

A Comparison of the Efficiency of Aquatic Macroinvertebrate Sampling Tools Used in Lotic Environmental Impact Assessment of Human Activities in A Tropical Mountain Stream in Eastern Uganda

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Abstract

The study was aimed at comparing the efficiency of three macroinvertebrate sampling tools used in lotic environmental impact assessment of River Sipi including Surber sampler, rock-filled basket and Kick net sampling tools. The efficiency of the sampling tools was based on the data collected by each sampling tool, which was in turn used to calculate the Relative variation (RV) (efficiency), diversity, richness, and relative abundance, time taken to sort macroinvertebrate per sample and taxa assemblage. Data was analysed using a two-way ANOVA that was performed under the R Development Core Team 2010. The results indicated that despite the fact that all the sampling tools were efficient in sampling macroinvertebrates with $RV < 25\%$, both kick net and rock-filled basket obtained closely comparable efficiency results with Relative Variation ranging between 0.54-2% for the kick net and 1.4-3.6% for rock filled basket. Of the three sampling tools, the kick net was the most efficient, collecting a greater diversity of macroinvertebrate taxa (1.81 ± 0.04) and a greater number of specimens (1444), abundance ($66.4 \pm 0.25\%$), greater taxa richness (11 ± 0.41) but required the biggest amount of time to sort macroinvertebrates (26 ± 0.71 minutes). In conclusion, the rock-filled basket served as the next best alternative efficient sampling tool to kick net in sampling macroinvertebrates.

Key words: Artificial substrates, Kick net, Relative variation, Rock-filled basket, sampling tools

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Introduction

Benthic macroinvertebrates are widely used as the best organisms to biomonitor stream ecosystem health and biodiversity altogether. This is because they are the best indicators of aquatic environmental quality and are found in all aquatic habitats (Buss et al., 2015). They also integrate, physico-chemical and biotic conditions of lotic systems and, above all, they are easy to sample using the available and appropriate sampling tools (Buss et al., 2015).

A number of macroinvertebrate sampling tools have been developed for use in Biomonitoring aquatic resources especially in lotic systems including but not limited to; D-frame nets, Hess samplers, kick nets, rock-filled baskets, porn grabs, U-nets, dragnets and Surber samplers (Buss et al., 2003; Dickens & Graham, 2002; Storey 1991; DePauw et al., 1986). Each of these sampling tools offer a varying degree of efficiency in sampling aquatic macroinvertebrates. Since there is no universally and internationally or region-specific agreed sampling tool for bio assessment, most researchers over the world choose the sampling tools based on availability and affordability without considering efficiency of the tools used. This at times may lead to over or underestimation of macroinvertebrate community assemblage. It is important that efficiencies of several sampling tools are known and comparison made to determine the best two or three alternative sampling methods that are approved for a specific region's water resources bioassessment. This would be a step towards identifying internationally accepted efficient sampling tools (Cunha et al., 2019).

An efficient sampling tool should requires less time for sample processing, more so in rapid bioassessment of streams. The sample processing time also depends on the quality of organisms and the amounts of debris collected in a sample, which depends on the type of the sampling tool used (Cunha et al., 2019; Southwood 1978). A sampling tool that is not efficient makes sampling

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process more tedious, laborious, and expensive and requires long time to process a sample as it may let in more detritus with the sample or may be so small that you need so many replicates to make up one sample.

In Uganda, most sampling teams have used either kick net, Surber sampler or D-net. However, the tools of these sampling approaches have in most cases suffered from limited accessibility to some microhabitats of streams especially during bankful discharges of stream thereby requiring much time to process the samples (Kasangaki et al., 1991). If alternative sampling tools are employed, and their efficiencies tested and known, then they would be used as alternatives against the common sampling tools. If they are better in terms of efficacy than the most commonly used as alternative tools used when situations warrant especially where some micro habitats are inaccessible or when then bankful discharge of stream limits kick net and D-nets or Surber samplers. Some rivers are dangerous in that they habit snakes and crocodiles. Nevertheless, the use of the D-nets, Surber sampler and kick nets require that the sampling manpower has to step in water to kick substrates. It is important to note that D-net and Surber sampler are small in size, which limits their setting on rough substrates and deep waters, which therefore requires that an alternative method that can produce comparable results be tested for efficiency and performance. When a reliable alternative is found in this region, it is universally considered an efficient sampling tool for sampling Uganda's lotic system.

