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# Longitudinal malacological monitoring, with a parasitological survey, reveals new schistosomiasis transmission foci in deforested sites in Western Uganda

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## Abstract

**Background** In sub-Saharan Africa, the epidemiology of schistosomiasis, a waterborne parasitic disease, is often altered by environmental and land-use changes. In the upland area of Western Uganda, ongoing destruction of the Bugoma Forest is transforming many lotic freshwater habitats, potentially favouring intermediate hosts of schistosomes. We therefore undertook a longitudinal malacological monitoring study for *Biomphalaria* and *Bulinus*, keystone intermediate host snails, to describe actual or potential transmission of schistosomiasis.

**Methods** Starting in June 2023, snail sampling was conducted regularly, every other month, at nine sites for eighteen months, until October 2024. To bolster this, an analysis of satellite imagery of vegetation and land-use change over the previous 20 years was undertaken. At each inspection site water temperature, conductivity and pH were noted, alongside on-site observations of human and animal activities. Snails were identified by morphology, with inspection for shedding *Schistosoma* cercariae, which were later identified by molecular analysis.

**Results** Mean water temperature, conductivity and pH were 23.5 °C ( $\pm 1.8$ ), 235.0  $\mu\text{S}$  ( $\pm 81.7$ ) and 6.9 ( $\pm 0.3$ ), respectively. A total of 2524 *Biomphalaria* snails at seven sites and 422 *Bulinus* snails at one site were examined. Abundance of *Biomphalaria* snails was greater ( $p < 0.01$ ) in sites where the forest had been cleared, with human encroachment evident. *Schistosoma* cercariae were shed from 0.4% of *Biomphalaria* and 1.9% of *Bulinus* with monthly maxima of 3.1% and 3.7%, in December 2023 respectively. In August 2024, among local school-aged children ( $n = 41$ ), the prevalence of intestinal schistosomiasis was 56.1% whereas urogenital schistosomiasis was absent. Further to the presence of *Schistosoma mansoni* cercariae, molecular analyses also revealed *Schistosoma bovis* and *Schistosoma rodhaini*. Satellite imagery showed land-use change with the Bugoma central Forest reduced by 18.0% and shrub land by 57.0% due to human agricultural activities, revealing an expanding zone of putative schistosomiasis transmission in upland areas.

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**Conclusion** This changing endemicity of human and animal schistosomiasis to new areas, some 15 Km away from Lake Albert, needs to be addressed by the Uganda Ministry of Health and National Forestry Authority. Further targeted malacological and schistosomiasis surveillance, at finer geographical resolutions, will pinpoint new foci and better guide local interventions.

**Keywords** Molecular epidemiology, Bugoma forest, Deforestation, *Biomphalaria*, *Bulinus*, Invasive snails, Population displacement, Disease control

## Background

Schistosomiasis, commonly known as Bilharzia currently afflicts many millions of people in sub-Saharan Africa (SSA) [1, 2]. It is a water-borne Neglected Tropical Disease (NTD) attributable to infection with trematodes of genus *Schistosoma*, transmitted by certain freshwater intermediate host snail species [1]. *Schistosoma mansoni* and *Schistosoma haematobium* cause intestinal and urogenital schistosomiasis, respectively. Schistosomiasis can cause overt chronic morbidities sometimes leading to permanent disability and death if not treated promptly with praziquantel, the only available anthelmintic [2]. In addition to human schistosomiasis there are other species of schistosomes e.g. *Schistosoma bovis* in livestock and *Schistosoma rodhaini* in wildlife [1, 2]. At a species-level, schistosome cercariae are morphologically indistinguishable, for example, *S. mansoni* and *S. rodhaini* can be transmitted by *Biomphalaria* whereas *S. haematobium* and *S. bovis* can share certain *Bulinus* [1, 3, 4] and require identification using molecular methods [5–7].

Schistosomiasis is most endemic throughout many parts of SSA due to poor hygiene, inadequate access of poor communities to safe water and sanitation amenities and a suitable climate for parasite development [2, 3]. Indeed, ongoing land-use changes often favour the creation of new aquatic habitats suitable for later colonisation by intermediate host snails [1, 3]. Here, we place special attention on the upland forested area of the Lake Albert region of Uganda, originally covered by Bugoma and Budongo Forests. This forested canopy once provided substantial shading of cooler aquatic habitats beneath that were unsuitable for intermediate host snails. By contrast, the warmer and unshaded lentic habitats along open shorelines of Lake Albert nearby provided extensive intermediate host snail habitats with hyperendemic intestinal schistosomiasis in lakeshore communities [8]. Over the last few decades, like other forests in Uganda, Bugoma Forest is threatened by an increasing human population. There has been continuous destruction due to agricultural expansion and the settlement of displaced populations [9–12]. It is predicted that by 2040, 50% of the area of Bugoma Forest will be destroyed and occupied by human settlements [10, 13]. Such vegetation loss opens the ground to direct sun heat, causing increased temperatures and general environmental

changes which can favour schistosomiasis transmission [14].

