

Land-Use Practices Affect Water Quality Parameters and Mayfly (Order Ephemeroptera) Assemblage Along River Nzoia (Kenya)

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ABSTRACT

Several river ecosystems are undergoing varied land-use practices, whose monitoring should be continuous. This study evaluated the influence of land-use practices on water quality and macro-invertebrate taxa, specifically the mayfly (order Ephemeroptera) assemblage, along the River Nzoia in Kenya. Four dominant land-use activities were identified as undisturbed, sugarcane growing, settlement, and industrial activities. All the physicochemical water quality parameters displayed significant ($P < 0.05$) spatial variations. Areas with industrial activities had low DO, as well as high BOD, TA, pH and conductivity, settlement and sugarcane growing areas had high levels of phosphates and nitrates. Land use patterns dictated the macro-invertebrate community structure, where sites with low disturbances had high composition, abundance and diversity and were dominated by order Ephemeroptera, Plecoptera, and Trichoptera (EPT). The distribution of mayfly was significant relative to land-use practice ($P < 0.05$), where undisturbed sites followed by industrial sites had the highest occurrence and abundance of mayfly taxa, suggesting the occurrence of more tolerant species of mayfly in sites near industrial areas. Dominance of *Baetis*, and *Caenis* in undisturbed sites and settlement areas, coupled with *Heptagenia* and *Ephemerella* dominance in the sugarcane growing region, but none of the mayfly taxa dominated industrial sites, suggests that they are influenced by anthropogenic activities. PCA plots showed a clear distinction between land-use practices, with ephemeroptera taxa composition being clearly distinguished in the tri-plot. The present study indicates that different types of land-use practices within the study area caused changes in the abundances of the macro-invertebrates and, particularly, mayfly taxa. Thus, all stakeholders should formulate immediate policies that will reduce human impacts on the water quality in River Nzoia. There is also a need to sensitize the local community members to avoid harmful activities along the River Nzoia.

Key words: Macroinvertebrates; Metrics; Human activities; River Nzoia (Kenya), ephemeroptera taxa

INTRODUCTION

The pivotal role of rivers in provisioning of vital ecosystem services, including drinking water, habitat for biotic community, conservation of biodiversity, and attenuation of downstream fluxes of sediments, water, organic carbon, and nutrients, is well known (Erős and Lowe, 2019). Rivers in high altitude areas conventionally occur in catchment areas where land use activities should be minimal. In the contemporary world as well as in the past, the fingerprints of numerous land use activities within proximity of the riverine environments (Thai-Hoang *et al.*, 2022; Ajwang, 2023; Zhang *et al.*, 2023) have created long-lasting legacies of the negative impacts of human activities on land near rivers (Li *et al.*, 2020; Li *et al.*, 2022). These land use activities include agriculture (Edegbene *et al.*, 2020; Comte *et al.*, 2022), urbanization and urban development (Qi *et al.*, 2020; Bi *et al.*, 2023), industrial development (Zhang *et al.*, 2022; Zhu *et al.*, 2022), large-scale water abstraction (Jiang *et al.*, 2022; Zogaris *et al.*, 2023), or a combination of various land use practices (Ren *et al.*, 2022; Islam *et al.*, 2023). By virtue of the strong interconnections between the riverine environment, their ecotones and fluvial ecosystems, land use activities may alter the hydrological regime, damage habitats, as well modify ecological structures and processes within the lotic aquatic ecosystems (Brontowiyono *et al.*, 2022; Yao *et al.*, 2023). Therefore, monitoring of water quality changes at the riverine ecosystems remains a prerequisite for managing human-induced land use changes.

Traditional methods of monitoring the ecological integrity of the riverine ecosystems is usually performed using physical and chemical methods (Farrell-Poe, 2005; Omer, 2019). Monitoring of surface waters using physicochemical parameters provides conditions of the land use practices over a shorter time span (Tanjung and Hamuna, 2019; Ewaid *et al.*, 2020). Hitherto, the unreliability for long-term monitoring of water quality changes remains the drawback of using physical and chemical water quality parameters for monitoring purposes. Moreover, the results of water quality do not account for the impacts of land use activities from non-point sources (Bhatia *et al.*, 2018; Islam *et al.*, 2018). Incorporating suitable biological methods and/or metrics ensures long-term approaches in monitoring of water quality due to land use activities (Simon, 2020; Ustaoglu *et al.*, 2020). In electing appropriate methods, selecting biological organisms that are easy and affordable to collect is more convenient to achieve the best biological monitoring programme (Parmar *et al.*, 2016; Motlagh and Yang, 2019).

Macro-invertebrates assemblage may provide an integrative measure of scientifically defensible evidence of environmental condition (Mzungu *et al.*, 2022) including reflection of land use activities on the water quality (Sripanya *et al.*, 2023). Several quantifiable population attributes that assess macro-invertebrate assemblage, structure, composition, and response have been evaluated, and some have been found to massively correlate with anthropogenic activities (Tampo *et al.*, 2021). Measuring changes in macroinvertebrates' characteristics only at the population level may allow only indirect inference to be made with regard to the cause of the population decline. Assemblages of macroinvertebrates together with metrics of the population attributes are useful surrogates for ecosystem attributes, reflecting on anthropogenic impact (Retnaningdyah *et al.*, 2023; Sripanya *et al.*, 2023). The use of macro-invertebrates in the assessment of water quality in Kenya is historical (Mathooko, 1988; Mathooko and Mavuti, 1992; Ndaruga *et al.*, 2004; Kibichii *et al.*, 2007) and even in recent years (Lubanga *et al.*, 2021; Chamia and Kutuny, 2022; Fekadu *et al.*, 2022). Several quantifiable attributes that assess macro-invertebrate assemblage, structure, composition, and functional response have been evaluated, and some have been found to massively correlate with water quality changes (Tampo *et al.*, 2021).

Aquatic insects have gained prominence in studies aimed at monitoring water quality through biological ways (Prommi and Payakka, 2015; Sitati *et al.*, 2021). Research into this realm has consistently established that aquatic insects belonging to Ephemeroptera (mayfly), Plecoptera (stonefly) and Trichoptera (caddisfly), generally referred to as (EPT), show immense sensitivity to water quality changes (Abong'o *et al.*, 2015; Oruta *et al.*, 2017; Fekadu *et al.*, 2022). Insects belonging to these three groups cannot tolerate degraded water conditions, and thus their high abundance in a particular localized region of a water body depicts high water quality integrity (Mzungu *et al.*, 2022). Among the EPT community of aquatic insect taxa, mayflies (order Ephemeroptera) have shown much higher sensitivity to water quality changes compared to their counterparts in this taxa and thus are currently being given leeway as biological water quality monitors (Maina *et al.*, 2021). Mayflies are good bio-indicators of the freshness in water quality due to their ability to swell in water of good biological quality (Mir *et al.*, 2021). However, land-use practices that introduce changes in water quality may also negatively impact the mayfly abundance and community attributes.

