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Akbar A Ganatra

International Centre for Insect Physiology and Ecology <https://orcid.org/0000-0001-7861-0958>

Faith Jebiwot Kandie

Moi University

Ulrike Fillinger

ICIPE: International Centre for Insect Physiology and Ecology

Francis McOdimba

Egerton University - Njoro Campus: Egerton University

Baldwyn Torto

ICIPE: International Centre for Insect Physiology and Ecology

Werner Brack

Helmholtz-Zentrum für Umweltforschung UFZ: Helmholtz-Zentrum für Umweltforschung UFZ

Matthias Liess (✉ matthias.liess@ufz.de)

Helmholtz Centre for Environmental Research GmbH – UFZ. Department of System-Ecotoxicology,
04318 Leipzig, Germany <https://orcid.org/0000-0002-3321-8909>

Henner Hollert

Goethe University Frankfurt: Goethe-Universität Frankfurt am Main

Jeremias Becker

Umweltbundesamt

Research

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Calibration of the SPEAR_{pesticides} bioindicator for cost-effective pesticide monitoring in East African streams

Akbar A. Ganatra^{1,2}, Faith Jebiwot Kandie^{1,3,4,5}, Ulrike Fillinger¹, Francis McOdimba^{1,2}, Baldwyn Torto¹, Werner Brack^{3,5}, Matthias Liess^{6, 7}, Henner Hollert⁷, Jeremias Becker^{6,7}

¹International Centre of Insect Physiology and Ecology (*icipe*), Human Health Theme, 00100 Nairobi, Kenya

²Egerton University, Biological Sciences, 20115 Egerton, Kenya

³Goethe University Frankfurt, Department Evolutionary Ecology and Environmental Toxicology, Institute of Ecology, Evolution and Diversity, Faculty Biological Sciences, 60438 Frankfurt, Germany

⁴Department of Biological Sciences, Moi University, P.O. Box 3900-30100, Eldoret, Kenya.

⁵Department of Effect-Directed Analysis, Helmholtz Centre for Environmental Research (UFZ), Permoserstrasse 15, 04318 Leipzig, Germany.

⁶Helmholtz Centre for Environmental Research GmbH – UFZ. Department of System-Ecotoxicology, 04318 Leipzig, Germany

⁷RWTH Aachen University, Institute for Environmental Research (Biology V), 52074 Aachen, Germany

corresponding author: Matthias Liess: matthias.liess@ufz.de

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Ecotoxicology, Bio-indicator, pesticide pollution

Abstract

Background

Pesticides are washed from agricultural fields into adjacent streams where even short-term exposure causes long-term ecological damage. Detecting pesticide pollution in streams thus requires the expensive monitoring of peak concentrations during run-off events. Alternatively, exposure and ecological effects can be assessed using the SPEAR_{pesticides} bioindicator that quantifies pesticide-related changes in the macroinvertebrate community composition. SPEAR_{pesticides} has been developed in Central Europe and validated in other parts of Europe, Australia and South America; here we investigated its performance in East African streams.

Results

With minimal adaptations of the SPEAR_{pesticides} index, we successfully characterized pesticide pollution in 13 streams located in Western Kenya. The East African SPEAR_{pesticides} index correlated well with the overall toxicity of 30 pesticides (maximum toxic unit = maximum environmental vs. median lethal concentration) measured in stream water ($R^2 = 0.53$). Similarly, the SPEAR_{pesticides} index correlated with the risk of surface run-off from agricultural fields (as identified based on ground slope in the catchment area and the width of protective riparian strips, $R^2 = 0.45$). Unlike other bioindicators designed to indicate general water pollution, SPEAR_{pesticides} was independent of organic pollution and highly specific to pesticides. In 23% of the streams, pesticides exceeded concentrations considered environmentally safe based on European first tiered risk assessment.

Conclusions

Increasing contamination was associated with considerable changes in the macroinvertebrate community composition. We conclude that pesticides need to be better regulated also in developing countries. SPEAR_{pesticides} provides a straightforward and cost-efficient tool for the required monitoring of pesticide exposure in small to medium streams.

Background

In 2020, the worldwide application of agricultural pesticides is expected to increase from 2 million tonnes to 3.5 million tonnes annually (Sharma et al. 2019). Pesticide pollution is considered one of the main drivers for the global decline in the abundance and diversity of insects, plants and birds (Geiger et al. 2010, Beketov et al. 2013, Sánchez-Bayo and Wyckhuys 2019). There is increasing evidence that the pesticide-driven impairment of biocenoses also affects valuable ecosystem services ranging from pollination (Rundlöf et al. 2015) to leaf-litter degradation (Schaefer et al. 2011b) and to the biological control of agricultural pests (Talebi et al. 2008, Roubos et al. 2014) and of pathogens in freshwater (Becker et al. 2020a). For the United States and the European Union, where pesticides are used on a large scale, extensive literature on exposure and effects in the environment is available from academic research and from regulatory risk assessment (Sharma et al. 2019). In developing countries, regulation of plant protection products is often poor and information on pesticide pollution is scarce, though recent evidence suggests that pesticide usage has been increasing (Sharma et al. 2019). As agricultural producers grow more conscious about the use of synthetic pesticides and their toxic effects (Hernández et al., 2013), more effort in the assessment and mitigation of pesticide pollution is urgently needed.

Freshwater macroinvertebrates in small streams are at particular risk, since streams collect pesticide loads from agricultural fields in the catchment area (Münze et al. 2015). Moreover, many freshwater arthropods are highly sensitive to insecticides and fungicides (Von Der Ohe and Liess 2004, Van Dijk et al. 2013, Morrissey et al. 2015). However, the detection and quantification of pesticides in streams is challenging, as exposure occurs typically in short pulses due to spray drift and particularly due to surface run-off from agricultural fields following heavy rainfall (Liess et al. 1999). Such short-term exposure peaks (in the range of hours) drive long-term effects on the macroinvertebrate community for months (Liess and Schulz 1999, Cold and Forbes 2004, Beketov et al. 2008). Therefore, pesticide measurements

from grab samples of stream water and sediment at random time points tend to considerably underestimate the magnitude of pesticide exposure. Realistic environmental monitoring in streams must capture the exposure peaks by run-off event-triggered sampling (Liess et al. 1999) or by continuous passive sampling over extended periods of time (Münze et al. 2015). However, such studies are labor-intensive and expensive (Vrana et al., 2005). In addition to the chemical analysis of samples for many compounds, samplers need to be installed, protected and regularly accessed in remote areas. These challenges limit the feasibility of monitoring pesticide pollution based on chemical analyses, particularly in developing countries.

