Using aquatic insect communities (Arthropoda: Insecta) to assess water quality in Kob Srov Lake, Phnom Penh, Cambodia

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ទួលន័យសង្ខេប

សត្វល្អិតទក់ជាស្ទូចនាក់វេសោស្ត្រដ៏មានសារ:សខានក្នុងការបង្ហាញពគុណភាពទក។ យេងបានធ្វើការសង្កេតបម្រេបម្រួលរបាយនេ សត្វល្អិតទឹក និងទំនាក់ទំនងរបស់វាជាមួយនឹងលក្ខណៈរូប-គីមីនៃប៉ារ៉ាម៉ែត្រទឹកដើម្បីវាយតម្លៃគុណភាពទឹកនៅបឹងកប់ស្រវ រាជធានី ភ្នំពេញ។ សំណាញ់ដៃ និងអន្ទាក់សិប្បនិម្មិតត្រវបានប្រើដើម្បីប្រមូលសត្វល្អិតទឹកពីទីតាំងសិក្សាចំនួន ៧ កន្លែងនៅបឹងកប់ស្រវ។ អនុគមន៍លីនេអ៊ែរ (Multiple linear regression) ត្រវបានប្រើដើម្បីសិក្សាពីទំនាក់ទំនងរវាងចំនួនអំបូរ ចំនួនឯកត្ត: និងភាព សម្បូរបេបនេសត្វល្អត់ទក់ដោយប្រេតម្លេបារាមេត្រលក្ខណៈរូប-គមនេទក់។ តម្លេសន្ធស្សនជវសាស្រ្ត (Biological Monitoring Working Party index) និងតម្លេមធ្យមក្នុងអប្វរមួយត្រូវបានប្រេដេម្បីថាតិថ្នាក់គុណភាពទឹកនៅតាមទីតាំងសក្សា។ យេងបាន ប្រមូលសត្វល្អធមកសរុបចន្ទន ២,២៨៣ ក្បាល ស្ថតក្នុង ១៨ អប្វរ នង ៦ លដាប។ សត្វល្អតទកដេល មានចន្ទនច្រេនជាងគេនោ ទីតាំងសិក្សាទាំងអស់គឺ Chironomids (Diptera) (មានលើសពី ៧៧% នៃចំនួនសំណាកសត្វល្អិតទឹកសរុប)។ សត្វល្អិតទឹក ដែលមានចន្ទនតចិជាងគេគស្ថតក្នុងលដាប Lepidoptera។ ចំនួនអបូរ ចំនួនឯកត្ត: និងភាពសម្បូរបេបនេសត្វល្អត់ទឹកមានភាព ខុសគ្នានោទតាងទាង ៧ កន្លេង ហេយរាមានទនាកទនងទោនងបារាមេត្ររូប-គមជាកលាក។ ចន្ទនអបូរ ចន្ទនឯកត្ត: នងភាពសម្បូរ បេបនេសត្វល្អធុទកមានទនាក់ទនឯវជ្ជមានជាមួយអុកសុសេនរលាយក្នុងទក ខណ:ដែលចំនួនអបូរ និងចំនួនឯកត្ត:មានទនាក់ទនង អជ្ជេមានជាមួយសតុណ្ហភាពទក់មានកម្រត់ពមនល្អទោមធ្យមនោក្នុងទតាងផ្សេងៗគ្នានេបង។ លទ្ធផលនៃការសក្សា របសយេងបានបង្ហាញថា ការធ្វេខ្យប្រសេរឡេងនូវកម្រត់អុកសុសេនរលាយក្នុងទក នងសតុណ្ហភាពទក់នងធ្វេខ្យប្រសេរឡេងនូវ គុណភាពទក នងបង្កេនបូពុយឡាស្យុងសត្វល្អតទកនោក្នុងបងកបស្រូវ។

Abstract

Aquatic insects are effective bioindicators of water quality. We investigated the spatial distribution of aquatic insects and their relationships with physico-chemical water parameters to evaluate water quality in Kob Srov Lake, Phnom Penh. Hand nets and artificial substrate traps were employed to sample aquatic insects at seven sites across the lake.

