobenthic community diversity parameters between the six sampling sites (A-F) in the three streams. SR= Species Richness, H'=Shannon-Weiver diversity index, J'=Pielou's evenness index, and N=Total abundance (n=8 for each site in each stream).

Stream	Sampling site	SR	H'	J'	N
Gurugoda Oya	A	2	0.6061	0.5738	455
	B	3	0.6304	0.6745	298
	C	5	1.5600	0.9690	40
	D	5	1.0820	0.6720	226
	E	6	1.2010	0.6703	102
	F	8	1.5480	0.7445	72
Rakwatte Ela	A	10	1.7320	0.7524	74
	B	16	2.1840	0.7626	641
	C	18	2.2040	0.7911	585
	D	15	2.2920	0.8465	596
	E	19	2.4890	0.8452	591
	F	15	2.1930	0.8066	385
Aswathu Oya	A	5	1.5230	0.8502	65
	B	7	1.5970	0.8206	62
	C	8	2.1500	0.9336	22
	D	6	1.8200	0.8284	58
	E	6	2.0430	0.8874	36
	F	8	2.0830	0.9044	27

Spatial variation of abundance of macrobenthic taxa

Of the macrobenthic taxa recorded, tubificids and chironomids dominated the macrobenthic community. These tubificids and chironomids contributed 64% and 26%, 14% and 9%, and 17% and 37% to the microbenthic assemblage at the Gurugoda Oya, Rakwatthe Ela and Aswathu Oya respectively (Figure 3). The abundance of these two taxa varied significantly among the three streams, with Gurugoda Oya recording their highest abundance while Aswathu Oya recording the lowest abundance ($p<0.05$; Tukey post hoc pairwise tests after one-way ANOVA) (Figure 4).

In all three streams, the highest abundance of both tubificids and chironomids was recorded in the wastewater canal (i.e., site A), while their abundance was the lowest at the control site (site C) as well as the furthest downstream site (site F) ($p<0.05$; Tukey pairwise test after one-way ANOVA) (Figure 5).