As an alternative approach, pesticide exposure can be indirectly derived from its observed effects on the macroinvertebrate community composition. The SPEAR_{pesticides} (“SPECies At Risk”) bioindicator has been developed to quantify decreases in the proportion of those taxa considered to be vulnerable to pesticides, as compared to reference conditions (Liess and von der Ohe 2005, Liess et al. 2008). For this task, stream macroinvertebrates have been classified as vulnerable or non-vulnerable taxa based on ecotoxicological traits (see methods). Because the SPEAR_{pesticides} index describes the proportion of vulnerable taxa (weighted by individual number), it has no unit and does not facilitate the identification of individual toxic compounds in the environment. However, SPEAR_{pesticides} values can be translated to an estimated toxic pressure for macroinvertebrates in the more informative form of toxic units (see methods). Since the SPEAR_{pesticides} value is driven by long-term effects, it can be derived from a single community sample per site. SPEAR_{pesticides} thus offers a potential cost-effective technique for the monitoring of pesticide pollution in small to medium streams that may be specifically valuable in developing countries.

SPEAR_{pesticides} has been developed for temperate streams in Central Europe. Thus, the studied species composition, pesticide exposure patterns and ecological conditions may differ from those in other continents such as in sub-Saharan Africa. Malherbe et al. (2018) found only a non-significant response of SPEAR_{pesticides} values to pesticide pollution in South African streams. In contrast, SPEAR_{pesticides} has been successfully applied and validated to assess pesticide pollution in various streams ranging from Southern to Northern Europe (Liess et al. 2008), and (after minor modification) in Australia (Schaefer et al. 2011a) and Argentina (Hunt et al. 2017). The SPEAR_{pesticides} concept is based on ecotoxicological traits rather than taxonomic relations and thus provides a mechanistic linkage of pesticide stress and the community response; in contrast to classical taxonomy-based bioindicators, trait-based approaches can overcome issues with natural taxonomic variability and may thus be applicable even across different climatic regions (Menezes et al. 2010). For freshwater macroinvertebrates in the Southern hemisphere, information on ecotoxicological traits is very scarce (Schaefer et al. 2011a, Malherbe et al. 2018). However, Wang et al. (2014) found that related saltwater invertebrates from temperate and tropical regions differ only slightly in their acute sensitivity to toxicants. Moreover, SPEAR_{pesticides} can be applied at the family taxonomic level (Beketov et al. 2009), and palearctic and afrotropical streams share most of their macroinvertebrate families (Ochieng et al. 2020). Assuming that families of freshwater macroinvertebrates from temperate and tropical streams may generally share their vulnerability to pesticides, SPEAR_{pesticides} may be therefore used also in sub-Saharan Africa.

Here we applied the SPEAR_{pesticides} bioindicator to macroinvertebrate samples from 13 small to medium streams in Western Kenya. The SPEAR_{pesticides} values were compared to the toxic pressure of 30 pesticides in water samples concurrently collected during the rainy season (Kandie et al. 2020). Additionally, we tested the specificity of the SPEAR_{pesticides} index for

pesticide effects and compared its performance to alternative bioindicators for environmental stressors.

Materials and methods

Study area

The Lake Victoria South Basin in the western part of Kenya (Fig. 1) is characterized by a tropical climate with a major rainy season between March and June and a minor rainy season between October and December. The rainy seasons are separated by dry months, particularly in the lowlands close to the shore of Lake Victoria. The area is densely populated, but poorly developed in terms of infrastructure and sanitation. It is dominated by food production, mainly maize, for local markets with small scale fields that often range close to the unfortified banks of streams. However, large scale commercial farming also exists in the form of irrigated rice fields, sugarcane and particularly tea plantations (Wambugu and Muthamia 2009). During the rainy seasons, streams turn red from silt loads indicating heavy erosion and a high potential for surface run-off from agricultural fields.

Figure 1: Location of the sampling sites in the study area of Western Kenya, East Africa. Site locations are displayed with the site code and colour-filled to annotate the relationship between predicted pesticide pollution (TU_{max}) using $SPEAR_{pesticides}$ and measured pollution in water samples using LC-HRMS in Kandie et al. (2020). Akbar Ganatra. “ $SPEAR_{pesticides}$ study sites”. “World Topographical map” & “World Hillshade”. February 2nd, 2021. <https://arcg.is/1fiDn9>.

Pesticide measurements

The sampling and analysis of pesticide residues has been described in detail in Kandie et al. (2020). In brief, 48 sites covering small to large streams, oxbow lakes, irrigation channels, reservoirs and rice fields were sampled during the rainy season in September – October 2017. A single 500 mL water sample per site was collected in a pre-cleaned glass. Solids were allowed to settle for 1 min, before aliquots of 1 mL were transported to the laboratory in amber glass vials at -4°C. Chemical analysis was performed by directly injecting 100 µL of the water sample into a high performance liquid chromatography system (Thermo Ultimate 3000 LC) coupled to a high resolution mass spectrometer (QExactive Plus, Thermo). The water samples were subjected to target screening for 428 chemicals and to suspect screening for 233 additional substances. This analysis included 162 hydrophilic pesticides (active substances and metabolites), but no highly hydrophobic compounds such as pyrethroid insecticides for which different sampling techniques are required. Raw data was processed using MZmine (Version 2.38, Pluskal et al., 2010) and further confirmation and quantification done using TraceFinder 4.1 (Thermo) as detailed in Kandie et al. 2020. Physicochemical water parameters were measured *in situ* (flow velocity, temperature, dissolved oxygen, phosphates, pH, turbidity) and from additional water samples in the laboratory (nitrate, nitrite, ammonium, carbonate hardness).

In the present study, we only considered pesticides and converted the observed concentrations in the water samples to toxic units in order to quantify the toxic pressure for freshwater macroinvertebrates. The toxic unit (TU; Sprague 1970) describes the environmental concentration *conc* of a pesticide *i* in relation to its median effective or lethal concentration (EC_{50} or LC_{50} , concentration that affects or kills 50 % of individuals in an acute toxicity test)

Eq 1:

$$TU_i = \frac{conc_i}{LC_{50_i}}$$

For the TU calculation we generally referred to the LC_{50} for the water flea *Daphnia magna* after constant exposure for 48 h as reported in the Pesticides Properties Data Base (PPDB, 2019). *D. magna* was selected as a reference because it belongs to the more sensitive freshwater macroinvertebrates and because most toxicological data are available for this species due to its use in regulatory risk assessment (Brock and Van Wijngaarden 2012). However, for some pesticides (particularly neonicotinoid insecticides), *D. magna* turned out to be considerably more tolerant than other macroinvertebrates (Brock and Van Wijngaarden 2012). Therefore, we additionally extracted the LC_{50} for the saltwater shrimp *Americamysis bahia* after 96 h exposure from the PPDB data base in case it was available. In cases where *D. magna* was highly insensitive ($LC_{50} > 10$ mg/L), we additionally took data from the ECOTOX data base (ECOTOX 2019) for the standard test species *Chironomus riparius* and *Hyalella azteca*. If the LC_{50} of *D. magna* was > 10 times higher than that of an additional species observed after 48 h or > 100 times higher after 96 h, we used the most sensitive additional species as reference (mean LC_{50} from different studies in ECOTOX, if available). For metabolites we used the LC_{50} of the parental compound if no toxicity data was available or if the metabolite was less toxic than the parental compound.