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Multiple linear regressions were applied to explore associations between the taxonomic richness, abundance and diversity of aquatic insects with physico-chemical parameters, whereas Biological Monitoring Working Party index values and average scores per taxon were used to rank water quality at study sites. We collected 2,283 aquatic insects belonging to 18 families in six orders. Chironomids (Diptera) dominated all study sites (representing >77% of all individuals sampled), whereas lepidopterans were the least represented order. Patterns of taxonomic richness, abundance and diversity varied across the lake and were related to specific physico-chemical parameters. Taxonomic richness, abundance and diversity were positively associated with dissolved oxygen, whereas the first two metrics were negatively associated with water temperature. Water quality ranked from poor to moderate at different locations across the lake. Our results suggest management efforts focusing on enhancing dissolved oxygen levels and water temperature would improve water quality and support aquatic insect populations in Kob Srov Lake.

Keywords Aquatic insects, Biological Monitoring Working Party, Diptera, water quality.

Introduction

Aquatic ecosystems are threatened by various humaninduced factors including chemicals from agricultural, industrial and mining activities, as well as sewage water discharged from households into the water bodies (Dudgeon, 2000; Yule & Yong, 2004; Gopal, 2005). Agricultural practices, particularly the use of chemical fertilizers, pesticides and livestock manures are the main sources of water pollutants such as heavy metal ions (Rashid *et al*., 2023). These pollutants have resulted in water contamination and waterborne diseases including typhoid fever, cholera, and diarrhoea (Cairns & Pratt, 1993), consequently affecting the health of humans and other organisms. As a result, various methods including the use of aquatic insects have been developed to analyse water quality impairment and assess the health status of aquatic ecosystems (Rosenberg & Resh, 1993; Xu *et al*., 2014).

 Aquatic insects occur in both terrestrial and mainly aquatic habitats (mostly freshwater habitats) including lotic systems (e.g., springs, streams and rivers) and lentic systems (e.g., lakes, ponds, wetlands and bogs) (Starr & Wallace, 2021; Vilenica *et al*., 2022). Aquatic insects have one or more life cycle stages which mostly occur in water in their egg and larval forms and migrate to terrestrial habitats in their adult stages (Dijkstra *et al*., 2013; Starr & Wallace, 2021; Vilenica *et al*., 2022). They are one of the most abundant and speciose groups, comprising approximately 130,000 species across 12 orders, representing more than 60% of freshwater species (Mayhew, 2007; Dijkstra *et al*., 2013). Some species are very vulnerable and sensitive to pollution, such as Ephemeroptera (mayflies), Trichoptera (caddisflies), Plecoptera (stoneflies) and Odonata (dragonflies & damselflies) (Lindenmayer et *al.,* 2000; Bonada *et al.,* 2006; Shafie *et al.,* 2017), whereas others such as Diptera can survive in very polluted waters (Cummin & Merrit, 1996; Hepp *et al*., 2013; Arimoro *et al*., 2018). These groups are important components of both aquatic and terrestrial ecosystems, serving as primary consumers, detritivores, predators and pollinators (Dijkstra *et al*., 2013; Vilenica *et al*., 2022). They also play a vital role in nutrient cycling and contribute significantly to the aquatic-to-terrestrial transfers of food and energy (Dijkstra *et al*., 2013; Starr & Wallace, 2021).

 Due to their sensitivity to environmental conditions, aquatic insects are widely employed as bioindicators to assess ecosystem health (Arimoro & Ikomi, 2008; Chowdhury *et al*., 2023). They are useful for assessing or monitoring anthropogenic stress on ecosystems, including sewage discharge, agricultural runoff, recreational activities, land clearing and urban development over extended periods (Cairns & Pratt, 1993). It has been reported that aquatic insects offer more accurate insights into the dynamics of water bodies or river systems than chemical data (Shafie et al., 2017). As such, many indices have been developed to monitor water quality, including diversity indices (e.g., Simpson diversity, Shannon diversity) and scoring system (i.e., Biological Monitoring Water Party (BMWP) (Armitage *et al*., 1983) and Belgian Biotic Index (de Pauw & Vanhooren, 1983).