Despite the observable weaknesses of the sampling tools mentioned above used in east Africa in general and Uganda in particular, comparative studies about their efficiency or performance in macroinvertebrate sampling has not been done. In this study therefore, we compared the efficiency and the overall performance of kick net (1M high x1M wide), Surber sampler (0.5x0.5M square

opening on a 2m long handle) and a rock-filled basket in sampling macroinvertebrates in river Sipi in eastern Uganda. This study was aimed at comparing the efficiencies of the three tools so that we could recommend the most efficient tool for use in Uganda's lotic environmental biomonitoring. In this case, a rock-filled basket was introduced as an alternative to the kick net and Surber sampler and a comparison was made about their performance and efficiency in sampling macroinvertebrates.

Materials and methods

Study site

The study was carried out on three study sites in three reaches of River Sipi that is in the upper reaches in the pristine where the river is in the protected natural tropical forest. Here, the substrate was dominated by rock bed, some pebbles, some woody debris and coarse organic substrates like rotting leaves and lianas and other vegetal substrates. The riparian vegetation was mainly dense tropical forest, hanging leaves in stream water. The average channel width was 1.4m, average channel depth was 12cm and the average flow velocity was 0.6m/s. The sampling point chosen was located at 657645.81mE, 147303.15mN. D.o was 11.2 ppm, pH was 7.4 and temperature was 18.2⁰C. In the middle reaches, the river is disturbed by arable activities. The riparian vegetation was dominated by banana plants and coffee trees. The major crops grown are bananas; coffee, beans, maize and the river substrates were dominated by boulders, sand, pebbles, cobbles and some coarse organic substrates. The average channel width was 3.1m., average channel depth was 18cm and the average flow velocity was 0.2m/s. The sampling point chosen was located at 644978.70mE, 147820.11mN. D.o was 8.6 ppm, pH was 8 and temperature was 21.8⁰C. In the lower reaches where the river dissects and is disturbed by the Nabongo urban influences, the

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substrates are dominated by sand, pebbles, shingle, gravel, little mud and boulders. The stream was devoid of riparian vegetation. The average channel width was 2.1m., average channel depth was 20cm and the average flow velocity was 0.4m/s. D.o was 7.8 ppm, pH was 7.9 and temperature was 23.3⁰C. The sampling point chosen was located at 644391.39mE, 147735.40mN.

Sampling Procedure

The sampling was launched in four campaigns of macroinvertebrate sampling; twice in the wet season and twice in the dry season from February 2020 – November 2020 using three sampling tools including kick net, Surber sampler and a rock-filled basket. Below is the description of how each sampling tool was used;

Kick net: This was made of a large square screen net mesh size 0.3mm, 1mx1m wide between two metallic poles with two handles measuring 1m from the top of the net frame. One person held the net downstream of the targeted microhabitat to be sampled against the water flow direction, while another person started kicking the substrates in the river water upstream from the kick net, thereby dislodging the macroinvertebrates, which were carried by the stream flowing water into the kick net. Kicking each microhabitat lasted for 3 minutes. Samples from the kick net obtained from the different micro habitats sampled on a given sampling site were pooled together to form one sample, put in a sampling bottle, 70% ethanol added, sealed, labeled and taken to the laboratory for further processing.

Surber sampler: An appropriate riffle or run was identified with depth equal or lower than that of the height of the sampler 40 cm wide × 40 cm long and 40 cm high with 0.16 m² catching area and a net of 0.3 mm mesh. The Surber sampler frame base was firmly set into the substrates with its opening faced upstream, against the water flow. One person disturbed the streambed in the area

inside the sampler's frame. Larger rocks/pebbles were individually inspected and cleaned of all macroinvertebrates using a hand. However, at some points, we used a kitchen scrubbing brush into the net while holding the net of the sampler. We ensured that all dislodged individuals could flow into the net. The remaining finer substrates were disturbed by hands up to a depth of 5cm. all the samples at each site were pooled together to form one sample, put in a sampling bottle, 70% ethanol added, sealed and taken to the laboratory for further processing.