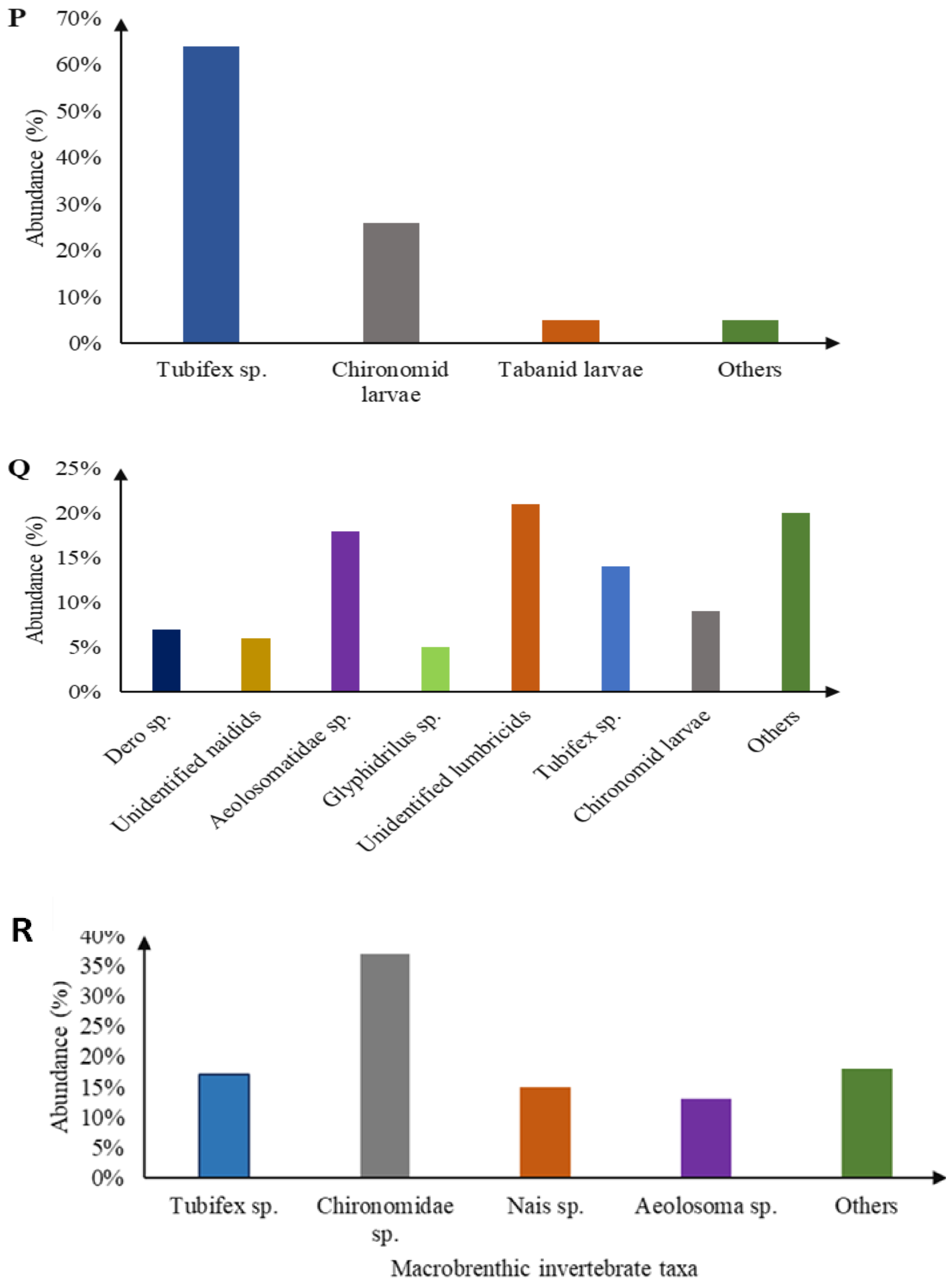

Fig 3 Composition of macrobenthic community in Gurugoda Oya (P), Rakwatte Ela (Q) and Aswathu Oya (R). The tubificids and chironomids dominated the macrobenthic community in the three study streams. Together, these two taxa contributed to 90%, 23% and 54% of the macrobenthic community in Gurugoda Oya, Rakwatte Ela and Aswathu Oya, respectively.

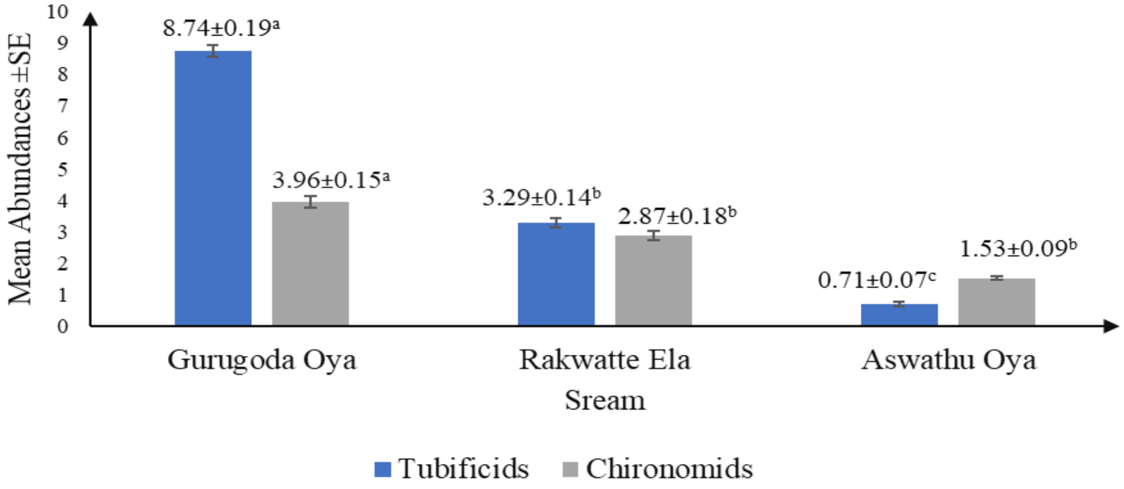

Fig 4 Spatial variation in mean abundance of tubificids and chironomids among the three study streams. Mean abundance ± SE are presented (n=48 for each stream). Different superscript letters indicate significant differences between the three streams ($p<0.05$, Tukey pairwise test after one-way ANOVA).