In Kenya, Massive extensions of human settlements, human population growth and increasing industrial activities along the River Nzoia Catchment have been reported (Nyilitya *et al.*, 2020; Kadeka, 2021; Tarus *et al.*, 2022). The intensification of human activities within the catchment of River Nzoia threatens water quality and functional integrity of the river, which occurs in the form of adjustments of biotic abundance and assemblage (Achieng *et al.*, 2021). Fortunately, the changes in biological assemblage can be used to decipher the extent to which humans are affecting the ecological integrity of the river through diverse land-use practices. Although several studies have employed aquatic macroinvertebrates to study changes in water quality along River Nzoia, surprisingly, very few of these studies have directly linked them to land-use practices. Indeed, most of the studies just describe spatial variations in macro-invertebrate assemblage without reference to land-use activities (Aura *et al.*, 2010; Aura *et al.*, 2011; Sitati *et al.*, 2021). There is also a consistent lack of studies on the mayfly assemblage relative to land-use activities. Prolonged deficiency of such data will hamper the protocol for the management and conservation of rivers undergoing differential land-use activities in Kenya.

MATERIALS AND METHODS

Study area land use classification

The study was conducted along the River Nzoia in the larger Nzoia Catchment in Kenya. River Nzoia catchment is located within the Lake Victoria Basin (Figure 3.1), lying at latitudes 1°30'N and 0°05'S, and longitude 34° and 35°45'E. The river is 320 km long with a riparian catchment area of 275 km² (Githui, 2008). It lies at 1134–2700 m above sea level. The rivers flow from the Kakamega Forest, and some of its tributaries pass through the Mt. Elgon region, the Cherangany Hills and the lowland areas of Mumias and Webuye, and finally drain into Lake Victoria. There are several sources of pollutants along the river located in urban areas in Bungoma, Webuye, Mumias, and Webuye. Bungoma is an urban area that has industrial effluents from the municipal region, Mumias and Nzoia discharge effluents from sugar mills, and at Webuye, there is the Pulp and Paper Mills. Herein, espoused is the role played by agricultural chemicals, especially Mumias sugar and pan paper mills, respectively.

The weather pattern within the catchment is typical of the equatorial climate type, where there is sunlight for 12 hours and darkness for 12 hours throughout the year. Temperature varies from a low of 8–12°C to a high of 25–29°C (Othieno Odwori, 2021). The lowest temperatures are recorded in September, and the highest temperatures are in March.

The rainfall in the region of study is bimodal and is fairly high, with a mean of 1200 to 1400 mm per year. Long rains fall between the end of the month of March to the end of April-May, while June to the end of August is a dry period that ushers in short rains in September, and afterwards is a long dry spell from October to the end of March.

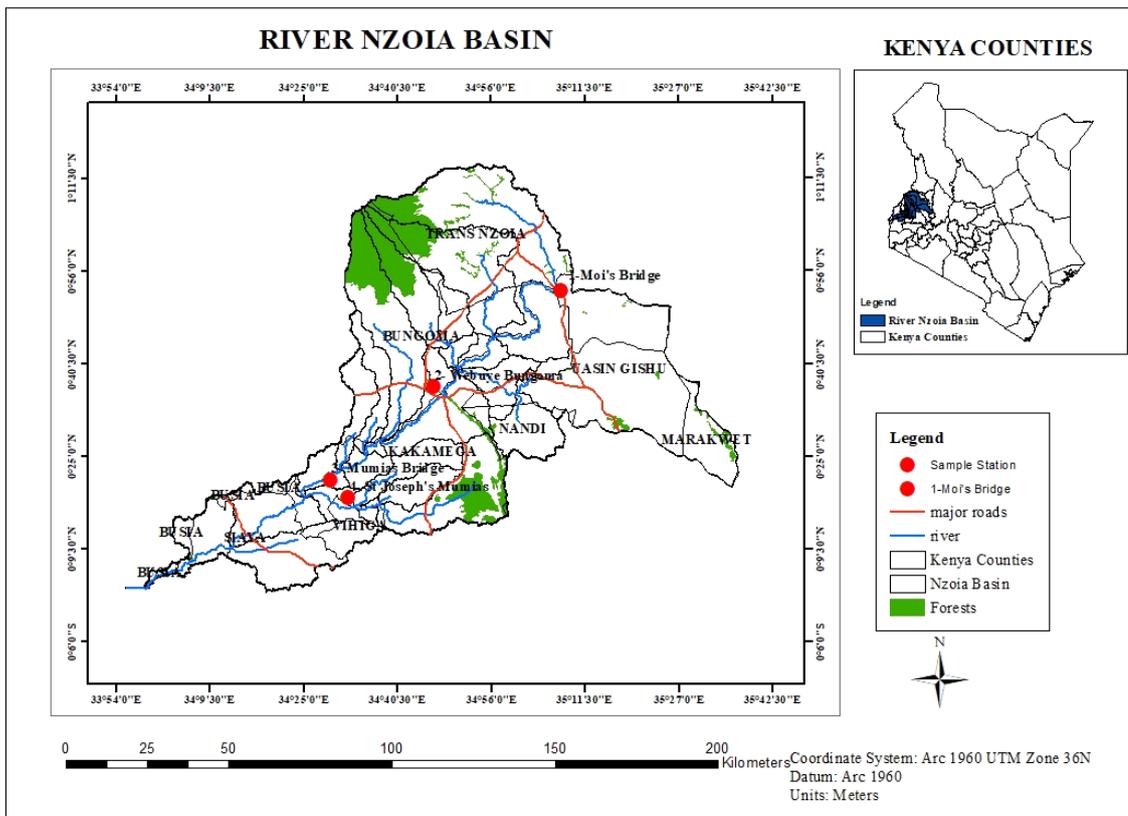


Fig. 1. Map of River Nzoia Basin, the study area and the sampling stations chosen for the study.

Selection of study sites

Based on the categorization of the major land use activities and the delineation of catchments, the study sites were selected. Undisturbed land use was found in the forest at latitude E035.07.272 and longitudinal N00.52.403; sugarcane regions were located at latitude N00.21.005 and E034.29.462, with large-scale sugarcane farming being the dominant activity; and areas near industrial activities were sampled at latitude E34.46.33 and longitudinal N036.036, hereby referred to as industrial regions. On the other hand, settlement (at latitude E034.20.049 and longitude N00.34.29.400) was the dominant land-use activity, with scattered subsistence farms serving as a backdrop. Bathing, subsistence farming, fishing, and sand gathering were notable activities in population areas.

Measuring physico-chemical and hydrological parameters

Five samples for physico-chemical parameters were taken for 8 months in January, March, May, June and August. All samples were measured in triplicate. During measurements of *in-situ* samples, samples for laboratory analysis were also collected in triplicate.

Physico-chemical parameters such as pH, dissolved oxygen (DO), electrical conductivity and total alkalinity were measured *in situ* using the JENWAY® 3405 electrochemical analyzer that had probes for each independent variable. Calibration of the probes was done before any sampling activity. Three measurements were done within the vicinity of the sampling points.

Water was collected in plastic bottles for measurement of BOD₅, nitrate and phosphates. Water samples were drawn from the selected sites in triplicate using labelled plastic bottles. The bottles were stored in cool boxes during transportation from the field to the water quality laboratory. In the laboratory, samples were refrigerated at 4 °C until analyzed using standard methods (Adams, 2017). In all analyses, 100 ml of the sample was used after filtration through Whatman No. 42 filters. BOD₅ was analyzed using the light and dark bottle method (Young *et al.*, 1981). NO₃-N was analyzed through flow injection spectrophotometric, diphenylamine sulphonic acid chromogene method (Asan *et al.*, 2008). PO₄²⁻ was analyzed using the standard ascorbic acid method, reduction of phosphomolybdic acid (Townes, 1986). Due to financial limitations, other parameters such as total nitrogen (TN), total phosphorus (TP), ammonia (NH₄), and total organic carbon (TOC) were not analyzed.