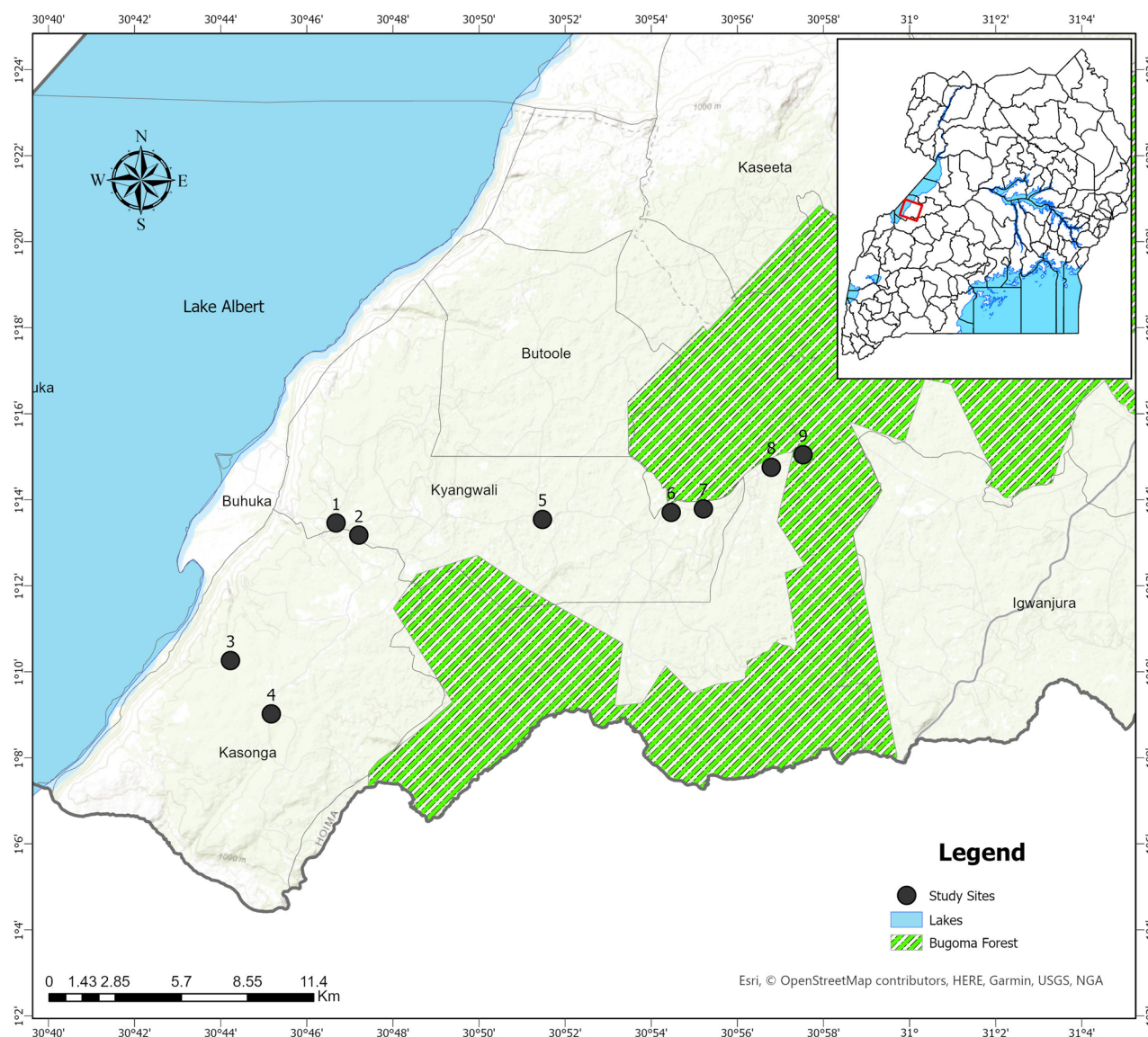
Kyangwali refugee settlement, located west of Bugoma Forest, has since 2012 had an increasing influx of displaced people from the Democratic Republic of Congo (DRC) and South Sudan, resulting in accelerated encroachment on the forest reserve [13, 15, 16]. With such increasing deforestation and population movement into the Albertine region [11, 12, 17], we conducted a detailed longitudinal malacological monitoring study, followed by a preliminary cross-sectional parasitological screen of local resident school-aged children, to investigate the potential for schistosomiasis transmission and spread to upland areas, likely driven by local environmental change.

We report on current findings in Kikuube District drawing attention specifically on land-use change, intermediate host snail dynamics augmented with molecular identification of encountered *Schistosoma* cercariae in snails.