The overall toxic pressure from pesticide mixtures in the water samples was summarized as the summed up (TU_{sum} , Sprague 1970) and as the maximum (TU_{max} , Liess and von der Ohe 2005)

toxic unit. TU_{max} and TU_{sum} were set to a minimum threshold of 10^{-4} for sites at which no potentially relevant concentrations were detected; this threshold is in the range of the limit of quantification for the most toxic pesticides in the chemical analysis (see supplementary material). Additionally, no effects of pesticides on freshwater macroinvertebrate communities have been generally observed in the field below a threshold range of TU_{max} between 10^{-3} and 10^{-4} (Schaefer et al. 2012).

The $SPEAR_{pesticides}$ concept

The $SPEAR_{pesticides}$ bioindicator has been developed to specifically quantify pesticide-driven deviations in the community structure from those observed under non-polluted reference conditions. Since its first publication in Liess and von der Ohe (2005), $SPEAR_{pesticides}$ has been constantly refined (Liess et al. 2008, Beketov et al. 2009). Here we refer to the latest version 2019.11 that includes some major improvements described in Knillmann et al. (2018).

$SPEAR_{pesticides}$ is based on a classification of freshwater macroinvertebrates in "Species At Risk" and in taxa being not at risk based on four traits that describe their vulnerability to pesticides: 1.) the average physiological sensitivity to various pesticides (s -value, mean of $\log_{10}(LC_{50})$ relative to the LC_{50} of the reference species *D. magna* or *Chironomus sp.*); 2.) the ability of autochthonous population recovery from reproduction (generation time); 3.) the ability of allochthonous recovery from recolonization (dispersal from non-polluted refuge areas); and 4.) the probability of being actually exposed (e. g. aquatic life stages during the main insecticide application season). Taxa with high sensitivity, low ability of autochthonous and allochthonous recovery and high probability of being exposed have been classified as being at risk, others as not at risk. A trait data base for Central European taxa, linked to a program for the calculation of $SPEAR_{pesticides}$ values is available with the software INDICATE (<http://www.systemecology.eu/indicate>). The data base comprises entries for individual species, but also for higher taxa where available data on lower taxonomic levels have been averaged.

The $SPEAR_{pesticides}$ index relates to the proportion of taxa at risk within the macroinvertebrate community and decreases with increasing toxic pressure (Knillmann et al. 2018):

Eq 2:

$$SPEAR_{pesticides} = \frac{\sum_{i=1}^n \log_{10}(4x_i + 1) \cdot y}{\sum_{i=1}^n \log_{10}(4x_i + 1)}$$

with x_i being the observed number of individuals of taxon i , and y being 1 for those taxa classified at risk and 0 for taxa not at risk. In this formula, $SPEAR_{pesticides}$ values can range between 0 (no species at risk, indicating high pesticide effects) and 1 (only species at risk). Abundant taxa are down weighted to limit the influence of populations with mass development so that the $SPEAR_{pesticides}$ index increases rather equally with both the incidence and the population density of vulnerable taxa. In order to facilitate interpretation, since version 2019.11, "raw" $SPEAR_{pesticides}$ values obtained from the equation above are divided by the average $SPEAR_{pesticides}$ value observed in non-polluted reference sites (at measured $TU_{max} < 10^{-4}$) from Germany (Knillmann et al. 2018):

Eq 3:

$$scaled\ SPEAR_{pesticides} = \frac{raw\ SPEAR_{pesticides}}{reference\ SPEAR_{pesticides}}$$

With $reference\ SPEAR_{pesticides} = 0.27$. This scaled $SPEAR_{pesticides}$ index ranges from 0 (no species at risk) to 3.7 (only species at risk) and indicates no toxic pressure at values > 1 . For European streams, an empirical relationship between $SPEAR_{pesticides}$ and the measured $\log_{10}$ -transformed TU_{max} during run-off events has been established to convert $SPEAR_{pesticides}$ values to the estimated toxic pressure (Liess et al. 2008, Knillmann et al. 2018). Because the community composition of Kenyan freshwater macroinvertebrates may differ from those in Germany, both the classification of individual taxa and the conversion to TU_{max} were subjected to revision in the present study (see results).

Application of $SPEAR_{pesticides}$ in the study area

We applied $SPEAR_{pesticides}$ to macroinvertebrate samples collected together with the water samples from Kandie et al. (2020, see above). The macroinvertebrate sampling has been described in detail in Becker et al. (2020a). In brief, macroinvertebrates were collected at the same site and day as the water samples, following a standardised approach adapted from the South African Scoring System 5 (SASS5, Dickens and Graham 2002). At each stream site, a 50 m stretch was sampled in four quadrants by two persons in parallel. Each quadrant was sampled for 7 minutes using sweep nets for the water surface and littoral habitats, and kick sampling for the benthic macroinvertebrates. Additionally, gravel, soil and mud (GSM) habitats were sampled for 1 minute per quadrant. Organisms were preserved in 70 % ethanol and identified to the family level under a dissecting microscope in the laboratory. $SPEAR_{pesticides}$ V. 2019.11 and the associated estimated TU_{max} were calculated using the software INDICATE v. 1.2.0 (<http://www.systemecology.eu/indicate>).

Application of other indicators of freshwater pollution

In order to compare the performance and specificity of $SPEAR_{pesticides}$ in the identification of pesticide pollution, we additionally applied the Biological Monitoring Working Party (BMWP) and the South African Scoring System (SASS5) bioindicators for freshwater quality. In contrast to $SPEAR_{pesticides}$, the BMWP and SASS5 indicators are based on a gradual instead of a binary classification system for the sensitivity of freshwater macroinvertebrates (mainly to low oxygen levels) ranging from 1 to 10. Like $SPEAR_{pesticides}$, the indicators have been established for freshwater families from a different region, but have been also applied in sub-Saharan Africa beyond their countries of origin (Bere and Nyamupingidza 2014, Ochieng et al. 2020)

The BMWP score system was developed in the 1980s and refined later to assess organic pollution in British streams (Paisley et al. 2014). For a refined assessment according to the European Water Frame Work Directive, ecological quality indices are derived from the BMWP score by comparing observed vs. expected values based on an elaborate stream classification system. Such a classification system is not available for Kenyan streams; therefore, we simply calculated the average score per taxon (ASPT) using the software ASTERICS – AQUEM/STAR Ecological River Classification System v. 4.0.4, (https://www.gewaesserbewertung.de/index.php?article_id=419&clang=0). SASS5 was originally developed in 1994 and refined in 2002 to determine the condition or ‘health’ of rivers in South Africa (Dickens and Graham 2002). As recommended, we calculated the Average Score Per Taxa (ASPT) using the score tables available in Dickens and Graham (2002).