Macro-invertebrate sampling

Over the course of eight months, five sampling occasions were conducted in January, March, May, June, and August to collect macro-invertebrate samples in tandem with physico-chemical parameters. In order to account for the study area's seasonality, the samples were taken during both the dry and wet seasons. Macro invertebrates were collected using Disturbance Removal Sampling Technique (DRST) and a Hess Sampler. The method involve defining a particular sampling area and holding the Hess sampler in water as one disturbs the substrate for approximately three minutes to ensure adequate dislodgement within the designated area and the macrobenthos were then washed down into the Hess sampler. At each site, three samples of Macro invertebrates were collected that is, from the middle and about 1m from the left and right banks. The Hess sampler was used in riffles, shallow sites with rocky substrates while an Eckman grab sampler was utilised in soft sediment. Large debris was then removed from the samples after careful washing the samples through a 500um mesh sieve of the attached organisms, and placed in lidded sample jars. The sample jars were then appropriately labelled, and fixed with 4% formalin (Kage, 2003), right in the field.

Laboratory processing

In the laboratory, the macro-invertebrate samples were filtered and washed free of sediments through a 250um sieve. The samples were spread evenly on a white tray for sorting (Kage, 2003). All the Ephemeroptera were sorted out and identified to the lowest genera level and preserved with 70% Ethanol in well labelled and lidded sample jars. Later the samples were taken to a specialist in Kenyan Museum for further identification of the specific names and confirmation of genera. The unidentified samples were taken to South Africa for further

attention where it was possible. The other samples were preserved in 70% Alcohol and kept in the University of Eldoret Museum.

Assessment of species diversity indices

In calculating the indices, abundance data were obtained by counting macroinvertebrates. Several key attributes were determined:

1. Absence/presence data of taxa of benthic macroinvertebrates at the various sampling sites relative to the human activities by counting the visible macroinvertebrates
2. The abundance of each macroinvertebrate at various sampling sites relative to human activities by quantification of the numbers determined
3. Shannon index (H') of benthic macroinvertebrates at the various sampling sites relative to the human activities using the formula: $H' = \sum_{i=1}^n P_i (\ln P_i)$ (Murphy, 1978)

Where H' = Shannon's diversity index; P_i = the abundance of the i^{th} species expressed as a proportion of total cover; n = number of species

1. Percentage of oligochaetes and chironomids compared to the total abundance of the macroinvertebrates
2. Percentage of Ephemeroptera, Plecoptera and Trichoptera (%EPT) compared to the total abundance of the macroinvertebrates
3. Percentage of mayfly (Ephemeroptera) compared to the total abundance of the macroinvertebrates

Data analysis

The data obtained after sample analysis were analyzed using STATISTICA 6.0 (StaSoft, 2001). All assumptions of the parametric test were achieved through a normality test using the Shapiro-Wilk W statistic (Shapiro and Francia, 1972). Differences in physico-chemical parameters among land-use practices were analyzed using One-Way Analysis of Variance (ANOVA). Significantly different means were discriminated using Duncan's Multiple Range test after ANOVA (Tallarida *et al.*, 1987).

Differences in macroinvertebrate community attributes (abundance, diversity, %OC, %EPT, %mayfly) were analysed using the nonparametric Kruskal–Wallis test, as the data did not meet normality assumptions. Analysis of the variation in the occurrence of macroinvertebrates among land-use practices was done using chi-square (χ^2). All results were declared significant at $P < 0.05$.

Principal Component Analysis (PCA) was used to analyze the spatial relationships between physico-chemical parameters relative to catchment land-use as well as the relationships between mayfly taxa relative to catchment land-use cover and the Interrelationship between land-use practices, water quality parameters and ephemeroptera (mayfly) taxa. This was done through the PCA procedure uses multiple regressions to fit attributes to an ordination space as vectors. The significance of Principal Axis Correlation coefficients was tested using a Monte-Carlo procedure. Discriminant functions of each attribute on land-use were determined through measures of constancy and fidelity. Constancy is the quantity of land-use areas where macroinvertebrate taxa occur, while fidelity is the ability of the macroinvertebrate attribute to predict variations.

RESULTS

Effects of land-use practices on physico-chemical water quality parameters in River Nzoia

A summary of the analyzed physico-chemical properties of riverine water relative to the four human activities along River Nzoia is shown in Table 1. All the analyzed physicochemical water quality parameters demonstrated significant ($P < 0.05$) spatial changes relative to land-use practices. The pH ranged from 3.5 to 8.1 and displayed significant ($F = 19.3223$, $df = 3$, $P = 0.0021$) differences relative to land use. The pH value was highest in sugarcane farms, while the pH of industrial areas was somewhat lower than 4.0.

Table 1. Physico-chemical attributes (mean \pm SEM) of river water with respect to land-use practices along River Nzoia from January to August 2020

	UNDS	INDS	SUGS	SETL	P-value
pH	6.59 \pm 0.72 ^c	3.99 \pm 0.27 ^a	7.85 \pm 0.74 ^d	5.94 \pm 0.73 ^b	0.0021
DO (mg/L)	12.71 \pm 0.00 ^c	3.49 \pm 0.35 ^a	5.40 \pm 0.45 ^b	4.87 \pm 0.44 ^b	0.0005
BOD ₅ (mg/L)	1.24 \pm 0.17 ^a	8.13 \pm 0.51 ^d	2.39 \pm 0.18 ^b	3.86 \pm 0.68 ^c	0.0015
Electrical conductivity (µS/cm)	32.4 \pm 4.9 ^a	341.5 \pm 6.22 ^d	148.8 \pm 25.9 ^c	99.4 \pm 14.9 ^b	0.0076
Total Alkalinity (mg/L)	3.91 \pm 0.48 ^a	19.72 \pm 2.22 ^d	7.94 \pm 1.06 ^c	6.41 \pm 0.78 ^b	0.0124
NO ₃ ⁻ (mg/L)	0.28 \pm 0.26 ^a	0.37 \pm 0.07 ^b	0.71 \pm 0.11 ^c	0.64 \pm 0.12 ^c	0.0035
PO ₄ ²⁻ (mg/L)	0.22 \pm 0.04 ^a	0.24 \pm 0.03 ^a	0.36 \pm 0.06 ^b	0.65 \pm 0.23 ^c	0.0128

¹Means with the same letters as superscripts are not significantly different ($P > 0.05$).

²SE: Standard Error of the mean

³UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

Macro-invertebrate species composition and abundance in Nzoia River

A total of 43 species belonging to 31 genera and 12 orders of macroinvertebrates were observed relative to the land-use activities. The macroinvertebrate genera occurrence in terms of presence/absence along a gradient of human activities along River Nzoia is provided in Table 2. There were significant differences in the occurrence of macroinvertebrates among sites (Chi-square; $\chi^2 = 9.234$, $df = 3$, $P = 0.0263$). The undisturbed sites had the highest occurrence of macroinvertebrate genera (36 genera), which was followed by occurrence in settlement areas (21 genera), sugarcane growing areas (20 genera), while sites adjacent to the industrial areas had the lowest macroinvertebrate occurrence (17 species).