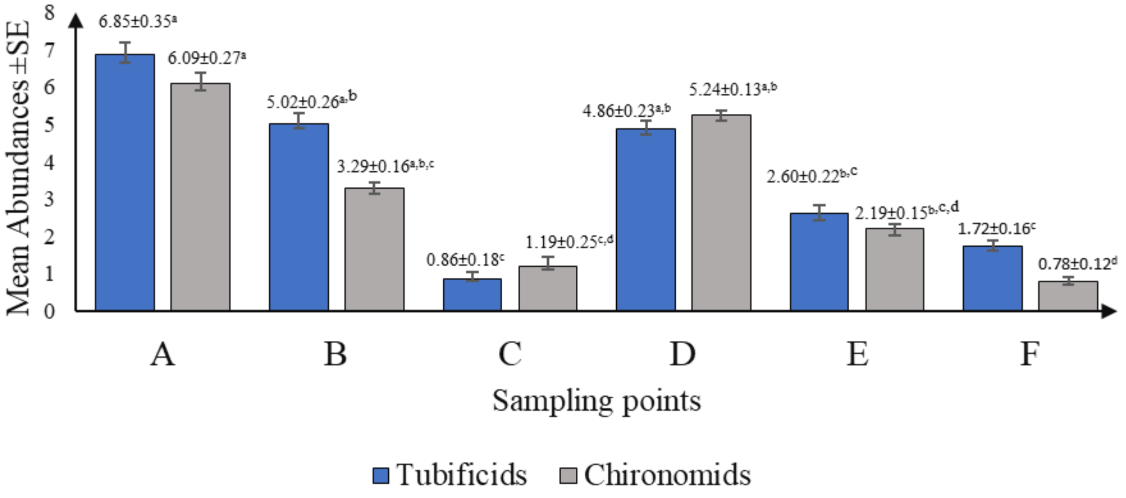

Fig 5 Spatial variation of tubificids and chironomids among the six sampling sites A-F. Different superscript letters indicate significant differences among the three streams ($p<0.05$, Tukey pairwise test after two-way ANOVA). Mean abundance ± SE are presented (n=24 for each site).

The cluster analysis further attested to the above results. The analysis separated the wastewater canal site (site A) from the other five sampling sites (sites B, C, D, E, and F) in the three streams. Of these five stream sites, the total abundance of macrobenthos in the upstream control site (site C) and the furthest downstream site (site F) were more similar to each other and clustered together at a 99.97% similarity level (Figure 6). The remaining two sites (sites D and E) were also clustered with this C & F cluster around a 95% similarity level. However, the macrobenthos in the effluent discharge point in the stream (site B) separated from the main cluster at around 84%

similarity level. Macrobenthos in the effluent canal (site A) separated from the above main cluster at a 44% similarity level, indicating that the macrobenthic taxa in the canal are unique in terms of their total abundance ($p<0.05$, one-way ANOSIM). This is due to the elevated abundance of both tubificids and chironomids in the wastewater canal in all three streams (Figure 4).