Rock-filled basket: We fabricated a total of 18 equal sided heavy duty plastic basket cubes (containers) of 4mm thickness and of an area of 0.125m³ (0.5m long x 0.5 m wide x 0.5m high each) with 16 perforated holes of 6cm diameter on each basket as described by Depauw et al., (1986). The basket was filled with artificial substrates that were locally available and found dominating each river sampling site usually dominated by rocks. The basket was then placed and enclosed round with a fish net mesh size 5cm to allow macroinvertebrates enter and colonize the substrates. The substrate –filled baskets were then placed in the sampling points of choice at each sampling site. For example, at the site in the upper reaches, one was placed in a riffle, another in a micro plunge pool, another in a run, and other two on bank sides. These baskets were tied with ropes on the riparian trees while others were tied on poles/wedges planted on the riverbank sides to keep them where they were placed without being swept away by the water current. These baskets were left in river for four weeks, a time that is enough for macroinvertebrates to colonize a habitat. After four weeks, these baskets were retrieved. On retrieval of each basket, a bucket was placed under it to prevent the loss of entrapped macroinvertebrate from the colonised substrate of the basket. At each site, all the macroinvertebrates from the 6 baskets were pooled together to form one sample, put in a sample bottle, and 70% ethanol added sealed awaiting laboratory processing.

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Surber sampler and kick net were only used to collect macroinvertebrates at the time of retrieving rock-filled baskets.

While in the laboratory, macroinvertebrates were identified and grouped into their respective taxonomic groups up to family level following the identification guides set by Merritt and Cummins (1998). Data about macroinvertebrates taxon diversity, richness, Rv, relative abundance and time for processing were obtained based on the laboratory data.

Taxon diversity and richness of macroinvertebrates was determined at each site using Shannon Weaver's Diversity Index (Shannon & Weaver, 1949) to compare the macroinvertebrate taxon diversity between various habitats associated with different wood treatments as follows:

$$H = - \sum_{i=1}^n \left(\frac{n_i}{N} \log_2 \left(\frac{n_i}{N} \right) \right)$$

Where,

H= Shannon Wiener index of diversity;

n_i = Total No. of individuals of a taxon;

N= Total No. of individuals of all taxa.

Relative abundance of taxa was calculated from the formula denoted by;

$$R.A = \frac{\text{Number of individuals of one taxon}}{\text{Total number of individuals on a site}} \times 100.$$

According to Taylor et al. (2001), processing time refers to the amount of time it takes to sort 100 macroinvertebrates individuals from a sample, which is denoted by the formula;

$$\text{Processing time} = \frac{\text{Duration (number of minutes) spent sorting macroinvertebrates}}{\text{Number of macroinvertebrates individuals sorted}} \times 100$$

Relative variation (RV) was estimated to determine the efficiency of each sampling tool in macroinvertebrate sampling. Relative variation is defined as the ratio of the standard error of the mean (SEM) divided by the mean (m) and presented as a percentage (%). This is denoted by the formula; $RV (\%) = (SEM/m) \times 100$ (Pedigo et al., 1982). Where SEM = standard error of the

mean, m = mean of the individual macroinvertebrates. The standard is 25%, beyond which a tool is considered inefficient. An RV of 25% is equivalent to a sampling error of 20% (Southwood, 1978), which is the maximum error level accepted in aquatic sampling (Elliot, 1972).

For comparison purposes, the collection sites were standardized by ensuring that each sampler was used to collect macroinvertebrates from stones in current, runs, pools, riffles and submerged coarse/ vegetal debris and that all sampling tools were used for a similar size of sampled area (10 m although not necessarily continuous) per site.

Supporting site characteristics as if dissolved oxygen, temperature, and pH for each site were determined in 'situ' using a multi-parameter analyzer model Consort C3010/C3030 dual channel. Measurement of velocity, depth, and width of the wetted channel on each site. Velocity was measured using buoyant dry sticks and a stop clock over a stretch of 5m. The channel width and depth were measured using a tape measure and wading rod.