Table 2. Information concerning presence (+)/absence (-) of various genera of benthic macroinvertebrates from January to August 2020

Order	Family	Genus	UNDS	SUGS	SETL	INDS
Pulmonata	Limnaeidae	<i>Lymnaea</i>	+	+	-	+
	Physidae	<i>Physa</i>	+	-	+	+
	Planorbidae	<i>Planorbis</i>	+	-	+	+
Coleoptera	Dytiscidae	<i>Coptotomus</i>	+	-	+	-

		<i>Ilybius</i>	+	+	-	-
		<i>Platambus</i>	-	+	-	-
	Gyrinidae	<i>Gyrinus</i>	+	-	-	-
	Haliplidae	<i>Haliphus</i>	-	-	+	-
Diptera	Anthomyiidae	<i>Limnophora</i>	+	-	-	-
	Chironomidae	<i>Chironomus</i>	+	+	+	-
	Tipulidae	<i>Tipula</i>	+	-	-	+
	Simulidae	<i>Simulium</i>	+	+	-	-
Ephemeroptera	Baetidae	<i>Baetis</i>	+	+	+	+
	Caenidae	<i>Caenis</i>	+	+	+	+
	Ecdyonuridae	<i>Heptagenia</i>	+	+	+	+
	Ephemerallidae	<i>Ephemerella</i>	-	+	-	+
	Leptophtebiidae	<i>Habrophlebia</i>	+	-	-	-
	Heptageniidae	<i>Epeorus</i>	+	+	-	+
		<i>Rhithrogena</i>	+	-	-	+
Hemiptera	Corixidae	<i>Collicorixa</i>	+	-	-	-
		<i>Corixa</i>	+	+	+	-
	Gerridae	<i>Gerris</i>	+	+	+	-
	Hydrometridae	<i>Hydrometra</i>	+	-	+	-
	Mesoralidae	<i>Mesorelia</i>	+	-	-	+
	Notonectidae	<i>Notonecta</i>	+	-	-	+
	Physidae	<i>Phymata</i>	+	-	+	-
Lamellibrandiat a	Sphaeriidae	<i>Sphaerium</i>	+	-	+	-
Odonata	Agridae	<i>Agrion</i>	+	+	+	+
	Cordulegasterida e	<i>Cordulegaster</i>	+	+	-	-
	Gomphidae	<i>Gomphus</i>	+	+	+	-
	Plactyenemididae	<i>Pyrrhosoma</i>	+	-	-	+

		<i>Enallagma</i>	+	-	-	-
		<i>Platycnemis</i>	+	+	-	-
Prosobranchiata	Valvatidae	<i>Valvata</i>	+	-	-	-
Trichoptera	Hydropsychidae	<i>Hydropsyche</i>	+	+	+	+
		<i>Tinodes</i>	+	+	+	+
Plecoptera		<i>Nemouridae</i>	+	+	+	-
		<i>Neoperla</i>	+	+	+	-
Oligochaeta	Lumbricidae	<i>Lumbricus</i>	+	-	+	-
Crustacea	Decapoda	<i>Eriocheir</i>	-	-	+	+
Frequency			36	20	21	17

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

The abundance of macroinvertebrate genera relative to land-use activities is provided in Table 3. Generally, there was no genera of macro-vertebrate exceeding 10% in relative abundance, while those whose abundance was more than 4% were *Baetis* (9.5%), *Caenis* (5.4%), *Hydropsyche* (4.7%), *Gerris* (4.5%), and *Sphaerium* (4.2%). Differences in the abundance of macro-invertebrate genera were significant relative to land-use practices (Kruskal-Wallis ANOVA; $H = 19.234$, $df = 3$, $P = 0.0004$). Undisturbed sites had the highest abundance of macro-invertebrate genera ($n = 5692$), which was dominated by *Baetis* (8.6%), *Caenis* (5.9%), *Tinoides* (5.3%), *Hydropsyche* (5.0%), *Heptagenia* (4.7%), and *Notonecta* (4.6).

Meanwhile, the settlement areas had the second most abundant macroinvertebrate ($n = 3182$), which was dominated by macroinvertebrates of the genera *Baetis* (16.8%), *Sphaerium* (11.6%), *Caenis* (11.5%), and *Chironomus* (9.5%). The areas within industrial regions had the least abundance of macro-invertebrates ($n = 1692$), which was dominated by genera: *Planorbis* (12.5%), *Baetis* (12.5%), *Rhithrogena* (10.8%) and *Tipula* (9.9%). In the sugarcane-dominated growing, the total abundance of macroinvertebrates ($n = 2510$) was lower than that of the settlement region but higher than the industrial region. The macroinvertebrate was dominated by *Platambus* (11.3%), *Cordulegaster* (10.9%), *Gerris* (9.2%), *Agrion* (9.0%), and *Ephemera* (4.2%).

Table 3. Information concerning the abundance of macroinvertebrates at the four land-use practices from January to August 2020

Order	Family	Genera	Landuse type			
			UNDS	SUGS	SETL	INDS
Pulmonata	Limnaeidae	<i>Lymnaea</i>	44	123	110	0
	Physidae	<i>Physa</i>	54	98	33	0
	Planorbidae	<i>Planorbis</i>	30	72	15	213
Coleoptera	Dytisadae	<i>Coptotomus</i>	26	89	27	159
		<i>Ilybius</i>	21	27	132	132
		<i>Platambus</i>	0	285	0	142

	Gyrinidae	<i>Gyrinus</i>	31	0	0	0
	Haliplidae	<i>Haliphus</i>	0	0	231	0
Diptera	Anthomyiidae	<i>Limnophora</i>	123	0	0	0
	Chironomidae	<i>Chironomus</i>	75	132	303	0
	Tipulidae	<i>Tipula</i>	27	0	0	168
	Simuliidae	<i>Simulium</i>	49	34	0	0
Ephemeroptera	Baetidae	<i>Baetis</i>	489	0	534	213
	Caenidae	<i>Caenis</i>	336	0	366	0
	Ecdyonuridae	<i>Heptagenia</i>	270	211	0	0
	Ephemerallidae	<i>Ephemerella</i>	201	213	0	0
	Leptophtebiidae	<i>Habrophlebia</i>	54	0	0	0
	Heptageniidae	<i>Epeorus</i>	69	33	0	44
		<i>Rhithrogena</i>	30	0	0	183
Hemiptera	Corixidae	<i>Collicorixa</i>	127	0	0	0
		<i>Corixa</i>	261	78	33	0
	Garridae	<i>Gerris</i>	159	231	201	0
	Hydrometridae	<i>Hydrometra</i>	138	0	15	0
	Mesoralidae	<i>Mesorelia</i>	228	0	0	15
	Notonectidae	<i>Notonecta</i>	264	0	0	54
	Physidae	<i>Phymata</i>	137	0	135	0
Lamellibranchiata	Sphaeriidae	<i>Sphaerium</i>	192	0	369	0
Odonata	Agridae	<i>Agrion</i>	212	225	84	33
	Cordulegasteridae	<i>Cordulegaster</i>	207	273	0	0
	Gomphidae	<i>Gomphus</i>	195	117	39	0
	Plactyenemididae	<i>Pyrrhosoma</i>	69	0	0	21
		<i>Enallagma</i>	78	0	0	0
		<i>Platycnemis</i>	228	156	0	0
Prosobranchiata	Valvatidae	<i>Valvata</i>	234	0	0	0
Trichoptera	Hydropsychidae	<i>Hydropsyche</i>	285	53	159	115
		<i>Tinodes</i>	301	45	48	101
Plecoptera	Hydropsychidae	<i>Nemouridae</i>	153	15	0	27
		<i>Neoperla</i>	225	0	228	51
Oligochaeta	Lumbricidae	<i>Lumbricus</i>	70	0	39	0
Crustacea	Decapoda	<i>Eriocheir</i>	0	0	81	21