Finally, we calculated the proportion of ephemeropteran, plecopteran and trichopteran insects on the overall individual number (EPT) and the species diversity (Shannon index) as more general descriptors of the freshwater macroinvertebrate community. Both descriptors are considered to decrease with increasing levels of water pollution (Knillmann et al. 2018).

Additionally we estimated the run-off potential of a site, i. e. a predictor for the risk of pesticide exposure from surface run-off (Schriever et al. 2007). We used a highly simplified approach by classifying sites on an ordinal scale based on the average local ground slope and the average width of riparian strips up to 2 km upstream as visible from online satellite imagery. Slope was classified as flat, low, medium and high. The run-off potential was then classified as follows: 1 (none): ≥ 30 m buffer strip, or ≥ 20 m buffer strip and flat or low slope; 2 (low): 21 – 30 m buffer strip, or 11 – 20 m buffer strip and flat or low slope; 3 (moderate): 11 – 20 m buffer strip, or 6 – 10 m buffer strip and flat or low slope; 4 (high): ≤ 10 m buffer strip, or ≤ 5 m buffer strip and flat or low slope.

Data analysis

Data analysis was performed using R version 3.6.2. Because SPEAR_{pesticides} has been developed for small and medium streams with flowing water, we limited our analysis to samples from natural streams (no artificial channels) with an average width of < 20 m and an estimated average flow velocity > 1 cm/s (based on the drift-body method). Two sites from a small river close to Kisii (PS42 and PS43, see supplementary information) were excluded because biological sampling took part during high water. These samples were characterized by low numbers of taxa and individuals, as well as unusually high pesticide concentrations as compared to the SPEAR_{pesticides} values, suggesting that sampling took place during a run-off event. Therefore, we considered neither the SPEAR_{pesticides} nor the pesticide data from this stream comparable to the other sites. Altogether, our analysis included 16 sites from 13 different streams. Three of the streams were sampled twice, with an average distance of ca. 2 km between both sites. Macroinvertebrate samples and toxic units from sites of the same stream were considerably more similar than those from different streams; to avoid pseudo-replication we therefore aggregated data from different sites of the same stream using the mean values.

The original and the revised SPEAR_{pesticides} index, as well as the associated estimated TU_{max} were compared to the TU_{max} from pesticide measurements using one-way linear regression; TU_{max} was log₁₀-transformed prior to the analyses. To assess the potential disturbance of the sampling sites by additional stressors other than pesticides, we investigated the distribution of the measured TU_{max} and physicochemical water parameter values using violin plots; the data were compared to thresholds for a good ecological status from the literature. To assess the performance and the specificity of SPEAR_{pesticides} for pesticides, we analyzed responses of the SPEAR_{pesticides} index and of the additional bioindicators and descriptors to the physicochemical parameters using one-way linear regression. Additionally, relations of the SPEAR_{pesticides} index and those physicochemical parameters that had a (marginally) significant effect on the SPEAR_{pesticides}, BMWP or SASS5 bioindicator were summarized using a principal component analysis (PCA). Finally, we performed pairwise correlations among the bioindicators and among those environmental variables that significantly affected these bioindicators in order to identify confounding factors.

Results

Pesticide pollution and effects on the macroinvertebrate community

We observed considerable pesticide toxicity to freshwater macroinvertebrates in our 16 study sites from 13 streams, quantified as the maximum toxic unit derived from chemical water analyses (TU_{max} , see methods). TU_{max} exceeded the threshold range for environmental effects of 10^{-4} to 10^{-3} (Schaefer et al. 2012) in nine and eight streams, respectively. The results indicate that pesticide pollution was a relevant environmental stressor in the study area. Pesticide toxicity was driven by the insecticides diazinon (7 sites, $TU = 10^{-2.36} - 10^{-1.81}$), pirimiphos-methyl (3 sites, $TU = 10^{-1.86} - 10^{-1.21}$), bendiocarb (1 site, $TU = 10^{-2.54}$) and imidacloprid (1 site, $TU = 10^{-3.47}$).

Overall, we identified 35 macroinvertebrate families in the streams. The proportion of taxonomic groups changed markedly with increasing pesticide toxicity (Fig. 2). Non- or marginally polluted sites with $TU_{max} < 10^{-3}$ were dominated by the ephemeropteran families *Baetidae* and *Heptageniidae*, the zygopter family *Coenagrionidae*, the heteropter families *Gerridae* and *Veliidae*, and by the coleopteran family *Gyrinidae*. These families occurred in at least half of the low-polluted streams and also showed the highest proportions on the mean number of individuals collected in a low-polluted stream (relative abundances). The same families occurred also in at least half of the highly polluted streams with $TU_{max} \geq 10^{-3}$, but additionally the ephemeropteran family *Caenidae*, the anisopteran family *Libellulidae*, the heteropter families *Naucoridae* and *Nepidae*, and snails of the family *Planorbidae* were observed in the majority of these streams. As the most prominent changes in the community composition, the mean relative abundance of *Baetidae* decreased from 56 % (range: 7 – 94 %) in lowly polluted streams to 10 % (range: 0.2 – 38 %) in highly polluted streams ($n = 5$ lowly and 8 highly polluted sites, $\chi^2 = 8.29$, $p = 0.004$ using a binomial generalized linear model). At the same time, *Planorbidae* increased from 0 % to 9 % (range: 0 – 36 %, $\chi^2 = 7.95$, $p = 0.005$) in highly polluted streams. Therefore, changes in the community composition with pesticide pollution were driven by the response of frequently occurring taxa, suggesting that $SPEAR_{pesticides}$ may perform consistently across random samples from different streams. The results are consistent with Reiber et al. (2020) who identified mayflies and snails among those macroinvertebrate taxa that most strongly decrease or increase with pesticide pollution in European streams, respectively.

Figure 2: The freshwater macroinvertebrate community composition changes in streams with high pesticide pollution. The stacked bars show the mean proportions of macroinvertebrate families on the overall number of individuals observed in lowly-polluted (maximum toxic unit $TU_{max} < 10^{-3}$) and in highly polluted streams ($TU_{max} \geq 10^{-3}$). The number of individuals ranged from 32 to 154 (mean: 94) in the 5 lowly polluted streams and from 37 to 462 (mean: 152) in the 8 highly polluted streams. Families that were recorded in at least 50 % of the streams are shown in bold. Taxa shown in blue have been classified as being at risk in the adapted $SPEAR_{pesticides}$ indicator for East African streams, taxa shown in red have been classified not at risk.