Data analysis

For comparison in differences of the macroinvertebrate assemblage metrics by different sampling tools, a parametric (ANOVA) approach was used. Before the comparison, a normality test using Shapiro-Wilk was applied to macroinvertebrate assemblage metrics. After all the data passing the normality test, two-way ANOVA was performed to assess the differences between means of dependent variables (assemblage metrics) from the different sampling tools. For those models where it was found to be significant under ANOVA, a post hoc test using Turkey's Honestly Significant Difference (HSD) test was done, all the statistical analyses were performed under the R Development Core Team 2010.

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Results

Macroinvertebrate totals obtained by different sampling tools at different sites

Data about the efficiency of the three sampling tools tested was categorized into two that is the macroinvertebrate assemblage arising from total numbers of individuals and taxa, and the averaged data obtained at different times by different sampling tools and sites, which we called average distribution of macroinvertebrate metrics, obtained using different sampling tools over time at different sites. These two sets of data were presented differently for clarity with each in a separate table. To start with, Table 1 shows how different macroinvertebrates totals were obtained using the three tools at the three sites.

Table 1: Macroinvertebrate totals obtained by different sampling tools at different sites

Order	Family	Natural forested site (upper reaches)			Agricultural site (middle reaches)			Urban site (lower reaches)		
		Kick net	Surber	Rock – filled basket.	Kick net	Surber	Rock – filled basket.	Kick net	Surber	Rock – filled basket.
Pelecypoda	Carbiculidae	21	4	7	9	-	7	-	-	-
	Perlidae	19	7	13	16	9	13	12	5	9
	Simulidae	21	-	-	9	-	-	12	-	-
Diptera	Chironomidae	-	-	-	107	5	18	348	9	18
	Tipulidae	54	7	13	16	11	13	10	6	17
	Hydropsychidae	113	31	62	55	18	62	17	12	77
Trichoptera	Hydrophilidae	58	19	37	34	9	23	-	-	11
	Glossosomatidae	-	-	-	9	17	9	-	-	-
	Oligoneuridae	103	12	23	29	9	23	-	-	2
Ephemeroptera	Perlodidae	-	-	-	-	-	-	9	-	16
	Ephemeralidae	91	13	32	24	8	17	-	-	3
	Heptagenidae	-	-	-	-	17	-	19	9	22
Odonata	Coenagrionidae	23	-	-	8	-	-	-	-	-
	Libellulidae	25	5	9	16	-	9	16	-	-
	Aeshnidae	47	6	11	6	-	11	-	-	-
Annelida	Leeches	-	-	-	-	8	-	11	4	7
Crustacean	Palaemonidae	21	4	33	6	4	2	-	-	-
	Potamonautidae	23	3	15	-	-	-	-	-	-
Hemiptera	Belostomatidae	-	-	-	16	-	22	9	8	-
Plecoptera	Peltoperlidae	-	-	-	-	-	-	2	-	-
Number of orders	9	7	6	6	7	6	7	8	6	5
Number of families	20	13	11	11	15	11	13	11	7	10
total number of individuals	2380	619	111	255	360	115	220	465	53	182

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In Table 1, we have a total of 2380 animals from the three sampling sites, of which 985 were harvested from the forested site, 695 from agricultural site and 700 from urban site. These represented 20 families and 9 orders. At all the three sites, the Kick-net sampling tool fetched more macroinvertebrate individuals than other tools, while the Surber sampler fetched the least number of macroinvertebrate individuals. Apart from the middle reaches, the distribution of macroinvertebrate families followed the order of macroinvertebrate individuals having the lowest number obtained by the Surber sampler as compared to the Kick net that had the highest number of families followed by the rock-filled basket. The Surber sampler maintained the same number of orders at 6 throughout the study area, while the number of orders obtained using kick net and rock-filled basket varied from site to site.