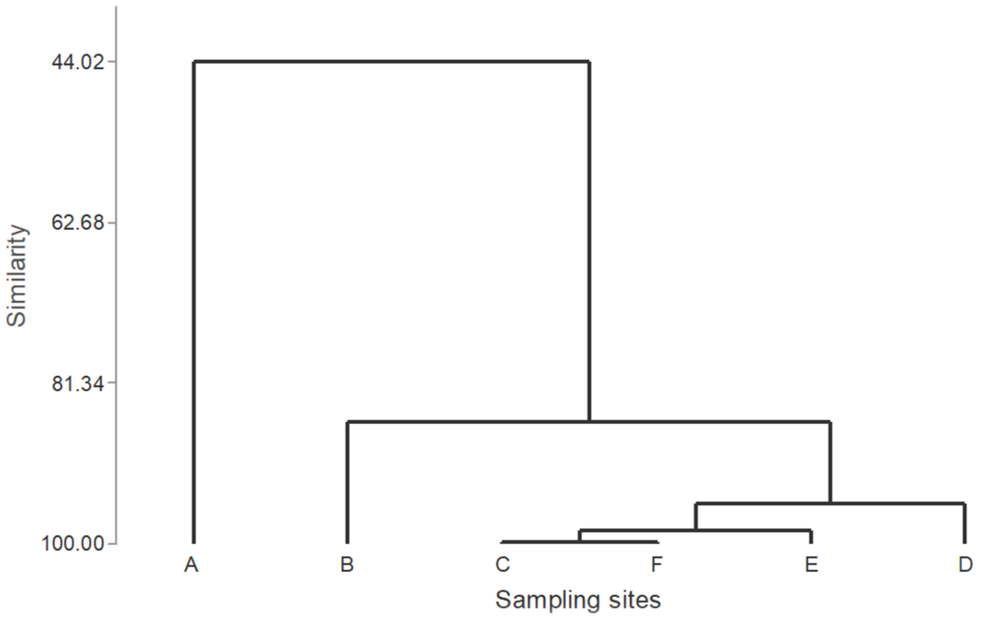

Fig 6 Dendrogram showing the clustering of macrobenthic taxa among the six study sites. Total abundance in each site for all three streams is considered for clustering (n=24 for each site). Site A in the effluent canal separated from the stream sites cluster (B, C, D, E and F) at a 44% similarity level. The sites B, C, D, E, and F clustered at 85% similarity.

DISCUSSION

The present study assessed the impacts of rubber factory wastewater on the water quality and the diversity of macrobenthic assemblages in three streams, *viz.* Aswathu Oya, Rakwatte Ela and Gurugoda Oya in the wet zone of Sri Lanka. Results revealed that the conductivity, COD, BOD_5, OMC, and TDS levels were elevated, and the DO level was reduced both in the wastewater canal (site A) and at the point of wastewater discharge in each stream (site B). Since rubber factory wastewater typically includes high levels of dissolved and suspended solids including organic and inorganic contaminants (Arimoro 2011; Jayashree Gopukumar, Murugan, Praseetha & Shruthi 2018) these two sites are supposed to be the most polluted compared to other sites in each stream. Results also revealed that the above water quality parameters did not vary significantly between the upstream control site (site C) and the furthest downstream site (site F) in all three streams. In addition, it was found that the abundance of the pollutant-tolerant tubificids and chironomids was also significantly high in sites A and B but low in C and F sites.

The increased OMC, COD and BOD_5 levels and decreased DO levels at both A and B study sites could be due to the high oxygen demand for chemical oxidation for the disintegration of pollutants in the factory wastewater aerobically as well as chemically (Adeogun, Chukwuka & Ibor 2011). Similar results have been reported from the studies carried out at Adofi River (Arimoro 2009), Oken River (Omoigberale *et al*. 2021) and Field 20 stream (Efiong and Eze 2004) in Nigeria, where all these three streams received rubber factory effluents. The aesthetic appeal of overlying water is also reduced in the wastewater canal due to changes in its colour, taste, and odour by the rubber factory effluents (Efiong and Eze 2004; Ajinde, Antai & Nosa-Obamwonyi 2017). Such changes were personally observed in the effluent canal during the field sampling at Aswathu Oya.

Contrary to the drastic COD, BOD_5, DO, conductivity and TDS changes in sites A and B, the pH values at all study sites remained almost neutral and slightly varied across all sampling sites. Thus, the pH may not be a significant pollution-determining agent in the three streams subjected to the exposure of rubber factory effluents. Similar results have been observed by Omoigberale *et al.* (2021), where the rubber factory effluent discharges have little impact on the acidity or alkalinity of stream water.

Except for BOD_5, TDS and conductivity, all measured water quality parameters exhibited a modest variation across the D, E and F downstream sites. The BOD_5, TDS and conductivity showed a relatively higher spatial variation among the six sampling sites than the other measured parameters. For example, these three parameters were significantly higher in the effluent canal and at the effluent discharge point in each stream than in the upstream control site C and the furthest downstream site F. They can be identified as the major pollution determinants in rubber factory effluents. Arimoro (2009) also found that BOD_5 level and conductivity are the primary determinants of stream water pollution induced by rubber factory effluents.