Adaptation of $SPEAR_{pesticides}$ to East African streams

Most of the 35 macroinvertebrate families sampled were automatically linked to existing taxa in the trait database provided in the software INDICATE (see Tab. S2 in the supplementary material). Only three families were not existing in the database because they do not occur in Central European streams; these families were manually linked to higher taxa available in the database: *Pisuliidae* was linked to Trichoptera Gen. sp., *Oligoneuridae* was linked to

Ephemeroptera Gen. sp., and *Chlorocyphidae* was linked to Zygoptera Gen. sp. *Culicidae* was automatically linked to Diptera Gen. sp., but traits are missing for this taxon (due to a lack of data because dipteran species are too heterogeneous to be aggregated), and they were therefore automatically excluded from the calculation of $SPEAR_{pesticides}$.

Though developed for European streams, the existing $SPEAR_{pesticides}$ index v. 2019.11 from Knillmann et al. (2018) a showed reasonable correlation with the measured pesticide toxicity (TU_{max}) from chemical water analysis when applied to the Kenyan macroinvertebrate communities ($R^2 = 0.43$, Fig. 3a). The correlation of the $SPEAR_{pesticides}$ index with toxic pressure increased when we re-classified two taxa based on additional information for Kenyan taxa. First, the damselfly family of *Coenagrionidae* has been classified at risk in the European $SPEAR_{pesticides}$ index v. 2019-11, based on a relatively high physiological insecticide sensitivity (s -value = -0.24). This value is only slightly above the threshold for sensitive taxa ($s \geq -0.36$). The classification was based on toxicity data for the European species *Eschnura elegans*. However, acute toxicity tests with the insecticides diazinon and imidacloprid on freshwater macroinvertebrates collected in the study region of Western Kenya revealed that the tolerance of local coenagrionid species is comparable to those of other taxa such as *Notonectidae*, *Chironomidae* and *Dytiscidae* that have been classified as insensitive (Becker et al. 2020a). Accordingly, we re-classified *Coenagrionidae* as being insensitive (s -value = -0.4); consequently, the taxon moved from the category “at risk” to “not at risk”.

Second, in $SPEAR_{pesticides}$ v. 2019.11, the heteropterid family *Corixidae* was considered as non-exposed due to a surrounding layer of air (physical lung) that potentially protect individuals from pesticide exposure in the water. However, in acute toxicity tests we observed high insecticide sensitivity of *Corixidae* from the study region (Becker et al. 2020a). Thus, we re-classified *Corixidae* as being exposed, so that the taxon moved from the category “not at risk” to “at risk”. Results of toxicity tests with additional 13 taxa collected in the study region from Becker et al. (2020a) were in accordance with the existing classification in the $SPEAR_{pesticides}$ trait data base.

With these changes, we were able to adapt the $SPEAR_{pesticides}$ index to East African streams so that it explained the observed TU_{max} considerably better than the European $SPEAR_{pesticides}$ index v. 2019.11 ($R^2 = 0.53$, Fig. 3b). The East African $SPEAR_{pesticides}$ index decreased with increasing measured pesticide toxicity as follows:

Eq. 4:

$$SPEAR_{pesticides} = -0.43 * \log_{10}(\text{background } TU_{max}) - 0.02$$

After solving equation 4 for TU_{max} , the East African $SPEAR_{pesticides}$ index may be used to predict background toxicity in streams from the observed macroinvertebrate community composition:

Eq. 5:

$$\text{background } TU_{max} = 10^{\frac{SPEAR_{pesticides} + 0.02}{-0.43}}$$

Figure 3: SPEAR_{pesticides} indicates pesticide pollution in tropical streams of Western Kenya. Pesticide pollution was quantified as the maximum toxic unit (TU_{max} , restricted to $\geq 10^{-4}$) out of 30 pesticides measured in water samples collected during the rainy season but not during peak exposure. Freshwater macroinvertebrates were identified to the family level. (a) Application of the European SPEAR_{pesticides} index v. 2019.11 from Knillmann et al. (2018) to the Kenyan samples; $R^2 = 0.43$, $F = 8.15$, $df = 1$, $res. df = 11$, $p = 0.016$, intercept = 0.62, slope = -0.37. (b) Application of the SPEAR_{pesticides} index after adaption to East African streams. $R^2 = 0.53$, $F = 12.35$, $df = 1$, $res. df = 11$, $p = 0.005$, intercept = -0.02, slope = -0.43.

The mean TU_{max} predicted from equation 5 and the mean observed background TU_{max} coincided well (difference by a factor of < 2.5 or of ≤ 0.4 orders of magnitude ($= \log_{10}$ -transformed TU_{max}) across the whole range of observed toxicity, Fig. 4a). Across all data points, the ratio of estimated vs. measured TU_{max} ranged from 0.2 to 19.6, i. e. the predicted toxicity deviated from the measured toxicity by < 1.3 orders of magnitude in all investigated streams.

We related the East African SPEAR_{pesticides} index to background pesticide toxicity measured in water from grab samples collected during the rainy season, but outside run-off events. In contrast, in Europe the SPEAR_{pesticides} index has been related to the short-term peak toxicity measured during run-off events following heavy rainfall (Liess and von der Ohe 2005, Liess et al. 2008, Knillmann et al. 2018). In temperate streams of Europe and North America, peak pesticide toxicity is approximately four times as high as the background toxicity during the season of main pesticide exposure in early summer (Williams et al. 1995, Kreuger 1998, Liess et al. 1999, Schäfer et al. 2008, Münze et al. 2017). Assuming that the same ratio of peak vs. background toxicity may hold also in tropical streams, we applied a correction factor of 4 to relate the East African SPEAR_{pesticides} index to an estimated peak exposure during run-off:

Eq. 6:

$$\text{estimated peak } TU_{max} = 4 * 10^{\frac{SPEAR_{pesticides} + 0.02}{-0.43}}$$

The response of the East African SPEAR_{pesticides} index to the estimated peak exposure in Kenya could then be compared to the response of the latest European SPEAR_{pesticides} index v. 2019.11

to peak exposure in Germany (Knillmann et al. 2018). The East African $\text{SPEAR}_{\text{pesticides}}$ index produced considerably higher numbers as compared to the European index, particularly when pesticide toxicity was low (Fig. 4b). The more pronounced increase in the East African $\text{SPEAR}_{\text{pesticides}}$ index with decreasing pesticide exposure indicates differences in the composition of European and East African macroinvertebrate communities and in their response to pesticides. Therefore, the use of a separate conversion scheme from $\text{SPEAR}_{\text{pesticides}}$ to TU_{max} in afrotropical streams, as established with equation 5 and 6, is justified.

To assess the reproducibility and robustness of the East African $\text{SPEAR}_{\text{pesticides}}$ index, we extended our data set to all sites sampled in the study area of Western Kenya (see Becker et al. 2020a). The 48 sites from 40 water bodies included habitats for which $\text{SPEAR}_{\text{pesticides}}$ was not designed such as artificial irrigation channels, rice fields, reservoirs, oxbow lakes and large rivers. We still observed a relation of the East African $\text{SPEAR}_{\text{pesticides}}$ index with TU_{max} that was very similar to those in Fig. 3b, but the non-explained variation increased considerably ($R^2 = 0.13$, $F = 5.55$, $\text{df} = 1$, $\text{res. df} = 38$, $p = 0.024$, intercept = 0.13, slope = -0.31).