At the forested site found in the pristine stage of the river, the most abundant family of all that were harvested using a Kick-net was Hydropsychidae (Trichoptera) with 113 individuals, while the smallest was Perlidae (pelecypoda/scrapper) with 19 individuals. At the same site, Hydropsychidae also dominated the catch by Surber and by rock-filled basket-sampling tools with 31 and 62 individuals respectively. Of all the sites, Potamonautidae were only found at the forested site, while Peltoperlidae were the rarest with only 2 individuals and only obtained by the Surber sampler at the urban site. Chironomidae were only found in the middle and lower reaches but none was found in the forested site. At the agricultural site, Chironomidae dominated the catch by kick net with 107 individuals while Hydropsychidae dominated the catch by Surber and rock-filled basket with 18 and 62 individuals respectively.

At the urban site, Kick net fetched the highest number of orders (8) and families (11), as compared to rock-filled basket that fetched 5 orders and 10 families as well as Surber sampler that fetched 6

orders and 7 families, but also harvested the highest number of macroinvertebrate individuals (465) of which 348 were chironomidae as the most dominant family. Chironomidae also dominated the Surber macroinvertebrate catch with only 9 individuals. 77 individuals of Hydropsychidae on the other hand, dominated the macroinvertebrate catch at the urban site by rock-filled basket. The rock-filled basket provided a stable habitat for fast mobile Trichoptera (hydropsychidae) as opposed to the sand and loose cobbles at this site.

Average distribution of macroinvertebrate metrics obtained using different sampling tools over time at different sites

Since we had more than one factor variables (site and metric), a two-way ANOVA was appropriate in explaining whether independent variables (sampling tools) interacted with the dependent variables (macro vertebrate metrics). In addition, the results were summarized in Table2 before explanation.

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Table 2: Average distribution of macroinvertebrate metrics obtained using different sampling tools over time at different sites

Site	Sampling tools	Average Total number of macroinvertebrate individuals	Average Time spent in sample sorting samples (minutes/100 macroinvertebrate individuals)	Relative Variation (RV=SE M/M*100%)	Average Macroinvertebrate taxon Diversity	Relative Abundance(%)	Taxon Richness (Effective No. of families)
Forested site	kick net	154.75±1.11 ^d	26±0.71 ^c	0.7	1.64±0.01 ^a	62.8±0.48 ^b	9±0.41 ^b
	Rock filled basket	63.75±1.11 ^e	18±1.08 ^d	1.7	1.515±0.15 ^a	25.9±0.52 ^e	5±0.41 ^c
	Surber	27.75±2.81 ^f	12±0.71 ^e	10.1	1.33±0.02 ^b	11.3±0.19 ^h	6±0.00 ^d
Agricultural site	Kick net	90±1.78 ^a	19±1.22 ^a	2	1.81±0.04 ^a	51.8±0.41 ^a	11±0.41 ^a
	Rock filled basket	55±1.96 ^b	17±0.35 ^a	3.6	1.79±0.05 ^a	31.7±0.18 ^e	8±0.41 ^c
	Surber	28.75±2.63 ^c	14±0.82 ^b	9.2	1.29±0.04 ^b	16.5±0.29 ^f	7±0.41 ^d
Urban site	kick net	116.25±0.63 ^g	24±1.08 ^f	0.54	1.67±0.05 ^a	66.4±0.25 ^d	7±0.71 ^c
	rock filled basket	45.5±0.65 ^h	14±0.82 ^g	1.4	1.49±0.23 ^a	26±1.78 ^e	6±0.41 ^c
	Surber	13.25±2.69 ⁱ	9±1.08 ^h	20.3	1.32±0.02 ^b	7.6±0.25 ^g	3±0.41 ^e

Different metrics with different letters (a, b, c, d, e, f, g, h, i) in the same column indicate significant differences at P<0.05. SEM means standard error of mean, M is mean based on total individuals sampled by each tool at a site.