These streams also appear to have a remarkable self-purification capacity after receiving effluents from rubber factories. In general, the COD, BOD_5, OMC, TDS and conductivity showed a decreasing trend along the downstream sites from the effluent discharge point in the stream (site B). Contrary to this, the DO showed an increasing trend. Similar observations were made by both Arimoro (2009) and Omoigberale et al. (2021) in their studies. Furthermore, none of the above water quality parameters significantly differed between the upstream control site C and the furthest downstream site F. Although many of the water quality parameters in the stream water are altered upon receiving the rubber factory effluents as in site B (hence depleted the water quality), it is evident that the water quality reacquired the status quo within a stretch of 150 m along all the three streams assessed in this study. This downstream water healing distance could have been less than 150 m during the rainy season when the stream flow is heavy and fast. Therefore, it is recommended that the assessment be conducted during the rainy season to determine this water healing distance.

Parallel to the water quality changes, all three streams showed almost similar variation patterns in species diversity (H’), species richness (SR), and species evenness (J’) of macrobenthic assemblages among the six sampling sites. The lower SR, H’, and J’ were recorded in sites A and B (relative to the reference site C), indicating a relatively low number of specific macrobenthic taxa. However, it was interesting to note that the highest total abundance (N) of macrobenthos was recorded in these two sites. Careful analysis of the macrobenthic species composition in all six study sites in the three streams revealed that the pollutant-tolerant taxa, mainly the tubificids (e.g., *Tubifex, Limnodrilus, Aulodrilus, Bothrioneurum*) and chironomids were predominating in these two sites. However, they were recorded in lesser numbers in the remaining study sites in all three streams.

The elevated abundance of tubificids and chironomids in sites A and B could be attributed to the unfavourable water quality parameters induced by the rubber factory effluents. Drastic reduction in water and sediment quality causes the diminishment of pollution-sensitive macrobenthic taxa with a preponderance to pollution-tolerant taxa (tubificids and chironomids) to flourish under low competition and predation (Višinskiene and Bernotiene 2012; Arimoro et al. 2015; Ibezute, Ibezute & Asibor 2016). It is also possible that water quality-stressed environments such as sites A and B, as observed in this study, disrupt the life cycle, osmoregulation, oxygen uptake ability, reproduction, food supply, and migrations of such sensitive macrobenthic taxa, thus threatening their survival (Lokhande, Singare & Pimple 2011; Shimba, Mkude & Jonah 2018).

It was also interesting to note that the SR, H', and J' increased gradually from site B towards the downstream sites D, E and F as the water and sediment quality improved. Hence, the water and sediment quality changes and the benthic community changes induced by the rubber factory effluents are never permanent and disappear within a relatively short stretch of 150 m down along each stream. This may be due to the dilution of the rubber factory effluents by the large amount of water carried by the stream (Arnon, Avni & Gafny 2015). A much more rapid water quality improvement process could be expected during the rainy season when the water flow in the three streams is heavy and fast during the rainy season. However, this area needs further investigation.

Although the tubificids and chironomids are significantly associated with the rubber factory effluent-impacted sites, the results of the present study revealed that their abundance gradually decreased from site B towards the furthest downstream site F parallel to the improvement of the water and sediment quality along the three streams. Therefore, these two taxa could be considered as bioindicators to detect the impacts posed by rubber factory effluents.

The results showed that, of the three streams assessed, Gurugoda Oya is the most polluted stream, where the water is characterised by the highest COD, BOD_5, pH, TDS, conductivity, and the lowest DO levels as well as increased abundance of tubificids and chironomids of the other two streams. Aswathu Oya, on the other hand, was the least polluted stream, with the lowest BOD_5, pH, OMC, TDS, conductivity, and the highest DO, as well as a relatively low abundance of tubificids and chironomids than the other two streams. According to Ranathunge (2011), the low SR, H', and extremely high abundance of tubificids and chironomids in Gurugoda Oya could be due to the continuous release of partially treated rubber factory effluents from the raw rubber processing facilities in the associated rubber estates.

CONCLUSION

The current study assessed the impacts of rubber factory effluents on the water quality and macrobenthic assemblages in three wet zone streams at three different temporal frames (i.e., 2001, 2011 and 2021/2022). Results revealed that the changes posed by the effluents from raw rubber processing factories to the water quality and the macrobenthic assemblages in streams are always temporary and disappear within a relatively short stretch along the streams. Further, the current study highlights the importance of tubificids and chironomids as pollution indicators to assess such environmental concerns the rubber industry possesses.

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