Figure 4: Performance of the East African $\text{SPEAR}_{\text{pesticides}}$ index. (a) Background pesticide toxicity (maximum toxic unit, TU_{max}) predicted with the East African $\text{SPEAR}_{\text{pesticides}}$ index correlates with the measured background toxicity in Kenyan water samples. $R^2 = 0.54$, $F = 12.97$, $\text{df} = 1$, $\text{res. df} = 11$, $p = 0.004$, intercept = -0.41, slope = 0.80. TU_{max} was restricted to $\geq 10^{-4}$, because lower toxicity could not be measured due to detection limits for pesticides. TU_{max} has been $\log_{10}$ -transformed, so that differences are presented in orders of magnitude. (b) Relation of the East African $\text{SPEAR}_{\text{pesticides}}$ index with the estimated peak pesticide toxicity during run-off in Kenyan streams, as compared to the relation of the European $\text{SPEAR}_{\text{pesticides}}$ index v. 219.11 with measured peak toxicity in German streams (data from Knillmann et al. 2018). Peak exposure in Kenyan streams was estimated by multiplying the measured background toxicity with a correction factor of 4 (see main text for justification).

Specificity of the East African SPEAR_{pesticides} index to pesticides

To assess the potential impact of confounding factors and the specificity in the response of the adapted East African SPEAR_{pesticides} index to pesticide pollution, we first investigated the potential influence of additional physicochemical parameters on the macroinvertebrate community. A comparison of the observed physicochemical water parameters with values recommended for good water quality from the literature revealed that pesticide pollution was indeed one of the dominant stressors at the sampling sites (Fig. 5). Additionally, in many streams we observed very high levels of phosphate and turbidity, as well as low levels of carbonate hardness and dissolved oxygen, which may have contributed in shaping the macroinvertebrate community.

Figure 5: Distribution of physicochemical parameter values at the study sites. The violin plot shows the kernel probability density of the data points across the parameter values. White points indicate the median, black boxes the interquartile range, and black lines 1.5 x the interquartile range. Light-grey boxes indicate the range of values considered typical or recommend for streams with good water quality.

Next, we assessed the sensitivity of the adapted $\text{SPEAR}_{\text{pesticides}}$ index for East Africa to such additional stressors. The $\text{SPEAR}_{\text{pesticides}}$ index decreased significantly not only with increasing pesticide toxicity (measured TU_{max} and TU_{sum}) and run-off potential, but also with carbonate hardness, conductivity and turbidity (Tab. S1 in the supplementary material). However, pesticide toxicity was significantly correlated with each of these confounding factors (Tab. S2 in the supplementary material). All these factors were associated with a dominant environmental gradient identified by the first principal component in a principal component analysis (PCA). This gradient explained 61 % of the total variation and likely reflects a range of different stream types (Fig. 6). On one end, lowly polluted, fast-flowing mountain streams showing high $\text{SPEAR}_{\text{pesticides}}$ values (streams nr. 27, 28, 31 and 32) were running in comparably steep valleys through intensely cultivated tea plantations but were protected from run-off by wide buffer strips so that run-off potential and pesticide toxicity was low. On the other end, lowland streams were slowly flowing and showed higher run-off potential due to surrounding agriculture with only small buffer strips. These streams showed high pesticide toxicity, as well as sediment (turbidity) and phosphate pollution that were associated with low $\text{SPEAR}_{\text{pesticides}}$ values and high conductivity, respectively.

Figure 6: Principal component analysis of the East African $\text{SPEAR}_{\text{pesticides}}$ index and those environmental variables that (marginally) significantly correlated with any of the investigated bioindicators. The first principal component (horizontal axis) explains 60.7 % of the overall variation and was considered to represent a gradient of stream types. The second principal component (vertical axis) additionally explains 14.7 % of the overall variation and was considered to represent a gradient

of high water level. Each point represents a stream (identified by its number, see Tab. S4 in the supplementary material), colors refer to the average SPEAR_{pesticides} value obtained from each stream.

Though all these confounding stressors increased simultaneously with the first principal component (considered as stream type), they could be disentangled along the second principal component (Fig. 6): On one end, we observed slow flowing streams with clear water but high conductivity and carbonate hardness. Streams on the other end showed high flow velocity, turbidity and phosphate concentrations, but low conductivity and carbonate hardness. We consider the second principal component to represent a gradient of raised water level at the day of sampling. Raised water levels may have increased flow velocity, as well as turbidity and phosphate levels due to erosion, and decreased conductivity and carbonate levels due to dilution with rainwater. In contrast, neither run-off potential, nor pesticide toxicity (TU_{max}) or SPEAR_{pesticides} responded to this potential gradient of water levels. First, run-off requires not only high water but also nearby agricultural fields. Second, raised water does not mean flooding in this context, which might have been associated with pesticide pollution from run-off (flooded sites were excluded from the analysis, see methods). The SPEAR_{pesticides} index was thus most closely associated with pesticide toxicity and run-off potential, indicating indeed a high specificity of the adapted SPEAR_{pesticides} indicator for pesticide pollution in Western Kenyan streams. Each of the higher principal components explained only $\leq 8\%$ of the total variance, and they were thus not further analyzed.

Finally, we compared the performance of SPEAR_{pesticides} in indicating pesticide pollution to those of other bioindicators and commonly applied descriptors of the macroinvertebrate community. Pesticide toxicity most strongly correlated with the East African SPEAR_{pesticides} index ($R^2 = 0.53$), followed by the European SPEAR_{pesticides} index ($R^2 = 0.43$) and the average score per taxon (aspt) of the BMWP indicator ($R^2 = 0.33$, Tab. S1). Correlation with the aspt of the SASS5 indicator was lower ($R^2 = 0.19$) and not significant. In contrast to SPEAR_{pesticides} that was only related to pesticides and their associated stressors (see above), the BMWP and SASS5 indicators were additionally related to phosphate pollution and temperature but did not significantly correspond to run-off potential. The EPT index (cumulative proportion of *Ephemeroptera*, *Plecoptera* and *Trichoptera*) showed a significant but low negative response to pesticide pollution ($R^2 = 0.10$), turbidity and carbonate hardness, but increased with flow velocity. The Shannon index for species diversity increased with pesticide pollution, but the response was only marginally significant and may relate to the generally concurrent increase in species richness and pesticide pollution from the spring to more downstream sections (Minshall et al. 1985).

The different bioindicators correlated with each other. The East African SPEAR_{pesticides} index increased with the aspt of the BWMP and SASS5 indicator and with the EPT index, but decreased with species diversity (Tab. S2). BMWP and SASS5 were clearly correlated with each other, but not with the EPT index and species diversity. The EPT index decreased with increasing species diversity.