## Methods and materials

### Study area

Our study was conducted in the Lake Albert zone of Kikuube District in Western Uganda from June 2023 to October 2024. The area has two distinct ecological zones: fishing communities along the shoreline of Lake Albert, in the floor of the rift valley some 1,000 m lower in altitude, and the upland side of the escarpment, which was once a large conservation area covered by Bugoma Forest (Fig. 1). In 1967, a small portion of the forest was cleared to establish the Kyangwali refugee settlement to accommodate displaced populations from Rwanda and Congo. Since 2012, the camp has continuously expanded with a regular influx of refugees, characterising the encroached area into the early, (gradually cleared 1967–2005), and the later forest-cleared zones where accelerated forest destruction started in 2010 [18]. Many internal migrant populations from other locations of Uganda have also continued to encroach onto the forest for subsistence agriculture. Environmental studies in the Albertine area have predicted that 50% of forest cover could be lost by 2040 [10, 13]. To control schistosomiasis locally, mass treatment of communities on the shoreline of Lake Albert



**Fig. 1** Map of the study area showing snail sampling sites in Bugoma, Kikuube District

started in 2003, while those communities in the forested upper area were typically regarded as non-endemic. As this upland area now has several swamps, marshes and exposed streams, originally under the forest canopy, it needs to be periodically monitored and examined. All the eight swamps in the encroached area known to hold water throughout the year were selected for malacological monitoring. The sites also had visible human settlement around them. Sites [1–4], in the earlier encroached area had dense settlements and subsistence crop gardens without trees while those [5–8] in the later encroached had sparse human settlement, crop gardens and scattered trees. An additional site 9 at the edge of the forest with no human settlement was included among the later encroached to represent the forest environment (Fig. 1).

### Study design

We conducted the study in a cascaded investigative approach, using iterative analysis of results from one step to later inform another. The first stage was longitudinal monitoring of snail populations, taking physico-chemical water parameters and noting natural *Schistosoma* infections. Snail sampling was conducted at each of the selected nine sites once in two months for 18 months. This step aimed to determine the distribution of snails in the sites under encroachment and to understand human settlement patterns around the points where snails were observed to shed *Schistosoma* cercariae. The second stage was molecular characterization of the *Schistosoma* cercariae collected from intermediate host snails to determine their species and risk to human or animal health. The third stage was a preliminary parasitological screen

of available local resident school-aged children (SAC) in households within 200 m radius from the sites with infected snails to assess their *Schistosoma* infection status. The fourth and last stage was a general retrospective description of observed environmental change in the last twenty years using satellite imagery.

#### Environmental parameters at snail collection sites

We observed and noted environmental parameters such as vegetation, status of deforestation, human water contact activities, and presence of animals. Physicochemical water parameters (temperature, conductivity and pH) were recorded using a handheld multi-parameter meter (HI98130, Hanna instruments, Leighton Buzzard, UK). Prevailing weather conditions and water level changes at the time of snail collection were noted.

#### Snail collection and identification

The geographical location coordinates of each site were taken using a Garmin GPS receiver (e-trex, Garmin, Olathe, USA). Snails were collected using standard two-meter handle scoops with a 2.0 mm mesh. The sampling was semi-quantified, with two collectors scooping at a site for 30 min, picking all *Biomphalaria* and *Bulinus* species snails found into a plastic cup [19]. The sampling frequency and time of scooping by two collectors was the same at each site. Each round of sampling was conducted in all sites in the same week. During the 30 min, each collector scooped constantly and picked snails without a break. The snails were identified to genus using a morphological key [20]. The *Biomphalaria* and *Bulinus* species snails from each site were placed in labelled plastic cups for transportation to the field lab for cercarial shedding experiments.

#### Cercarial shedding

Screening *Biomphalaria* and *Bulinus* species snails for natural schistosome infection was conducted in the field. Individual snails were first washed and placed in a cell culture plate shedding tray in 2.0 ml of water and exposed to indirect sunlight at intervals of 2–4 h [4, 21, 22]. Using a dissecting microscope, the *Schistosoma* cercariae from each shedding snail were first identified by morphology and thirty picked singly by micro-pipette onto an FTA card using methods in [22], and a selection also preserved in 70% ethanol within 1.5 ml tubes. Every snail shedding *Schistosoma* cercariae was preserved in absolute ethanol for molecular analysis.

#### Molecular identification of *Schistosoma* cercariae

The cercariae were identified by molecular methods using the samples on FTA card, collections in ethanol, or the preserved shedding snail tissue to ascertain whether they were of human or animal origin.

Identification of *S. rodhaini* was conducted at the Natural History Museum, UK, where DNA was extracted from each single cercaria ( $n = 10$ ) using the Qiagen DNeasy Blood and Tissue Kit (Qiagen, Manchester, UK) following the manufacturer's protocol. Individual cercariae were molecularly characterised using three molecular markers: a partial mitochondrial *cox1* region (~ 900 bp), the nuclear ITS rDNA (ITS1-5.8 S-ITS2 ~ 915 bp) region, and a partial region of the 18 S rDNA (289 bp) region, to determine the *Schistosoma* species. The PCR primers and parameters used for each marker are detailed [23]. Polymerase chain reactions (PCR's) were performed in 25  $\mu$ l reactions using Illustra PuReTaq™ Ready-To-Go™ PCR beads (GE Healthcare). Amplicons were visualised on a 0.8% gel-red agarose to identify PCR-positive reactions, which were then purified and Sanger sequenced. The identification nucleotide sequence was uploaded to the National Center for Biotechnology Information (NCBI-NIH) GenBank repository.

*Schistosoma bovis* Identification was conducted at the Liverpool School of Tropical Medicine, UK. Genomic DNA was extracted from the foot muscles of one sample of *Bulinus* snail which shed cercariae in the field, using the hexadecyltrimethylammonium bromide (CTAB) method [24] followed by phenol/chloroform DNA extraction and ethanol precipitation. Molecular identification of cercariae was performed using a high-resolution melt (HRM) species specific real-time PCR assay, based on previously described methods [7, 25].