From Table 2, the highest average number of macroinvertebrate individuals was obtained using kick net sampler with 154.75±1.11 at the forested site, while the least average was obtained using Surber sampler with 13.25±2.69 individuals from the urban site. It can be noted that higher macroinvertebrate numbers were obtained from the forested site by all sampling tools and went on reducing at the agricultural site whereas the urban site recorded the least number of macroinvertebrates by all sampling tools. This implies that irrespective of the sampling tool used, forested site had a bigger macroinvertebrate population than the agricultural and urban sites. Based

on ANOVA results, the average number of macroinvertebrates varied significantly from site to site and from sampler to sampler at $P < 0.05$.

At all sampling sites, Kick net required more time for sorting macroinvertebrates compared to the Surber that required the least amount of time. At the forested site, kick net required 26 ± 0.71 minutes to sort macroinvertebrates, while rock-filled basket required 18 ± 1.08 minutes as compared to Surber that required only 12 ± 0.71 minutes. At the agricultural site, Kick net required less time to sort macroinvertebrates (19 ± 1.22 minutes) than at the urban site (24 ± 1.08 minutes). Since there were more macroinvertebrates corresponding with more time, we can propose that the more the macroinvertebrates, the more the time needed. The ANOVA results showed that, rock-filled basket and kick net at the agricultural site had a similar distribution of time required to sort the macroinvertebrates at $P > 0.05$. That notwithstanding, the time required to sort macroinvertebrates significantly varied from site to site and sampling tool to sampling tool at $P < 0.05$.

Based on relative variation (RV), all the three sampling tools were efficient with their $RV < 25\%$. In comparison of the three, at all sites however, Kick net was the most efficient tool with a RV not exceeding 2%. Rock filled basket was the next best with a RV close to that of a kick net that is not exceeding 3.6% that was recorded at the agricultural site. Despite collecting fewer macroinvertebrate, the Surber sampler registered the lowest efficiency in collecting macroinvertebrates with RV ranging from 9.2% recorded at the agricultural site to 20.3% recorded at the urban site. This implies that kick net is capable of collecting more macroinvertebrates irrespective of the nature of the microhabitat as long as the river is wadeable. The second-best alternative to the kick net is the rock-filled basket rather than Surber sampler. At the urban (lowest reach sampling sites, there were more pools than runs and riffles, which affected the effectiveness

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of the Surber sampler by dropping it to 20.3%, while it is here where the kick net was most efficient given its size and height that was not affected by water levels, thereby increasing its efficiency (RV) to 0.54%.

The highest macroinvertebrate taxon diversity was recorded by the kick net at the agricultural site with 1.81 ± 0.04 while the lowest diversity was registered by Surber sampler at the same site with 1.29 ± 0.04 . Taxa diversity was lowest at the urban site, possibly due to the urban stresses that affect the survival rate of macroinvertebrates. This pattern, however, does not hold for the least disturbed forested site. The distribution of taxa diversity collected by Surber sampler did not vary from site to site ($P > 0.05$) but significantly differed from diversity recorded by other samplers at $P < 0.05$. This difference of Surber from other tools but similarity at different sites could be attributed to the size of the Surber.

Despite the fact that urban site had the lowest diversity, it registered the highest relative abundance of taxa of the three sites by all the three sampling tools. This could be because the few taxa occurred in higher numbers. The highest relative abundance was associated with the kick net with $66.4 \pm 0.25\%$ recorded at the urban site, while the lowest was recorded by a Surber sampler with $7.6 \pm 0.25\%$ recorded at the same site. No site had a similar distribution of relative abundance ($P < 0.05$), but the rock filled basket had a similar distribution of relative abundance at all sites at $P < 0.05$.

The highest taxa richness of macroinvertebrates was associated with kick net at all sampling sites, while the lowest richness was associated with Surber sampler. The highest richness was 11 ± 0.41 families recorded at agricultural site using kick net, while the lowest was 7 ± 0.41 families at the same site using Surber sampler. Rock-filled basket had a similar distribution of taxa richness

at different sites at $P > 0.05$, but this significantly differed from the rest of the sampling tools and sites at $P < 0.05$.