Discussion

We adapted the SPEAR_{pesticides} bioindicator for the quantification of pesticide exposure to afrotropical conditions and demonstrated its use in thirteen Kenyan streams. A detailed step-by-step guidance for the application of the East African SPEAR_{pesticides} index is provided in the

supplementary material. In the following, we discuss the performed modifications of SPEAR_{pesticides} and the results from the present case study.

Pesticide pollution in Western Kenya

Pesticides are an important stressor to freshwater macroinvertebrates in small and medium streams of western Kenya. Our results confirm earlier conclusions from various freshwater habitats of the same study area (Kandie et al. (2020); in that study, the chemical freshwater pollution was assessed and the ecological risk (TU_{sum}) identified was highest for macroinvertebrates due to insecticide exposure. Pesticide toxicity observed in streams of the present study was comparable to those in European landscapes characterized by intensified agriculture (Becker et al. 2020b). In three out of 13 streams, we observed toxic units exceeding the threshold of 10^{-2} that is considered safe according to the first tier of governmental risk assessment in the European Union (EFSA PPR 2013). The results illustrate a need for the monitoring and regulation of pesticide application in order to reduce pesticide exposure in freshwater. In Kenya, plant protection products are sold at relatively low prices without the need for a certificate of competence from retailers, making them widely available to small farmers who are then not informed of the necessary precautions needed to comply to the proposed environmentally safe use. This includes products containing active substances that have been banned in many high income countries, such as the non-selective insecticide diazinon that can be applied on a wide range of crops (<https://ec.europa.eu/food/plant/pesticides/eu-pesticides-database/>) and was driving toxicity in most of our sampling sites.

Adaptation of SPEAR_{pesticides} to East African streams

Effects on the macroinvertebrate community composition were successfully quantified even with the non-modified SPEAR_{pesticides} indicator v. 2019.11 developed for Europe. SPEAR_{pesticides} values decreased approximately log-linearly with increasing pesticide toxicity when the maximum toxic unit (TU_{max}) ranged from 10^{-4} to $10^{-1.5}$. At lower TU_{max} values SPEAR_{pesticides} leveled off, confirming a threshold of 10^{-4} to 10^{-3} TU_{max} below which no effects on freshwater macroinvertebrates have been observed (Schaefer et al. 2012).

Reclassification of the families *Coenagrionidae* and *Corixidae* based on toxicity tests with Kenyan species (Becker et al. 2020a) improved the correlation of SPEAR_{pesticides} with TU_{max}. We suggest determining whether such a reclassification may increase the link between toxicity and invertebrate community composition also in European streams. SPEAR_{pesticides} values from both the European and the adapted East African index were considerably higher in Kenya than those observed in Central European streams (Knillmann et al. 2018), particularly when pesticide toxicity was low. We speculate that in East African streams, the natural proportion of non-vulnerable taxa may be lower. Indeed, macroinvertebrate communities in most Central European streams are dominated by amphipod crustaceans of the genus *Gammarus* sp., and by mayflies of the *Baetis rhodani* / *vernus* group (Becker and Liess 2017). Both groups are considered non-vulnerable to pesticides and often contribute to more than 50 % of individuals. In contrast, amphipod crustaceans were missing in Kenya, confirming previous observations e. g. from Elias et al. (2014). Additionally, we classified mayfly families observed in Kenya as vulnerable (confirmed by a strong decrease with increasing pesticide toxicity, and consistent with most other, less abundant European mayflies). Hence, SPEAR_{pesticides} values in East Africa are associated to higher levels of pesticide toxicity than similar SPEAR_{pesticides} values in Europe, and we adapted the link between the East African SPEAR_{pesticides} index and TU_{max} accordingly.

Using the East African SPEAR_{pesticides} index, the measured pesticide toxicity (TU_{max}) could be predicted with a precision of 1.3 orders of magnitude in each of the streams. This variation covered not only uncertainties in the SPEAR_{pesticides} approach, but also in the measurement of pesticide concentrations and in their conversion to toxic units. Considering the generally high levels of variability and uncertainty in ecotoxicology, as reflected by an assessment factor of 100 (2 orders of magnitude) in first tier European risk assessment (EFSA PPR 2013), results from chemical analyses and from the application of SPEAR_{pesticides} coincided reasonably well.

Specificity of SPEAR_{pesticides} for effects of pesticides

The overall variability in values of the East African SPEAR_{pesticides} index was explained to 53% by the measured pesticide toxicity, and to 45% by the estimated run-off potential based on catchment slope and width of buffer strips. The estimated run-off potential was also closely associated with pesticide toxicity and thus provides a fast and simple method for the identification of potential sampling sites of interest, and for the verification of calculated SPEAR_{pesticides} values: Sites where the estimated pesticide toxicity based on SPEAR_{pesticides} does not fit to the estimated run-off potential may be heavily affected by additional stressors and should be investigated further.

Additional stressors that affected the East African SPEAR_{pesticides} index included carbonate hardness, conductivity and turbidity. All these confounding factors increased with pesticide toxicity along a gradient of different stream types but could be disentangled along a second, independent gradient of increasing water levels. While carbonate hardness, conductivity and turbidity varied with water levels presumably due to erosion and dilution, the SPEAR_{pesticides} index, pesticide pollution and run-off potential did not, because raised water levels alone do not increase run-off without nearby agricultural fields. Thus, our results show that indeed pesticide toxicity and not confounding factors is driving the SPEAR_{pesticides} index.

In contrast to SPEAR_{pesticides}, other bioindicators for the assessment of freshwater quality have not been designed to specifically indicate effects of pesticides (Dickens and Graham 2002, Paisley et al. 2014). As expected, the BMWP and SASS5 scoring system therefore responded to a broader range of stressors including phosphate and sediment pollution that may be associated with oxygen depletion. Similarly, the EPT index and the Shannon index for species diversity most strongly responded to stressors other than pesticides.

Our application of SPEAR_{pesticides} is not the first attempt to establish or apply bioindicators for the assessment of freshwater quality in East Africa (Masese et al. 2009, Shimba and Jonah 2016, Ochieng et al. 2020). These case studies illustrate the growing interest in the use of bioindicators for freshwater monitoring but did not explicitly consider effects of a specific stressor such as pesticide pollution. Apart from our study, the only application of SPEAR_{pesticides} in sub-Saharan Africa we are aware of has been described in Malherbe et al. (2018). The authors applied a previous version of the European SPEAR_{pesticides} index and the Australian SPEAR_{pesticides} index (Schaefer et al. 2011a) to macroinvertebrate samples from the Crocodile River and the Harts River in South Africa. The sampling sites were located upstream, adjacent to and downstream of two large irrigation schemes. The SPEAR_{pesticides} index decreased with increasing estimated pesticide toxicity ($R^2 = 0.26$) but the correlation was not significant.