 Aquatic insects in freshwater bodies have been extensively researched, but remain relatively poorly studied in stable freshwater systems such as lakes. Few papers have investigated water quality in the Lower Mekong Basin in Cambodia to date. Those that have include studies on the spatial heterogeneity of macro-invertebrates (Sor *et al*., 2017) and aquatic insect communities (Sor *et al*., 2021). Other studies have examined aquatic insect diversity and its relationship to water quality in urban ponds in Phnom Penh (Chhy *et al*., 2019), alongside studies on aquatic Ephemeroptera (Chhorn *et al*., 2020) and Coleoptera (Doeurk *et al*., 2022) in relation to freshwater quality in southwest Cambodia. Additionally, research has been undertaken on aquatic Hemiptera (Zettel et al., 2017) and Polyphaga checklist (Freitag *et al*., 2018).

 Although water quality was recently examined in Kob Srov Lake (KSL) in the northern area of Phnom Penh in terms of heavy metal contaminations (Kev *et al*., 2019), this has yet to be assessed in relation to aquatic insect communities. The latter is necessary for enhancing understanding of the ecological context and implications of water pollution at the site. We consequently aimed to assess water quality in KSL using aquatic insect communities as bioindicators. To this end, we first examined the spatial distribution of aquatic insects in the lake, then quantified relationships between aquatic insect diversity and selected chemical parameters. Finally, we assessed water quality in the lake using the BMWP system. Our overall purpose was to provide baseline information to local inhabitants and authorities about water quality in KSL and the need for improvement of wastewater management strategies in surrounding areas.

Methods

Study area

This study was conducted in the KSL in the northern area of Phnom Penh City (11.638969°N, 104.818686°E; Figs. 1–2), Cambodia. Kob Srov Lake encompasses approximately 26 km² and is located in the Sen Sok and Posenchey Districts of the city. While the lake is artificial and was created to protect Phnom Penh from flooding during in the wet season, it has recently also received wastewater from the western side of the city and been subjected to agricultural and other human activities which have degraded water quality. The lake is an important reservoir, not just because it receives excess water from Phnom Penh but because it stores a large amount of agricultural wastewater. Further, the lake supplies water to people around the lake for crop production, aquaculture, livestock and fishing activities and also serves as a biofilter to clean water (Kev et al., 2019) before draining into the Tonle Sap River at Prek Pnov.

Sampling design & data collection

Aquatic insects and water quality data were collected from seven sites in the KSL from 10 to 16 March 2018 (Figs. 1–2). Three samples were collected from each site, resulting in 21 samples for the study. Collection of a single sample took about 20 minutes, including measurement of water parameters and collection of aquatic insects.

Fig. 1 Location of seven study sites at Kob Srov Lake, Phnom Penh, Cambodia.

 Aquatic insects were sampled using two methods, namely hand nets and artificial substrate traps. The hand nets had an opening area of 30 x 30 cm, a depth of 92 cm and a mesh size of 1 mm and was used to collect insects in water or reed vegetation along the shoreline. Artificial substrate traps were employed to sample insects in deeper waters, with a single trap left at each study site for approximately four weeks. Following collection, insect samples were placed on a white tray in the field and rinsed with water for sorting and screening. These were then transferred with forceps into labelled containers containing 75% or 80% ethanol. In the laboratory, insects were sorted and identified to the family level using taxonomic keys (Dudgeon, 1999; Yule & Yong, 2004). Larger aquatic insects were examined by naked eye, whereas smaller insects were examined using an Olympus SZ51 dissecting microscope.