Total abundance			5692	2510	3182	1692
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¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

The total abundance of each order in River Nzoia is shown in Figure 2. Ephemeroptera was the most abundant at 24% relative abundance, followed by Hemiptera (15.9%), Odonata (14.8%) and Coleoptera (10.0%). There were significant differences in the abundance of macro-invertebrate orders relative to the land-use activities (Kruskal-Wallis ANOVA; $H = 76.312$, $df = 33$, $P < 0.0001$). Higher abundance of Ephemeroptera, Hemiptera, Odonata, Trichoptera and Plecoptera was recorded in areas that are less disturbed. In the settlement areas, Lamelibranchiata, Coleoptera, Diptera and Oligochaetes dominated. In sugarcane growing areas, Pulmonata, Coleoptera, and Odonata were more dominant. Meanwhile, no macro-invertebrate-dominated areas within the industrial activities.

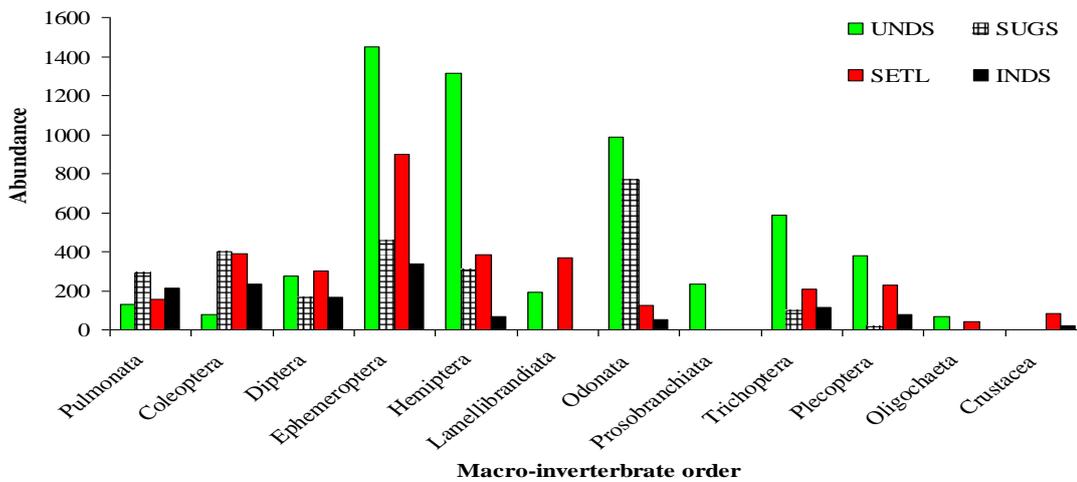


Figure 2. Total abundance of benthic macro-invertebrates' orders in Nzoia River from January to August 2020

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

The benthic macroinvertebrate community species diversity under different land-use attributes is shown in Figure 3. The highest species diversity occurred in the undisturbed areas, followed by sugarcane growing areas, while industrial areas had low species diversity. The high diversity of species in the least disturbed regions indicates that macroinvertebrates prefer less polluted sites in the river ecosystem, as previously stated.

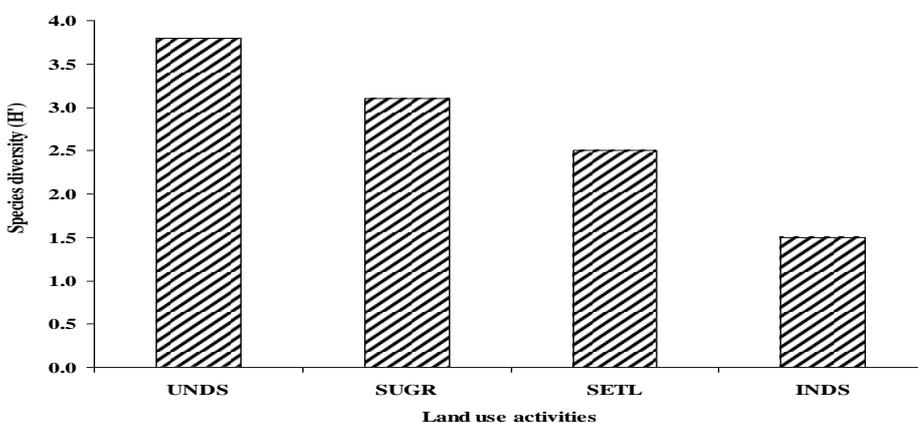


Figure 3. Species diversity relative to various land-use practices in Nzoia River from January to August 2020

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

The overall %EPT was highest in the industrial areas and lowest at the undisturbed sites, with the other two sites being intermediate (Figure 4).

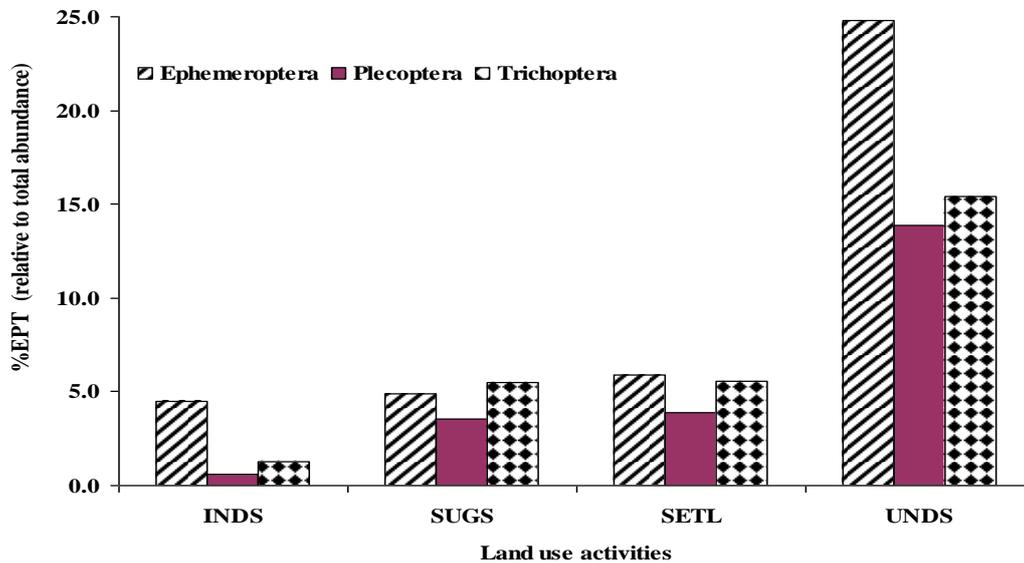


Figure 4. The %EPT relative to various land-use practices in River Nzoia from January to August 2020

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

Influence of land-use on the composition and diversity of mayfly

The results showing the presence/absence of mayfly taxa are shown in Table 4. There were significant differences in the abundance of the species among the sampling sites (Chi-square, $\chi = 7.987$, $df = 3$, $P = 0.0193$). Undisturbed sites had the highest occurrence of mayfly, followed by sites near industrial activities, and the least in settlement regions.