Molecular identification of *S. mansoni* was conducted at the Africa Museum Laboratory, Belgium. Extraction of genomic DNA (gDNA) from *Biomphalaria* snails shedding *Schistosoma* cercaria was carried out using the E.Z.N.A.® Mollusc DNA Kit (OMEGA bio-tek, Inc.), a modified CTAB based method [26]. The multiplex RD-PCR to identify the parasite shed by the snail was done using a protocol described by Schols et al. [6]. The *Schistosoma*-specific Cytochrome c oxidase subunit I (COI) marker primer (5'–3') F-CAGGGGTTTCAAGTCTAAT TGGAT, R-CAAATAATAACATCGTTATTCTCTCTGG was used in the sequencing PCR [6, 27]. The *S. mansoni* identification sequence was uploaded to the NCBI-NIH repository for reference.

#### Screening school-aged children (SAC) near snail sites

At two sites where snails were shedding *Schistosoma* cercariae was conducted by the study team and Kikuube District health department as a public health response to the risk flagged by malacology. Community leaders and village health workers mobilised and referred school-aged children from households within a 200-meter radius around the infected snail sites to the Kyangwali Local government Health facility for schistosomiasis screening. The lot quality assurance sampling (LQAS) for

determining local schistosomiasis prevalence and need for mass treatment was used to screen at least 15 school-aged children [28, 29]. At the site where *B. pfeifferi* snails shed *Schistosoma* cercariae, we suspected *S. mansoni* transmission. Forty-one school-aged children provided faecal samples for screening using the Kato-Katz method while for the site where *Bulinus forskalii* shed the cercariae, urine samples were screened for *S. haematobium* using the syringe filtration technique of 10 ml urine collected during mid-morning [2].

#### Land use-land cover change (LULC) analysis (2000–2021)

At each snail monitoring site, we obtained information from local leaders about the history of forest destruction and human settlement. We took photographs of the surroundings to capture anthropogenic activities. We visited the Bugoma Conservation Association to obtain more historical records of Bugoma deforestation. Using remote sensing and satellite imagery, we analysed environmental change in the previous twenty years (2000 to 2021) to describe the extent of vegetation loss and anthropogenic activities using the 2000 land use/land cover (LULC) data obtained from the Global Land Cover 2000 (GLC2000) project. This dataset was derived from SPOT-VEGETATION imagery and classified according to the FAO Land Cover Classification System [30]. For the 2021 LULC, the ESA WorldCover 10 m v200 dataset (Tile N00E030) was utilized, which offers higher spatial resolution (10 m) and was generated from Sentinel-1 and Sentinel-2 satellite imagery [31]. Prior to analysis, both datasets underwent preprocessing in QGIS (v3.22.1; QGIS, 2021) to ensure spatial consistency. First, the imageries were reprojected to UTM Zone 36 N (EPSG:32636). Second, the ESA WorldCover data were resampled to a 1 km resolution using a majority-rule resampling method in QGIS to match the coarser scale of the GLC2000 product. This was done to address the resolution disparity between the GLC2000 (1 km) and ESA WorldCover (10 m) datasets.

#### Data management and analysis

The data sets were entered in excel. Statistical analysis was done using Stata (StataCorp, 2015. Stata Statistical Software: 14. College Station, TX: StataCorp LP). Simple descriptive statistics were used to compute infection rates. Comparison of *Biomphalaria* abundance and infection rates across sites and season was done using Kruskal Wallis test. Mean water parameters between the earlier and later encroached sites were compared using Welch T-test. The LULC was computed using R statistical software (v4.3.1; R Core Team 2023), leveraging specialized packages for geospatial and tabular data manipulation. The *terra* package (v1.8-44; Hijmans, 2025) was employed for raster-based calculations, including pixel-wise area computations. Change detection metrics

were computed to evaluate transitions between 2000 and 2021. Absolute changes in class area were derived by subtracting values of 2000 from those of 2021, while relative changes were expressed as percentages relative to the baseline. A Sankey diagram, generated using the *networkD3* package (V0.4.1; [32]), provided a visual representation of inter-class transitions, highlighting dominant land changes.

## Results

### Spatial and temporal snail distribution and infection

Of the nine sites, *Biomphalaria* snails were found at seven, while *Bulinus* at only one site. No snail was found at site 9 which was still under the forest cover. Out of a total 2524 *Biomphalaria* snails collected, 1872 (74%) were found in sites 1, 2 and 3 where the forest was cleared earlier and 651 (26%) in the sites 5,6,7 and 8 recently encroached. We found 422 *Bulinus forskalii* snails at a single site 4 only. No other *Bulinus* species was found. Abundance of snails was significantly greater ( $p = 0.002$ ) in sites ([1–3], and [4]) where the forest had been cleared at least 10 years ago by human encroachment compared to the areas cleared more recently.