Discussion

Macroinvertebrate totals obtained by different sampling tools at different sites

At all the three sites, the Kick-net sampling tool fetched more macroinvertebrate individuals than other tools while Surber sampler fetched the least number of macroinvertebrate individuals. Kick net had the highest number of families followed by the rock-filled basket. This is because the size of the kick net screen size of 1m^2 was large enough to trap more organisms as opposed to the 0.16m^2 area of Surber sampler we used. The findings of this study is in consonance with Brua et al. (2011) and Buss et al., (2015) who established that Kick net sampling is the most highly-effective tool for collecting an accurate representation of benthic macroinvertebrate communities and therefore, continues to be the standard choice of sampling protocol for use in river bioassessment procedures. The claim that it is the most effective tool for collecting the accurate representation of benthic macroinvertebrate communities is borne out of the fact that it disturbs a larger area of macroinvertebrate microhabitat.

The forested site found in the pristine stage of the river recorded the highest abundance of families recorded by all sampling tools compared to sites below it. The most dominant taxa were Hydropsychidae. Potamonautidae were only found habiting this site. Of all the sampling tools, kick net harvested the highest taxa abundance. This may be attributed to the environmental factors like better water quality in the less disturbed pristine forested site where D.O was high, turbidity and temperature were low, and pH was neutral (Kasangaki, et al., 2008). The submerged woody

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debris contribute with an increase in the complexity and heterogeneity of the habitat, serving as shelter and feeding place for a diverse fauna (Thomaz & Cunha, 2010).

At the urban site, Kick net fetched the higher number of orders (8) and families (11), as compared to rock-filled basket that fetched 5 orders and 10 families. It also harvested the higher number of macroinvertebrate individuals (465) compared to 182 of the counterpart tool. It can be noticed that rock-filled basket scored closer results to kick net. These results of rock-filled basket were obtained after leaving the artificial substrates in the baskets in water for only 4 weeks. If the artificial substrates in the baskets were given more time for macro colonisation, they probably would have yielded the same or even more macroinvertebrates than the kick net. Thorp et al., (1985) suggested 1-8 weeks for full colonisation by macroinvertebrate communities

There is need for comparing results obtained by more than one sampling tool during aquatic environmental bioassessment. For example, in a case where environmental conditions (such as the presence of dangerous wildlife, inaccessibility due to water depth or fast flow) prevent the use of kick net procedures, which require sampling personnel to enter the water, an alternative sampling tool, which produces data that is comparable would suffice and be considered equally accurate (Brua et al., 2011). From this study, based on the RV, the number of macroinvertebrate individuals and taxa obtained from kick net were closely comparable to those obtained by the use of rock-filled basket means provided that the rock-filled basket could fit in majority of microhabitats and unlike kick net, it would not require sampling personnel to enter water even during bankful discharge, it is plausible to rate it the next best alternative sampling tool after kick net.

Average distribution of macroinvertebrate metrics obtained using different sampling tools over time at different sites

It can be noted that higher average macroinvertebrate numbers were obtained from the forested site by all sampling tools and reduced at the agricultural site, while the urban site recorded the least number of macroinvertebrates by all sampling tools. This implies that irrespective of the sampling tool used, the forested site had a bigger macroinvertebrate population than the agricultural and urban sites. Humphreys et al., (1998) had earlier observed that the mean number of families per sample differed among sampling tools and sites with the greatest number of families being collected by rock-filled basket followed by snag and airlift sampler ranked as the third based on the number of families. In our study, the Kick net recorded the highest number of macroinvertebrates followed by the rock-filled basket. The differences might have been due to the differences in sampler sizes and or differences in sampler efficiencies.

At all the sampling sites, Kick net required more time for sorting macroinvertebrates than the Surber. At the forested site, kick net required 26 ± 0.71 minutes to sort 100 macroinvertebrates, while rock-filled basket required 18 ± 1.08 minutes as compared to Surber that required only 12 ± 0.71 minutes. At the agricultural site, Kick net required less time to sort macroinvertebrates (19 ± 1.22 minutes) than at the urban site (24 ± 1.08 minutes). The difference in time needed depended on the amount of detritus and amount of macroinvertebrates in a sample by a tool. The Surber sampler scooped less detritus and less macroinvertebrates, while the kick net scooped more detritus and macroinvertebrates that required more time to sort macroinvertebrates. A similar finding was established by Cunha et al. (2019) who showed that the sample processing time was

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faster for D-frame net samples than Surber sampler and square net samples because of smaller quantity of sediment and detritus (13.36 g) collected together with the organisms.