Table 4. Presence (+)/absence (-) of various genera of Ephemeroptera (Mayfly) taxa with respect to land-use practices along Nzoia River from January to August 2020

Family	Genus	INDS	SUGS	SETL	UNDS
Baetidae	<i>Baetis</i>	+	+	+	+
Caenidae	<i>Caenis</i>	+	+	+	+
Coenidaie	<i>Coenus</i>	-	-	-	-
Ecdyonuridae	<i>Heptogenia</i>	+	+	+	+
Ephemerallidae	<i>Ephemeralla</i>	+	+	-	-
Leptophtebiidae	<i>Habrophlebia</i>	-	-	-	+
Heptageniidae	<i>Epeorus</i>	+	+	-	+
	<i>Rhithrogenia</i>	+	-	-	+
	<i>Heptagenia</i>	+	-	-	-
Total occurrence		7	5	3	6

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

Absolute abundance (No. ind.) and relative abundance (%) of mayfly (Ephemeroptera) with respect to land-use practices along Nzoia River are shown in Figure 5. There was a significant spatial variation in the abundance of mayflies in relation to land-use practices Kruskal-Wallis ANOVA; $H = 1223.234$, $df = 18$, $P < 0.0001$). Dominance of *Baetis* (15.7%), *Caenis* (10.4%) and *Heptagenia* (8.3%) was observed in undisturbed sites. In the sugarcane growing region, the dominant genera of mayfly were *Heptagenia* (6.5%) and *Ephemerella* (6.6%), while in the settlement region, two species occurring in high abundance were *Baetis* (16.5%) and *Caenis* (11.3%). Meanwhile, at the industry sites, there was no dominant species except *Baetis*, which occurred at 6.6% and *Rhithrogena* (5.6%).

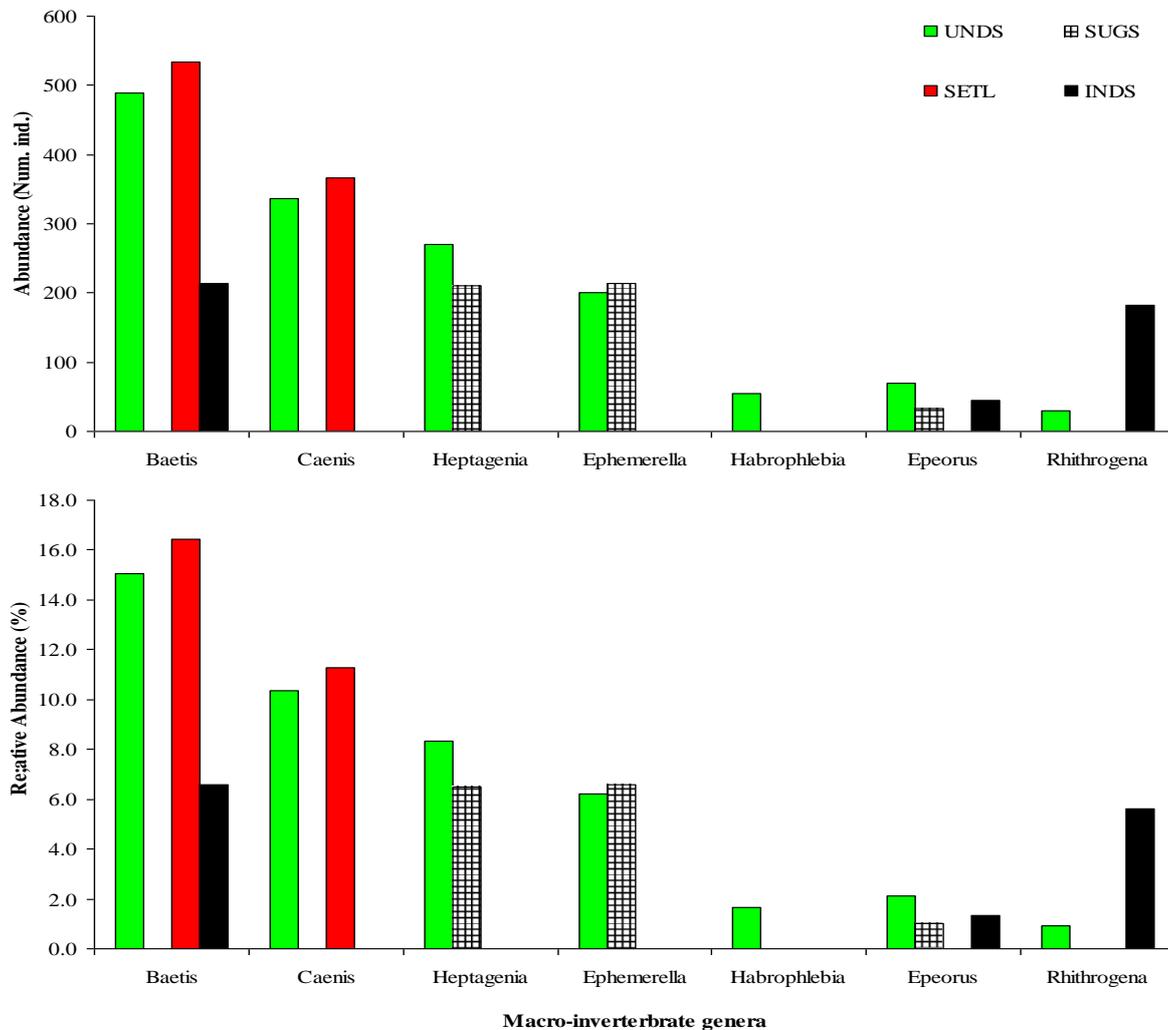


Figure 5. Absolute abundance (No. ind.) and relative abundance (%) of mayfly (Ephemeroptera) with respect to land-use practices along Nzoia River from January to August 2020

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

Interrelationship between land-use practices, water quality parameters and Ephemeroptera (mayfly) taxa

The relationships among water quality parameters and Ephemeroptera taxa composition at the sampling sites during the study are shown in Figure 6. There was a clear distinction between the land-use sites, with the ephemeroptera taxa composition being clearly distinguished in the PCA tri-plot. Areas of industrial activities were significantly correlated with DO, BOD, TA, pH and electrical conductivity, showing positive correlation with *Baetis*, *Coenus*, *Habrophlebia*, *Epeorus*, and *Heptagenia*, as well as with industrial sites. Settlement and sugarcane growing areas were correlated positively with nitrates and phosphates, as well as mayfly of genera *Ephemerella*, *Heptagenia*, and *Caenis*. Undisturbed areas did not control the population of any mayfly, as well

any water quality parameters. Phosphates and nitrates were positively associated with settlement and sugarcane growing. Meanwhile, undisturbed sites had no effects on any physico-chemical parameters.