 Five physico-chemical parameters were employed for our water quality assessment, namely dissolved oxygen (DO, as %), potential hydrogen (pH), turbidity (TBD, as FNU [Formazin Nephelometric Units]), water temperature (WT, in $^{\circ}$ C) and electrical conductivity (CON, in mS/m [molloSiemens per metre]). These were measured at each study site using a HI-7609829 multiparameter portable water quality meter (Hanna Instruments Ltd., Bedfordshire, UK) at water depths ranging from $0.1 - 0.5$ m.

Data analyses

We used three metrics to quantify the aquatic fauna of our study sites: taxonomic richness, abundance and

Fig. 2 Indicative images of study sites at Kob Srov Lake, Phnom Penh, Cambodia.

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Shannon-Wiener's diversity index (H). The latter H values were computed using the 'vegan' package (Oksanen *et al*., 2022) in R (R CoreTeam, 2022) and are based on taxonomic richness and abundance (Kerkhoff, 2010; Hamid & Rawi, 2017). Bubble plots were used to depict spatial distribution in these metrics across the seven sampling sites in the lake.

 We employed multiple linear regression to investigate the relationships between the above metrics and environmental parameters. In our model, insect richness, abundance and Shannon-Wiener's diversity index were dependent variables, whereas physico-chemical factors were employed as explanatory variables e.g., DO, pH, TBD, WT and CON. Prior to model construction, dependent variables were log-transformed to remove the effects of outliers and ensure data normality, whereas independent variables were normalized (by making the margin sum of squares equal to one; ranging from 0 to 1) to ensure the same scale across variables. We assumed that the logarithmic transformation enables normal data distribution. Afterwards, a stepwise variable selection approach based on Akaike Information Criterion (AIC) was employed to remove unimportant variables using the 'stepAIC()' function of the 'MASS package (Venables $&$ Ripley, 2002) in R (R CoreTeam, 2022). The influence of each environmental parameter on the distribution of the taxonomic richness, abundance and H was assessed using standardized regression coefficients. Model performance was assessed using coefficients of determination (R-square). The models were created using 'lm()' function of the 'stats' package in R (R CoreTeam, 2022).

 The BMWP system was used to score each aquatic insect family present in our seven study sites in KSL. Scores for individual families reflect their pollution tolerance based on knowledge of distribution and abundance. Pollution-intolerant families have high BMWP scores, whereas pollution-tolerant families have low scores (Armitage *et al*., 1983; Sivaramakrishnan, 1992). More specifically, scores ranging from 0–40, 41–70 and 71–100+ indicate very poor to poor (heavily polluted to polluted), moderate (moderately impacted) and good to very good water quality (clean but slightly impacted to unpolluted), respectively (Chesters, 1980; Mustow, 2002). The Average Score Per Taxon (ASPT) represents the average tolerance of all taxa within the community and is calculated by dividing the BMWP score by the number of families in a sample (Walley & Hawkes, 1996). More specifically, ASPT values ranging from 1–2.49 (water quality class III–IV), 2.5–3.99 (III), 4.0–5.49 (II–III), 5.5–6.99 (II), 7.00– 7.99 (I–II) and 8.0–10.0 (I) indicate 'poor', 'fairly poor', 'moderate', 'fairly good', 'good' and 'very good' water quality, respectively (Mustow, 2002). Thus, a high ASPT value denotes clean sites with a relatively large number of high-scoring taxa (Sivaramakrishnan, 1992).

Results

Community composition of aquatic insects

We recorded a total of 2,283 aquatic insects belonging to 18 families and six orders during sampling (Annex 1). These were dominated by Diptera (80.11%), followed by Hemiptera (12.44%), Odonata (5.52%), Coleoptera (0.96%), Ephemeroptera (0.79%) and Lepidoptera (0.18%). At a family level, members of Chironomidae were most dominant taxa (77.31%), followed by members of the Micronectidae (8.60%), Libellulidae (3.64%), Ceratopogonidae (2.67%), Belostomatidae (1.88%), Notonectidae

Table 1 Physico-chemical parameters recorded at sampling sites in Kob Srov Lake, Phnom Penh. Values are given as mean, min–max (based on three samples collected at each site).