According to South wood (1978), sampling techniques with RV >25% have low precision and those with RV values of 25% or lower have good sampling precision. This study based on relative variation (RV), all the three sampling tools were efficient with their RV<25%. In comparison of the three, at all sites however, the Kick net was the most efficient tool with a RV not exceeding 2%. Rock filled basket was the next best with a RV close to that of a kick net ie not exceeding 3.6% that was recorded at the agricultural site. Despite collecting fewer macroinvertebrate, the Surber sampler registered the lowest efficiency in collecting macroinvertebrates with RV ranging between 9.2% recorded at the agricultural site to 20.3% recorded at the urban site. This result indicated that the kick net was the most efficient at collecting macroinvertebrates.

The highest macroinvertebrate taxon diversity was recorded by the kick net at the agricultural site with 1.81 ± 0.04 while the lowest diversity was registered by the Surber sampler at the same site with 1.29 ± 0.04 , a result that contradicts that of Brua et al. (2011), who indicated that the kick net scored a lower Shannon–Wiener Diversity compared to the U-net. This is because the U-net that Brua et al. (2011) used had a big entrance and was dragged on the river's bed for longer distance and so fetched more and diverse taxa as compared to kick net. In our study, the kick net was bigger in surface area sampled than the Surber and rock-filled basket hence more diversity of macroinvertebrates obtained.

The highest relative abundance of macroinvertebrate taxa was associated with the kick net with $66.4 \pm 0.25\%$ recorded at the urban site, while the lowest was recorded by a Surber sampler with $7.6 \pm 0.25\%$ recorded at the same site. This finding is similar to the one by Turyahabwe et al.,

(2021), who noted that Kick-net sampling tools tended to have higher relative abundance of Chironomidae than other samplers did after correcting for multiple comparisons. The abundance is attributable to the larger communities in the different micro habitats being disturbed and irrespective of the size, these are mass dislodged and swept into the large net by water current since even the loose substrates in the river are scooped by the kick net screen. On the other hand, (Humphreys et al., 1998) found out that, the abundance of macro taxa collected by basket was far greater than that obtained by airlift or sweep. Mean abundance per sample ranged between 840-3019 for basket as compared to 68- 300 for other samplers like airlift and sweep samplers. This may be attributed to the area sampled and number of microhabitats sampled.

Despite the Kick-net sampling tool, collecting more taxa than Surber samplers based on species-sampling analysis, Stark (1993) in comparing the two sampling tools, found little difference in mean benthic macroinvertebrate taxonomic richness. In our study, the highest taxa richness of macroinvertebrates was associated with kick net at all sampling sites, while the lowest richness was associated with Surber sampler. The highest richness was 11 ± 0.41 families recorded at agricultural site using kick net, while the lowest was 7 ± 0.41 families at the same site using Surber sampler. This is because Surber was disturbed by environmental factors like bankful discharge and water levels rising higher than the sampler, sometimes failing the tool setting on the river's bed hence we ended up sampling fewer microhabitats especially during the wet season.

Conclusions

Based on the RV values obtained from this study, all the three sampling tools are efficient, but none of them is exhaustive in sampling the animals. This explains why despite portraying that the

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Surber was the least effective, it sampled some rare macroinvertebrate taxa that were not sampled or could be missed by the kick net that was considered best tool and rock-filled basket. This therefore means that before the choice of the tool to use is made, the purpose for sampling should be highlighted, otherwise we recommended that more than one tool should be applied for a give study. In comparison of results of our study, rock-filled basket obtained the results that were closely comparable to those of kick net.; We, therefore, concluded that rock-filled basket can either be used together with kick net or as an alternative tool for kick net especially where environmental conditions limit kick net. Of the environmental conditions that could limit the efficiency of kick net is the inaccessibility of riverbed may be due to bankfull discharges, fear of dangerous reptiles like snakes in water.

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