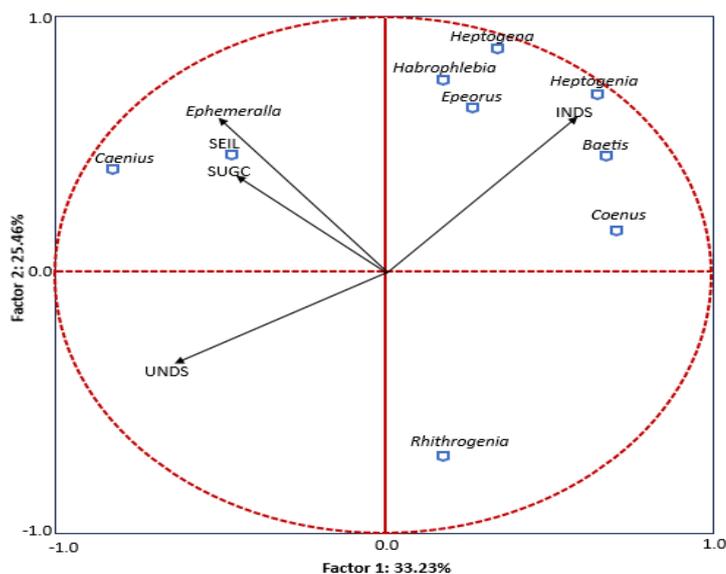


Figure 6: Principal Component Analysis (PCA) of mayfly taxa relative to catchment land-use cover along River Nzoia from January to August 2020

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

The interrelationships among water quality parameters, land-use and ephemeroptera taxa composition at the sampling sites during the study are shown in Figure 7. There was a clear distinction between the land-use sites, with the ephemeroptera taxa composition being clearly distinguished in the PCA tri-plot. There was a clear distinction between the land-use sites, with the ephemeroptera taxa composition being clearly distinguished in the PCA tri-plot. Areas of industrial activities were significantly correlated with DO, BOD, TA, pH and electrical conductivity, showing positive correlation with *Baetis*, *Coenus*, *Habrophlebia*, *Epeorus*, and *Heptagenia*, as well as with industrial sites. Settlement and sugarcane growing areas were correlated positively with nitrates and phosphates, as well as mayfly of genera *Ephemeralla*, *Heptogenia*, and *Caenius*.

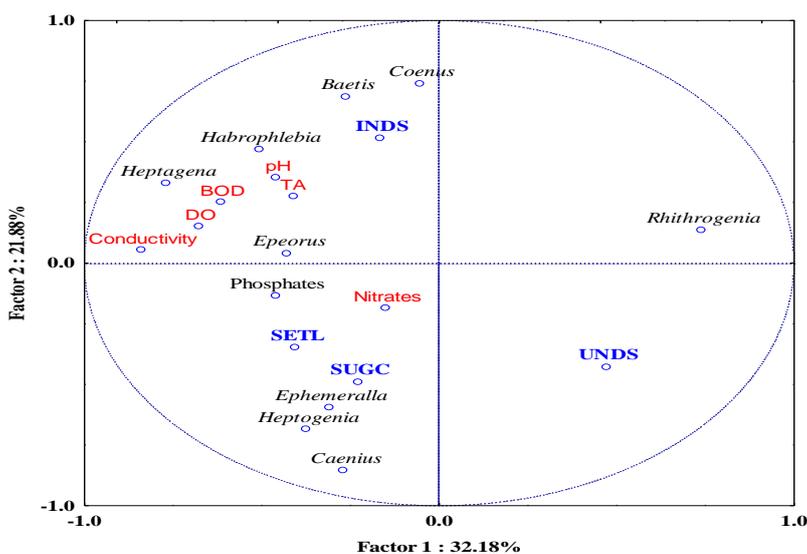


Figure 7. Principal Component Analysis (PCA) of mayfly taxa and water quality relative to catchment land-use cover along River Nzoia from January to August 2020

¹UNDS = Undisturbed sites, INDS = Industrial sites, SUGS = Sugarcane growing sites and SETL = Settlement locations

DISCUSSION

There were differences in physico-chemical parameters relative to land use activities along River Nzoia. The study, high pH values in the sugarcane growing zone, is most likely associated with alkaline conditions owing to the burning of sugarcane during harvesting, which produces ash which mixes with rainwater to form a weak alkaline solution (Trujillo-Narcía *et al.*, 2019). It is also probable that the fertilizers used in the sugarcane farms are mostly alkaline in nature, an assumption that needs further validation. The settlement areas had pH values between 4.6–6.7, indicating large variations from acidic to near neutral pH, which may be associated with the discharge of domestic wastes.

A total of 43 species belonging to 31 genera and 12 orders of macroinvertebrates were observed relative to the land-use activities. The rich biodiversity of macroinvertebrates in the undisturbed site could be attributed to the good water quality due to low human disturbances, as found in previous studies (Mathooko and Mavuti, 1992; Abong'o *et al.*, 2015; Gholizadeh *et al.*, 2021; Maina *et al.*, 2021). It must be highlighted that studies conducted in the upstream River Naromoru (Mathooko and Mavuti, 1992), River Omubira in Kakamega County (Mzungu *et al.*, 2022) and Thika (Maina *et al.*, 2021) found much higher species composition than in the current selected undisturbed sampling sites in the River Nzoia, suggesting that the river may be undergoing a more serious problem of water quality degradation.

The present study suggests that land-use practices are affecting species abundance since less disturbed sites had high genera abundance, mainly of the order Ephemeroptera, Plecoptera, and Trichoptera (EPT), as found in other studies (Menetrey *et al.*, 2010; Ab Hamid and Rawi, 2017; Firmiano *et al.*, 2017). Macro-invertebrates of the EPT taxa prefer less polluted sites and will be in higher abundance in areas with less human disturbance and pollution. The species count in areas undergoing human activities in the selected sampling sites in the River Nzoia rarely exceeds 50, perhaps due to the higher intensity of human activities, which may also serve as pollutants. No specific dominance of macroinvertebrates was observed in the region, and therefore occurrence of *Hydropsyche*, *Gerris*, *Heptagenia*, and *Spaherium* may have been possible because they are multivoltine, which allows them to be rapid colonizers (Merrit *et al.*, 2008). *Chronomus* sp., *Caenis* sp. and *Hydropsyche* sp. have been shown to increase in floodplain rivers (Chessman *et al.*, 2006), suggesting that the river has a tendency to flood during the rainy season. Meanwhile while *Baetis* sp. and *Caenis* sp. have been found in areas of intense food crop production (Johnson *et al.*, 2013; Gutzlera *et al.*, 2015). These patterns clearly indicate that different human activities contribute differentially to species abundance and somewhat validate the use of macroinvertebrate abundance in the study area to detect the influence of land-use practices.

The highest species diversity occurred in the undisturbed areas, followed by sugarcane growing areas, while industrial areas had low species diversity. The high diversity of species in the least disturbed regions indicates that macroinvertebrates prefer less polluted sites in the river ecosystem, as previously stated. The current results support the idea that agricultural activities and industrial land-use resulted in negative changes in water quality as reflected in the macroinvertebrate genera patterns.

The presence of low abundance of EPT in areas with varying land-use practices lends credence that human activities negatively impacted water quality and thus lower the quality of water, indicating some level of water pollution along the river, as established in other comparable studies (Camargo, 1992; Camargo, 1994; Živić *et al.*, 2009; Guilpart *et al.*, 2012). This could be attributed to the human disturbances that reduce canopy cover and introduce many pollutants into the water body. The low species diversity in settlement areas and sugarcane farms can be attributed to the use of various chemicals for agriculture, human wastes, together with a host of other activities that consequently lower the water quality.