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(1.58%), Coenagrinonidae (1.49%), Hydrophilidae (0.96%), Caenidae (0.44%), Protoneuridae (0.35%), Baetidae (0.35%), Gerridae (0.31%), Crambidae (0.18%), Syrphidae (0.09%), Stratiomyidae (0.04%), Gomphidae (0.04%), Veliidae (0.04%) and Nepidae (0.04%). Physico-chemical variables recorded at sampling sites are summarized in Table 1.

Spatial distribution of aquatic insects

Significant variation was observed in the taxonomic (family) richness, abundance and diversity of aquatic insects between sites (Fig. 3). More specifically, taxonomic richness was higher in the southeastern (site 1), south-central (site 2) and southwestern (site 5) portions of the lake, and lower in the northwestern (site 7) and north-central (site 6) areas of the lake. No aquatic insects were observed in the northeastern (site 3) and central (site 4) portions of the lake. Further, the southeastern (site 1) and south-central (site 2) areas of the lake supported signficantly higher insect abundance. Finally, the southwestern (site 5), northwestern (site 7) and northcentral (site 6) areas of the lake hosted the highest levels of diversity, followed by the southeastern (site 1) and south-central (site 2) areas.

Aquatic insect & water quality associations

Our multiple linear regression models yielded adjusted R2 values of 0.310, 0.480 and 0.170 for taxonomic richness, abundance and Shannon-Wiener's diversity, respectively (Table 2). Dissolved oxygen was positively related to taxonomic richness, abundance and Shannon-Wiener's diversity, whereas water temperature was negatively correlated with taxonomic richness and abundance.

Table 2 Standardized regression coefficients for taxonomic richness (SR), abundance (AB) and Shannon-Weiner's index values (H) for aquatic insects modelled against water parameters in Kob Srov Lake, Phnom Penh. Model performance for each is indicated by adjusted \mathbb{R}^2 values. Asterisks indicate significance at $p<0.05$.

Fig 3 Bubble plots indicating taxonomic richness, abundance and Shannon-Wiener diversity index values recorded at sampling sites in Kob Srov Lake, Phnom Penh.

Water quality in Kob Srov Lake

On the basis of BMWP and ASPT scores, water quality ranked as moderate (albeit decreasingly so) in the southeastern (site 1), south-central (site 2), southwestern (site 5) and north-central (site 6) portions of the lake, whereas it ranked as fairly poor in the northwestern portion (site 7) (Table 3). As before, the northeastern (site 3) and central

Table 3 Water quality scores and associated rankings for sampling sites in Kob Srov Lake, Phnom Penh.

(site 4) portions of the lake did not achieve biotic scores for water quality as no aquatic insects were recorded in these areas.

Discussion

Our study contributes to a better understanding of aquatic insects and water quality of Kob Srov Lake. We recorded 18 families belonging to six orders of aquatic insects and found that patterns of aquatic insect taxonomic richness, abundance and diversity varied across the lake. These three metrics were positively correlated with dissolved oxygen, whereas taxonomic richness and abundance were negatively correlated with water temperature, and water quality ranked as moderate to poor in different areas of the lake.

 Similar to studies of aquatic insects inhabiting freshwater ecosystems elsewhere (Balian et al., 2008; Sarikar & Vijaykumar, 2022), members of Diptera and Hemiptera were the most abundant taxa in our study. This is unsurprising because Diptera is typically the largest taxonomic group in representing half of all aquatic insects (Dijkstra *et al*., 2013). Members of Coleoptera and Lepidoptera were the least abundant taxa in our study, which is consistent with reports identifying these as the smallest taxonomic groups of aquatic insects (Dijkstra *et al*., 2013).