*Biomphalaria* species snails shedding *Schistosoma* cercariae were only found in the zone of the forest which was encroached earlier, at site 2 with 10 out of 809 (1.2%) and site 3 with 1 out of 472 (0.2%). *Bulinus* snails were only found at site 4, with 8 out of 422 (1.9%) shedding *Schistosoma* cercariae. None of the 651 *Biomphalaria* snails from recently encroached sites 5,6,7 and 8 shed cercariae. Overall, *Schistosoma* cercariae were shed from 0.4% of the *Biomphalaria* species and 1.9% of *Bulinus* with monthly maxima in December 2023 of 3.1% and 3.7%, respectively (Table 1).

The mean water temperature, conductivity and pH of the snail monitoring sites were 23.5 °C ( $\pm 1.8$ ), 235.0  $\mu$ S ( $\pm 81.7$ ) and 6.9 ( $\pm 0.3$ ) respectively, For the four sites ([1–3] and [4]) with earlier encroachment, the mean water temperature, conductivity and pH sites were 24.4 °C, 276.2  $\mu$ S and 6.9 respectively. On the other hand, the five recently encroached sites ([5–8] and [9]) had the mean water temperature, conductivity and pH as 22.8 °C, 202.0  $\mu$ S and 6.9, respectively (Appendix 1). The Welch 2-sample t-test showed significantly higher average water temperature in the earlier encroached sites compared to the recently cleared ( $p = 0.0001$ , 95% CI = 0.811–2.34).

### Epidemiological response

Among the 41 local resident school-aged children screened for intestinal schistosomiasis near site 2, where *Biomphalaria* snails shed *Schistosoma* cercariae, 19 were female and 22 male. The average age was 9.5, the youngest was 6, and the oldest was 14 years. The prevalence of *S. mansoni* was 56.1%. Out of the 23 infected children 13

[illegible]

**Table 1** (continued)

| Site (name)   | GPS (N°, E°) | Snail               | 2024 |     |     |     |     |     |          |     |          |     |          |       | Total infected |
|---------------|--------------|---------------------|------|-----|-----|-----|-----|-----|----------|-----|----------|-----|----------|-------|----------------|
|               |              |                     | June | Aug | Oct | Dec | Feb | Apr | Infected | Aug | Infected | Oct | Infected | Total |                |
| 9 (Nyakatete) | 1.250810 °   | <i>Biomphalaria</i> | 0    | 0   | 0   | 0   | 0   | 0   | 0        | 0   | 0        | 0   | 0        | 0     | 0              |
|               |              |                     | 0    | 0   | 0   | 0   | 0   | 0   | 0        | 0   | 0        | 0   | 0        | 0     | 0              |
| 30.95880 °    | 30.95880 °   | <i>Bulinus</i>      | 0    | 0   | 0   | 0   | 0   | 0   | 0        | 0   | 0        | 0   | 0        | 0     | 0              |
|               |              |                     | 0    | 0   | 0   | 0   | 0   | 0   | 0        | 0   | 0        | 0   | 0        | 0     | 0              |

(56.5%) were female and 10 (43.4%) male. The *S. mansoni* infection intensity ranged from 12.0 to 900 eggs per gram (epg) with overall mean eggs per gram of 109.

At site 4, out of the 41 children whose urine samples were screened, 22 were female and 19 were male. The youngest was 4 years and the oldest 14. The mean age of the village children screened was 8.1 years. No child was found to be schistosome egg-positive in urine.

#### Molecular identification of cercariae

The DNA sequencing of the cercariae shed by two snails from site 2, morphologically identified as *Biomphalaria pfeifferi* identified *S. rodhaini* (NCBI GenBank nucleotide accession number PZ697201) that infects rodents.

Analysis of tissue from another two infected snails from the same site identified *Biomphalaria sudanica* shedding *S. mansoni* (NCBI GenBank nucleotide accession number PZ697200) a human trematode parasite that causes intestinal schistosomiasis. No *Schistosoma* coinfections were found in any of the *Biomphalaria* species snails. Molecular melting curve analysis of the tissue of one cercariae shedding snail morphologically identified as *B. forskalii* collected from site 4, identified *S. bovis*, a schistosome of livestock, typically cattle.

#### Land use and land cover change in Bugoma forest area

The land use and land cover change (LULC) mapping yielded three primary outputs whose results indicate a pronounced forest and shrub land loss coupled with cropland and grassland expansion from 2000 to 2021 as shown in Table 2; Fig. 2.

#### Drivers of environmental changes

The major anthropogenic activities we observed were expansion of subsistence and commercial agriculture, the establishment of settlements by internally migrating populations, sugarcane growing, cattle grazing in the forest and settlement of refugees from Democratic Republic of Congo and South Sudan. Cutting poles for building huts and burning charcoal were also common. There was flooding and expansion of swamps and marshes near sites 5, 6, 7 and 8. The site 9, closest to the forest still had tree cover and had no snails (Fig. 3).