Undisturbed sites had the highest occurrence of mayfly, followed by sites near industrial activities, and the least in settlement regions. Apart from the sites near undisturbed sites, a higher occurrence of mayfly in the sites near industries compared to settlement and sugarcane growing is rather peculiar. It could, however, be possible that

these mayfly genera that inhabit those areas undergoing constant pollution develop to become tolerant and exist, but with low abundance, which is now widely documented (Edegbene *et al.*, 2020; Edegbene *et al.*, 2021; Hu *et al.*, 2022). This may be confirmed by reports that have indicated that there are some species of mayfly that may tolerate oxygen depletion (Firmiano *et al.*, 2017), high ammonia (Echols *et al.*, 2010), acidification (Šupina *et al.*, 2022), and extreme pH (Sivaruban *et al.*, 2020). Nevertheless, the current study cannot adequately verify this hypothesis since identification of the macroinvertebrates was done to genus levels, but determining which species tolerate pollution requires identification to species level.

It is still not clear why *Baetis* dominated undisturbed, settlement and industry sites but was absent in the sugarcane farm. Perhaps it is species-wide in distribution since among all the mayfly taxa, the genus *Baetis* is categorized as the most sensitive (Anuradha *et al.*, 2020; Elias, 2020; Strungaru *et al.*, 2021). *Baetis* sp. has been established as being very tolerant, with the ability to survive with constraints to a range of water parameters (Thakur *et al.*, 2023). Previously, it was established that abundance and distribution of *Baetis* sp. in several polluted rivers were mainly influenced by anthropogenic activities, which cause alterations in conductivity, dissolved oxygen, and nutrients (Strungaru *et al.*, 2021). Similarly, population dynamics of *Baetis* sp. are rarely decided by changes in the kind and accessibility of diet, but by the competition from other species inhabiting the sites (Thakur *et al.*, 2023). It has been established that the alteration in the water quality parameters is more likely to alter their abundance of *Baetis* sp., but may not eliminate them (Hamid *et al.*, 2021), which is similar to the present study. *Heptagenia* and *Habrophlebia* dominated the undisturbed region and settlement region, which shows that they respond to moderate pollution. *Rhithrogena* was more dominant in areas of wide industrial practice because this species has been established to tolerate a wide range of pollutants, including acid mines and heavy metals (Hamid *et al.*, 2021). Meanwhile, it also established that Species such as *Habrophlebia* occurred only in undisturbed sites and may be an indicator that they are not tolerant mayfly genera even low levels of pollution.

The present study indicates that different types of land-use practices within the study area caused changes in the abundances of the mayfly taxa. It is possible that undisturbed sites within the forested region allow for high abundance due to the presence of appropriate conditions necessary for the survival of larvae. However, the presence of diverse organic and inorganic pollutants in the areas undergoing human activities, there is a possibility that water quality may not be suitable for some species of mayfly or may allow for them to exist in low abundance. The highest species diversity occurred in the undisturbed areas, followed by sugarcane growing areas, while industrial areas had low species diversity. The high diversity of species in the least disturbed regions indicates that macroinvertebrates prefer less polluted sites in the river ecosystem, as previously stated. The current results support the idea that agricultural activities and industrial land-use resulted in negative changes in water quality as reflected in the macroinvertebrate genera patterns.

It is possible that undisturbed sites within the forested region allow for high abundance due to the presence of appropriate conditions for the survival of larvae. However, the presence of diverse organic and inorganic pollutants in the areas undergoing human activities, there is a possibility that the water quality may not be suitable for some species of mayfly. *Baetis* sp. It has been established that the changing land-use and alteration in the water quality parameters are more likely to alter their abundance of several species of mayfly (Hamid *et al.*, 2021). *Rhithrogena* was associated with any land-use activity since this species has been established to tolerate a wide range of waterbodies, including acid mines and heavy metals (Hamid *et al.*, 2021).

CONCLUSIONS

The present findings demonstrate that land use activities influence the physicochemical parameters and macroinvertebrate assemblage. All the physico-chemical water quality parameters displayed significant ($P < 0.05$) spatial variations. The concentration of DO was lowest and BOD highest in areas with industrial activities, followed by sugarcane farms, but was lowest BOD₅ in the undisturbed areas. The highest of NO₃⁻ and PO₄²⁻ occurred in sugarcane farms and settlement sites. Macroinvertebrate species composition was highest at the undisturbed sites, followed by sugarcane, while sites adjacent to the settlement and industrial areas had the lowest species numbers. There was a clear pattern showing that different land use patterns affected macro-invertebrate community structure, where sites with low disturbances had high composition, abundance and diversity and

were dominated by order EPT, while sites closer to industrial activities had low species composition, abundance and diversity. Meanwhile, mayfly occurrence was significant relative to land-use practice ($P < 0.05$). Undisturbed sites had the highest occurrence of mayfly, followed by sites near industrial activities, and the least in settlement regions, suggesting possible occurrence of more tolerant species of mayfly in sites near industrial areas. There were significant interrelationships between the land-use practices, water quality and Ephemeroptera taxa composition. The concentrations of DO, BOD, TA, pH and conductivity were positively associated with industrial activities, while the phosphates and nitrates were positively associated with settlement and sugarcane growing, leading to the conclusion that the present study indicates that different types of land-use practices within the study area caused changes in abundances of the mayfly taxa.

The macroinvertebrate richness in terms of family: genus: species ratio reflects that a loss of species in any site would mean loss of the entire genus (or family). This indicates the necessity of preserving all the sampled rivers if species or genus richness conservation should be a priority. It is also time to declare these selected sampling sites in the Upstream River Nzoia as endangered, and that the extinction will result in the extinction of unique macroinvertebrate species. As such, human activities along the Upstream River Nzoia should be regulated. The presence of a larger number of species tolerant to anthropogenic impacts could signal human-induced perturbations; thus, all stakeholders should formulate immediate policies that will reduce human impacts on the macroinvertebrate composition of the selected sampling sites in the River Nzoia.

RECOMMENDATIONS

Several recommendations were formulated based on the research findings as follows: First, there is a need to protect existing riparian forest strips by maintaining a natural canopy to control temperature and sediment input, as well as engaging local communities in monitoring EPT indicators every six to twelve months. This includes Capacity building of farmers on how to use controlled fertilizer application on sugarcane farms by introducing soil-testing-guided fertilization to reduce excess nutrient runoff. Secondly, there is a need to establish a catchment management committee to involve farmers, industries, NEMA, WRA, and KFS through public awareness programmes. This would include building the capacity to educate households on safe disposal and the ecological role of Ephemeroptera as bioindicators.

Third, farmers should be encouraged to plant napier grass and indigenous trees to trap sediments and agrochemicals, which serve as riparian buffers. And for this to happen, it is necessary for riparian demarcation and rehabilitation to remove illegal structures along the Nzoia River. Fourth, the government should introduce pollution effluent compliant monitoring monthly programmes for BOD, total suspended solids, and Ammonia at the factory discharge point. Fifth, there is also, a need to upgrade the effluent Pre-treatment system by introducing chlorine-free bleaching practices at Webuye Pan Paper industry. And, finally, there is a need to establish a pollution-controlled Buffer Zone by re-vegetating the river banks along the Nzoia River with water purifying plants such as reeds (*Phragmites Spp.*)

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