It should be noted that Malherbe et al. (2018) used a very coarse toxicity estimation based on the sampling site location, assuming that pesticide pollution increases from upstream to downstream of the irrigation system. Toxicity estimation thus did not consider pesticide input

from outside the irrigation scheme, whereas our data show that subsistence farming may considerably contribute to pesticide pollution. Therefore, the poor performance of SPEAR_{pesticides} in Malherbe et al. (2018) may be partly related to uncertainties in the assessment of pesticide exposure. Additionally, the authors sampled macroinvertebrates from large rivers, whereas SPEAR_{pesticides} has been developed for small to medium streams. When we tested the East African SPEAR_{pesticides} index with a larger data set of unsuitable habitats, our conversion scheme from SPEAR_{pesticides} values to pesticide toxicity turned out to be principally robust despite the small sample size used for development. However, the unexplained variance considerably increased, illustrating the importance to consider that the applicability of SPEAR_{pesticides} is limited to small and medium streams with flowing water and no heavy streambed degradation (Liess and von der Ohe 2005).

Conclusions

As shown, high impact of pesticides on freshwater organisms is not limited to regions with intensified commercial agriculture. Widespread pesticide pollution in Western Kenyan streams and the associated decline in vulnerable macroinvertebrates indicate an ecological risk also in areas dominated by subsistence farming. Potential negative effects on species diversity and on ecosystem services such as leaf-litter degradation and biological pathogen control illustrate the need to improve the risk management of pesticides also in developing countries. Monitoring is essential in this respect to identify hot spots of pesticide pollution for the targeted development of mitigation measures, and to evaluate the effectiveness of actions that have been taken. We adapted the SPEAR_{pesticides} bioindicator for the quantification of pesticide exposure in streams of East Africa. This tool provides a cost-efficient alternative to the complex sampling and analysis of chemicals and thus may facilitate large scale monitoring with limited resources in developing countries.

List of abbreviations

BMWP – Biological Monitoring Working Party index; SASS5 – South African Scoring System 5; EPT – Ephemeroptera, Plecoptera & Trichoptera index; SPEAR_{pesticides} – Species at Risk index for pesticides; TU – Toxic Unit

Declarations

Ethics approval and consent to participate

N/A

Consent for publication

N/A

Availability of data and material

All data generated or analysed during this study are included in this published article [and its supplementary information files].

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Authors' contributions

ML conceived the approach; AG, FK, UF, FM, BT, WB, ML, HH & JB designed the research; AG, FK and JB conducted the research; JB and AG analysed and interpreted the data; JB and AG drafted the initial version; AG, FK, UF, FM, BT, WB, ML & JB contributed to the final version - all approved the final version of the publication.

AG: Akbar A. Ganatra; FK: Faith Jebiwot Kandie; UF: Ulrike Fillinger; FM: Francis McOdimba; BT: Baldwyn Torto; WB: Werner Brack; ML: Matthias Liess; HH: Henner Hollert; JB: Jeremias Becker

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Competing interests

The author(s) declare no competing interests.

Authors' information (optional)

N/A

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Figures

Figure 1

Location of the sampling sites in the study area of Western Kenya, East Africa. Site locations are displayed with the site code and colour-filled to annotate the relationship between predicted pesticide pollution (TUMax) using SPEARpesticides and measured pollution in water samples using LC-HRMS in Kandie et al. (2020). Akbar Ganatra. "SPEARpesticides study sites". "World Topographical map" & "World Hillshade". February 2nd, 2021. <https://arcg.is/1fiDn9>.

Figure 2

The freshwater macroinvertebrate community composition changes in streams with high pesticide pollution. The stacked bars show the mean proportions of macroinvertebrate families on the overall number of individuals observed in lowly-polluted (maximum toxic unit $TU_{max} < 10^{-3}$) and in highly polluted streams ($TU_{max} \geq 10^{-3}$). The number of individuals ranged from 32 to 154 (mean: 94) in the 5 lowly polluted streams and from 37 to 462 (mean: 152) in the 8 highly polluted streams. Families that were recorded in at least 50 % of the streams are shown in bold. Taxa shown in blue have been classified as being at risk in the adapted SPEARpesticides indicator for East African streams, taxa shown in red have been classified not at risk.

Figure 3

SPEARpesticides indicates pesticide pollution in tropical streams of Western Kenya. Pesticide pollution was quantified as the maximum toxic unit (TU_{max} , restricted to $\geq 10^{-4}$) out of 30 pesticides measured in water samples collected during the rainy season but not during peak exposure. Freshwater macroinvertebrates were identified to the family level. (a) Application of the European SPEARpesticides index v. 2019.11 from Knillmann et al. (2018) to the Kenyan samples; $R^2 = 0.43$, $F = 8.15$, $df = 1$, $\text{res. df} = 11$, $p = 0.016$, intercept = 0.62, slope = -0.37. (b) Application of the SPEARpesticides index after adaption to East African streams. $R^2 = 0.53$, $F = 12.35$, $df = 1$, $\text{res. df} = 11$, $p = 0.005$, intercept = -0.02, slope = -0.43.

Figure 4

Performance of the East African SPEARpesticides index. (a) Background pesticide toxicity (maximum toxic unit, TU_{max}) predicted with the East African SPEARpesticides index correlates with the measured background toxicity in Kenyan water samples. $R^2 = 0.54$, $F = 12.97$, $df = 1$, $res. df = 11$, $p = 0.004$, intercept = -0.41, slope = 0.80. TU_{max} was restricted to $\geq 10^{-4}$, because lower toxicity could not be measured due to detection limits for pesticides. TU_{max} has been $\log_{10}$ -transformed, so that differences are presented in orders of magnitude. (b) Relation of the East African SPEARpesticides index with the estimated peak pesticide toxicity during run-off in Kenyan streams, as compared to the relation of the European SPEARpesticides index v. 219.11 with measured peak toxicity in German streams (data from Knillmann et al. 2018). Peak exposure in Kenyan streams was estimated by multiplying the measured background toxicity with a correction factor of 4 (see main text for justification).

Figure 5

Distribution of physicochemical parameter values at the study sites. The violin plot shows the kernel probability density of the data points across the parameter values. White points indicate the median, black boxes the interquartile range, and black lines 1.5 x the interquartile range. Light-grey boxes indicate the range of values considered typical or recommend for streams with good water quality.

Figure 6

Principal component analysis of the East African SPEARpesticides index and those environmental variables that (marginally) significantly correlated with any of the investigated bioindicators. The first principal component (horizontal axis) explains 60.7 % of the overall variation and was considered to represent a gradient of stream types. The second principal component (vertical axis) additionally explains 14.7 % of the overall variation and was considered to represent a gradient of high water level. Each point represents a stream (identified by its number, see Tab. S4 in the supplementary material), colors refer to the average SPEARpesticides value obtained from each stream.

Supplementary Files

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