#### Discussion

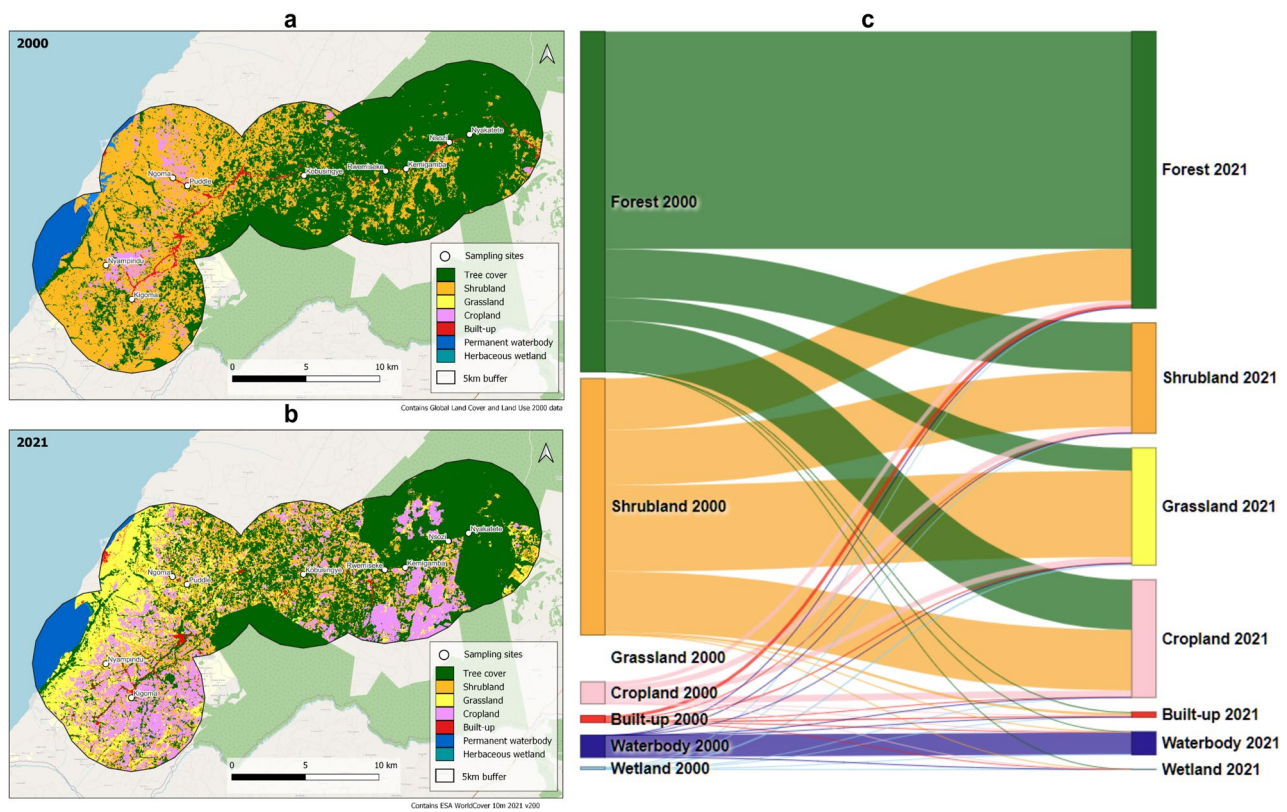
This study aimed to investigate the potential for schistosomiasis spread to the cooler upland areas of the Albertine in Western Uganda following environmental changes due to human encroachment on Bugoma Forest. Despite being a protected area, our analysis of satellite imagery shows that there was rather rapid encroachment on Bugoma Forest from 2000 to 2021 with the transition of extensive areas to grassland and cropland. We consider that this deforestation has resulted in environmental

**Table 2** LULC change in Bugoma Forest 2000 to 2021

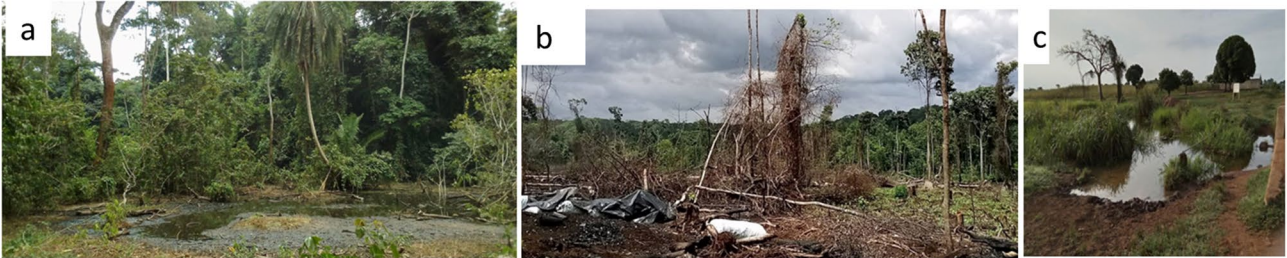
| Class     | Area                    |                         | Change Metrics       |           |
|-----------|-------------------------|-------------------------|----------------------|-----------|
|           | 2000 (km <sup>2</sup> ) | 2021 (km <sup>2</sup> ) | Δ (km <sup>2</sup> ) | %         |
| Forest    | 191.18                  | 155.47                  | -35.71               | -18.70%   |
| Shrubland | 144.19                  | 62.07                   | -82.12               | -57%      |
| Grassland | 0                       | 65.93                   | 65.93                | New class |
| Cropland  | 12.41                   | 66.15                   | 53.74                | 432.90%   |
| Waterbody | 12.47                   | 13.04                   | 0.57                 | 4.60%     |
| Wetland   | 1.45                    | 0.16                    | -1.29                | -89.10%   |

changes that favoured the local colonisation of *Biomphalaria* and *Bulinus* snails and with it the likely expansion of schistosomiasis transmission to upland areas away from the shoreline of Lake Albert.