More specifically, Chironomidae were the most dominant family in our study, accounting for over 77% of insects. These were ubiquitous across our sampling sites and evidently tolerant of differing environmental conditions, as suggested by Popoola & Otalekor (2011). Standing and low current waters and mud or sandy areas with high fine particle sizes are known to support higher diversity and abundance of chironomids (Doisy & Rabeni, 2001; Kubendran & Ramesh, 2016) and they are commonly found in high turbidity and polluted waters

(Armitage *et al*., 1995; Johnson, 1995). This is consistent with the dominance of the group at sampling site 2, which was characterised by such conditions.

Heterogeneous patterns of taxonomic richness, abundance and diversity were observed across the lake. Taxonomic richness was high along the southern periphery of the lake where various aquatic vegetation (water grass, small trees and water hyacinth) was observed. This likely provides favourable conditions for aquatic insects to shelter and forage, as well as refuges from predators (Andersson, 2014), allowing them to complete life cycles and providing feeding and nursing grounds. In contrast, the northern area of the lake hosted lower aquatic insect richness and abundance, possibly due to less aquatic vegetation and microhabitat, whereas no insects were found in the northeastern and central portions of the lake. This may be attributable to significant water disturbance and pollution as well an absence of vegetation in these areas.

 The physico-chemical parameters we measured offer good indications for determining aquatic insect communities. We found dissolved oxygen was positively related to taxonomic richness, abundance and diversity. This is consistent with previous studies that found that aquatic insect abundance was positively correlated with dissolved oxygen (Yahaya & Suleiman, 2017; Chhy *et al*., 2019). We also found that water temperature was negatively related to taxonomic richness and abundance. Water temperature appears to be an important factor impacting the ecosystem changes (Arim *et al*., 2007) and rising temperatures contribute to declines in the density and richness of aquatic insects (Vannote & Sweeney, 1980; Glazier, 2012). Dissolved oxygen levels also decrease when water temperature increase, likely due respiration and other processes such as organic matter degradation (Ebenebe *et al*., 2016). Higher temperature may cause

the water column to become more biologically active, which could lead to species in more tolerant groups (such as Diptera, Hemiptera and Odonata) thriving over less tolerant groups (e.g., Trichoptera and Ephemeroptera) (Ngodhe *et al*., 2014). Our data suggests that sensitive taxa require higher levels of dissolved oxygen for respiration, whereas tolerant organisms are capable of aerobic biodegradation of organic matter in lower oxygen aquatic environments. Further, most dipterans, hemipterans and odonates were found in water temperatures between 27° C and 34° C. This may attributable that these being able to shift their metabolic processes from aerobic to predominantly anaerobic at high temperatures (Hamburger *et al*., 1994).

 Our data suggests that water quality in Kob Srov Lake ranges from moderate in the southeastern, south-central, southwestern and north-central lake portions to fairly poor in the northwestern lake portions. The latter is likely due in part due to the fact that this area was dominated by tolerant taxa. Water quality ranked least in the northeastern and central portions due to the complete absence of aquatic insects. This may be attributable to disturbance from transport or sewage discharge from Phnom Penh, as these can lead to water degradation (Angelidis *et al*., 1995; Yang *et al*., 2004; Okumagba & Ozabor, 2014).

It is important to note that our results do not reflect the current status of Kob Srov Lake, but rather the conditions six years ago. Some parts of the lake have been experienced increased urbanization since this time. Irrespectively, our study highlights the important roles of aquatic insect diversity in association with dissolved oxygen and water temperate in indicating water quality in the lake. They also suggest measures are needed to improve poor water quality in certain areas of the lake for human and agriculture consumption. Management of wastewater will undoubtedly be important in this regard. Further assessments of water quality during both the dry and wet seasons with more sample sites would be of interest to evaluate seasonal variations in aquatic insects and water quality. Additionally, identification of aquatic insects to species level is advised to elucidate community structure and generate a checklist of aquatic insects inhabiting the lake and surrounding areas.

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Annex 1 Abundance of aquatic insect families recorded in Kob Srov Lake, Phnom Penh

Annex 1 Cont'd