We have demonstrated high local prevalence of *S. mansoni* among school-aged children near sites where *Biomphalaria* snails were found shedding *Schistosoma* cercariae. Settlement of displaced populations accelerated the forest encroachment and epidemiological risk of schistosomiasis spread to the new foci. The study further



**Fig. 2** An illustration of land-use and land cover change in Bugoma Forest in 20 years (2000–2021). **(a–b)**. About 18.7% of the forest trees were cut down to create land for farming. The shrubland surrounding the central forest was lost by 57%. **(C)**. Sankey diagram showing LULC transition. Open grassland and cropland were created following human encroachment on the forest



**Fig. 3** Photographic illustration of Bugoma Forest destruction: **a**. wetland under a forest at site 9, **b**. anthropogenic activities cause deforestation at site 6 **c**. Environmental change after forest loss favours *Biomphalaria* snail breeding and schistosomiasis transmission at site 3 within Kyangwali refugee settlement

shows that snail surveillance is an essential approach in monitoring the spread of new foci of schistosomiasis transmission. With the continuous deforestation and population movement, snail monitoring might be one of the cost-effective approaches schistosomiasis programs should strengthen for real time surveillance of schistosomiasis transmission. Although urogenital schistosomiasis is uncommon in Uganda [33], our findings show that there is a potential risk of *S. haematobium* spread in Kyangwali around the swamps in the refugee settlement where *Bulinus* snails are breeding. Deforestation of Bugoma Forest to open land for agriculture, with introduction of livestock, and human settlement are the likely major factors leading to creation of suitable habitats for the intermediate host snails. The observed effects of environmental change, e.g. flooding of swamps in the encroached parts of Bugoma Forest, coupled with influx of people from schistosomiasis endemic areas, and breeding of intermediate host snail species pose a potential epidemiological risk.

The significantly higher average water temperatures in sites that were encroached more than ten years ago than in sites which are close to the forest shows that there is rapid environmental change due to loss of the microclimate modification effects of trees. This supports the earlier reports of Twongirwe and Nuwangira who predicted that deforestation would cause rapid change in climatic elements in this region [10, 12, 17].

Abundance of snails was significantly greater ( $p = 0.002$ ) in sites where the forest was cleared earlier than in the recently opened areas and schistosome cercariae shedding was also found in *Biomphalaria* and *Bulinus* species in the same zone with monthly maxima in December 2023 of 3.1% and 3.7%, respectively. This means that snail invasion and colonization of swamps is likely to have been gradual after forest destruction. The infested sites were around the refugee settlement where human and animal contact with swamp water was high. On the other hand, deforestation at sites 6, 7, 8 was still ongoing, and human and animal contact was minimal. The site 9, at the forest edge did not have *Biomphalaria* species snails, perhaps because of the low water temperature and phytochemical properties of a wide variety of forest plant leaves which drop in the water. Some of these leaves might be potentially toxic or have repellent properties to snails as earlier reported [34, 35]. Our findings support the results of Oso et al. 2021, who also observed an increasing number new fresh water snail breeding sites following deforestation in Yawa [14].

Parasitological screening of children around the two human water contact sites where snails were found shedding schistosome cercariae was a valid epidemiological response to ascertain the disease-risk as flagged by malacology. The prevalence of *S. mansoni* among school-aged

children living near site 2 (Puddle) was 56.1% implying that detection of cercariae in snails was a real-time environmental signal of *S. mansoni* transmission. Without such malacological surveys the disease might spread and cause severe morbidity without early detection. Strengthening snail monitoring and building sustainable approaches for its implementation could be considered an integral component of schistosomiasis control to address challenges of emerging transmission as described in WHO intermediate host snail control response guidelines of 2017.

Although there was no case of *S. haematobium* amongst the 41 resident school-aged children examined, the survey was vital to rule out urogenital schistosomiasis in such refugee communities who might have originated from areas where the parasite might be endemic. The shedding of *Schistosoma* cercariae by *B. forskalii* indicates the importance of malacology in One-Health because this snail is a potential intermediate host of *S. haematobium* and *S. bovis*, yet the cercariae of the two are morphologically indistinguishable. Although formerly not recognised as a potential intermediate host of human schistosomes, a recent study, Gaye et al., 2023, has described emerging potential of *B. forskalii* to transmit *S. haematobium* in some parts of Africa [36]. The maxima of cercaria shedding (3.7%) in December 2023 occurred during flooding of site 4 (Kigoma swamp) and increase in the *B. forskalii* numbers. This snail species appears to thrive better during the rainy season than the dry season, when they perhaps undergo aestivation.

Molecular analysis identified cercariae shed by *B. forskalii* at site 4 to be *S. bovis*, a *Schistosoma* species that infects livestock typically cattle, and has been previously reported in animals on the Lake Albert shoreline [37]. The presence of this schistosome in snails explains why all the 41 screened children near site 4 were negative for urogenital schistosomiasis as noting cercariae from *Bulinus* species can be an important environmental flag. Nonetheless, with continuous arrival of more refugees from endemic areas of South Sudan and DRC, there remains a potential risk of introducing *S. haematobium* miracidia to local *Bulinus* snails though their compatibility with the parasite in Western Uganda is not well known [38]. In 2024, Tumusiime et al. also described high abundance of *Bulinus* species to be a potential risk for urogenital schistosomiasis transmission if *S. haematobium* is introduced in the Lake Albert region [39], though our study did not find any of the species they reported except *B. forskalii*.

Molecular identification of *S. rodhaini* and *S. mansoni* in *Biomphalaria* snails at the Site 2 explains the high prevalence of *S. mansoni* we found among the forty-one local resident school-aged children. Transmission of *S. mansoni* and *S. rodhaini* by the same snail species

at a single site has not been noticed before in the Albertine region of Uganda. Although uncommon, this points to potential future complications which might arise in schistosomiasis morbidity if the two *Schistosoma* species hybridise in the human host as speculated by Leger and Webster 2016 [40].

As a limitation of the study, we only screened children in two villages where snails shed *Schistosoma* cercariae. Conducting epidemiological surveys around all sites that yielded *Biomphalaria* species would provide additional information on the schistosomiasis risk in Bugoma, but this was beyond the study scope. Molecular confirmation of cercariae was conducted on few shedding snails, thus, we might have missed some other species. Future studies to characterise all forms of cercaria shed by intermediate host snails might provide insightful data.

## Conclusions

Unprecedented encroachment on Bugoma Forest in the last two decades has potentially accelerated environmental change and created suitable foci for *Biomphalaria* and *Bulinus* intermediate host snail species, with a consequential expansion of schistosomiasis transmission into new areas away from Lake Albert shores. Settlement of displaced people in the deforested areas also has the potential for the introduction of other human and livestock schistosomes. We therefore encourage the Ministry of Health to strengthen surveillance of schistosomiasis for early detection of new transmission foci and the National Forestry Authority of Uganda to better mitigate local deforestation.

## Abbreviations

|          |                                               |
|----------|-----------------------------------------------|
| DNA      | Deoxyribonucleic acid                         |
| PCR      | Polymerase chain reaction                     |
| Cox1/COI | Cytochrome oxidase 1                          |
| ITS      | Internal transcribed spacer                   |
| HRM      | High-resolution melting curve                 |
| LULC     | Land-use and land cover                       |
| DRC      | Democratic republic of congo                  |
| FTA      | Flinders technology associate                 |
| GPS      | Geographical positioning system               |
| LQAS     | Lot quality assurance sampling                |
| NCBI     | National center for biotechnology information |

## Supplementary Information

The online version contains supplementary material available at <https://doi.org/10.1186/s44329-026-00061-x>.

Supplementary Material 1

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## Author contributions

DOW, NB and JRS developed the research idea and framework. DOW, CK, BF, JRS, CUT, MO and DOW designed the methods and conducted field work. DOW, CK, BF, DK, MO, JRS, analysed the data and interpreted the results. DOW and CK prepared the initial manuscript. AJ, TH and BW conducted the molecular analyses and reviewed the manuscript. JRS, NB, CUT and AE critically reviewed the manuscript for intellectual content. CUT, JRS, TH and NB provided oversight and guidance throughout the research process. JRS, NB, AE and CUT wrote the grant which obtained NIH funding to support this study. All authors read and approved the manuscript.

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## Data availability

The key raw data supporting the findings of this study are included in the manuscript results and supplementary file. Sequences for molecular identification of *S. mansoni* and *S. rodhaini* can be accessed from the NCBI- NIH repository.

## Declarations

### Ethics approval and consent to participate

This was a sub study of the Uganda Schistosomiasis Multidisciplinary research centre project which obtained ethical approval from the Uganda National Council for Science and Technology (HS2568ES 24/3/2023) after initial approval by the Research Ethics Committees of the Uganda Virus Research Institute (GC/127/940 on 29-11-2022). The Research Ethics Committee of Mbarara University of Science and Technology further reviewed the environmental health scientific content and approved the sub-study (MUST-2024-1350 on 24/5/2024). The study was conducted in accordance with the Uganda National Council for Science and Technology (UNCST) regulatory framework, which operates on the foundational ethical principles of the Declaration of Helsinki. Persons conducting snail sampling wore chest waders and heavy-duty gloves to prevent exposure to cercariae. The district health team and local leaders at every administrative level of sampled sites were sensitized on the objectives and importance of the study to obtain permission to work in the area. Parasitological screening of children for schistosomiasis was a mandatory public health response by the Ministry of Health and Kikuube District Health Department, whose procedures adhered to the Helsinki Declaration (<https://www.wma.net/policies-post/wma-declaration-of-helsinki/>). Informed consent for the school-aged children to participate in screening was obtained from parents/guardians and community leaders, while children gave verbal assent. Children found infected with *S. mansoni* were treated at their local health centre using praziquantel provided by the Ministry of Health.

### Consent for publication

This is not applicable to the study because we did not collect identifying images or other personal or clinical details of participants that compromise anonymity.

### Competing interests

The authors declare no competing interests.

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