

Bwindi-Sarambwe 2018 Surveys

Monitoring Mountain Gorillas, Other Select Mammals, and Human Activities



MAX-PLANCK-GESELLSCHAFT



Bwindi-Sarambwe 2018 Surveys

Monitoring Mountain Gorillas, Other Select Mammals, and Human Activities

FINAL REPORT

December 2019

Authors: Jena R. Hickey, Eustrate Uzabaho, Moses Akantorana, Joseph Arinaitwe, Ismael Bakebwa, Robert Bitariho, Winnie Eckardt, Kirsten Gilardi, Jacques Katutu, Charles Kayijamahe, Elizabeth M. Kierepka, Benjamin Mugabukomeye, Altor Musema, Henry Mutabaazi, Martha M. Robbins, Benjamin N. Sacks, Gladys Kalema Zikusoka

Citation: Hickey, J.R., Uzabaho, E., Akantorana, M., Arinaitwe, J., Bakebwa, I., Bitariho, R., Eckardt, W., Gilardi, K.V., Katutu, J., Kayijamahe, C., Kierepka, E.M., Mugabukomeye, B., Musema, A., Mutabaazi, H., Robbins, M.M., Sacks, B.N., and Zikusoka, G.K. 2019. Bwindi-Sarambwe 2018 Surveys: monitoring mountain gorillas, other select mammals, and human activities. GVTC, IGCP & partners, Kampala, Uganda, 40p.

Table of Contents

Executive Summary.....	5
Introduction	9
Methods.....	10
Study Site	10
Bwindi-Sarambwe Ecosystem	10
Community Outreach & Engagement.....	11
Field Methods	11
Sweeps	11
Reconnaissance Routes “Recces”	12
Vegetation typing.....	13
Sample Collection	13
Laboratory Methods	14
DNA Extraction	14
Probability of Identity and Genotyping Error	15
Individual Assignment of Gorillas.....	16
Group Assignment of Gorillas	16
Parasite and Viral Analyses	17
Analytical Methods	18
Spatial & Tabular Analyses.....	18
Results.....	18
Reconnaissance Routes “Recces”	18
Mountain Gorillas	19
Spatial Analysis.....	19
Field Sampling and Genetic Data Analysis	20
Gorilla Group Composition and Minimum Number Detected.....	20
Parasites.....	24
Select Mammals.....	25
Human Activities	31
Discussion.....	33
Mountain Gorillas	33
Parasite and Viral Analyses	33
Select Mammals.....	34
Human Activities	35

Future Work 36

Conclusions 36

Recommendations 37

Acknowledgements 37

Literature Cited 39



Doreen Chemayek & Léonard Rwambibi sample dung, as Team Lead Léonard Mugiraneza oversees them during extensive training.

Bwindi-Sarambwe 2018 Surveys

Monitoring Mountain Gorillas, Other Select Mammals, and Human Activities

Executive Summary

Long-term monitoring of wildlife populations allows population trends to be characterized from periodic robust abundance estimates. Based on those derived trends, conservation status of the species and conservation efforts may both be assessed. As such, for decades researchers and Protected Area Authorities have estimated the abundance of mountain gorillas in both Bwindi Impenetrable National Park and the Virunga Massif through collaborative survey efforts. Here, we report on the results of the Bwindi 2018 surveys of mountain gorillas, other select mammals, and human activities. As in recent surveys (e.g. Roy *et al* 2014, Hickey *et al* 2019, Granjon *et al* in press), field teams walked pre-determined compass bearings termed 'reconnaissance trails' through the forest of the Bwindi-Sarambwe

ecosystem in two separate sweeps to ensure thorough coverage of all areas while searching for signs of mountain gorillas, other select mammals, and illegal activities. When a fresh or recent gorilla trail was detected, the teams followed it to locate, optimally, three recent nest sites for each gorilla group or solitary individual. At each of these sites, the teams collected fecal samples from gorilla nests (Photo 1) that were genotyped to individual for a minimum count of mountain gorillas in the Bwindi-Sarambwe ecosystem.

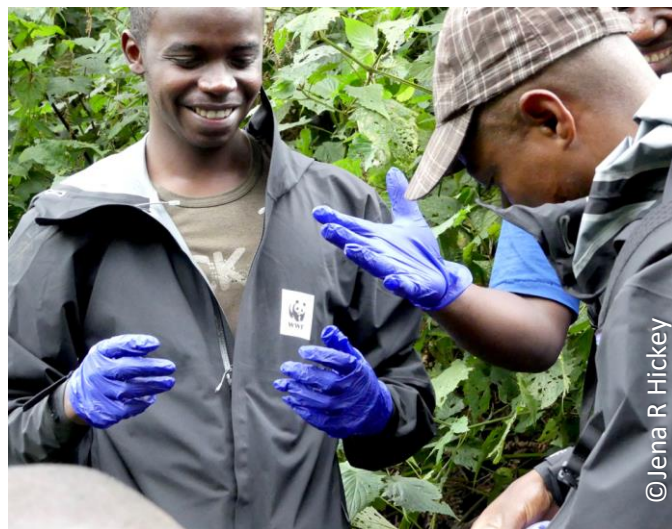


Photo 1. Team members prepare for biological sampling

We estimated a minimum count of 459 gorillas based on the number of unique consensus genotypes of unmonitored gorillas ($n=263$) detected during this survey plus the known number of monitored gorillas ($n=196$). A minimum count does not equate to a total population estimate because not all gorillas are detected in such surveys. For example, only 1 of 13 solitary individuals and 14 of 33 unmonitored groups were detected in both sweep 1 and 2. The remaining gorillas were only detected in one of the two sweeps. Therefore, detection probabilities and an associated total abundance estimate for the Bwindi-Sarambwe subpopulation of mountain gorillas are forthcoming in a separate document pending further mark-recapture analyses of the gorilla detection histories following similar previous approaches (Roy *et al* 2014, Granjon *et al* in press).

Compared to the 2011 survey estimate of 400 individual gorillas (an estimate that included correction factors for 37 infants and/or individuals that were potentially undetected in the genetic analysis; Robbins *et al* 2013, Roy *et al* 2014), the 2018 minimum count of 459 gorillas (that included no correction factors and is a true minimum) confirms that the Bwindi-Sarambwe mountain gorilla population grew during the intervening period.

Although two sweeps were conducted in 2011 for estimating the gorilla abundance, only one sweep in 2011 included the survey of other large mammals and human activities. Therefore, the 2018 Bwindi-Sarambwe Survey represents approximately twice the effort made in 2011 in terms of total kilometers walked while recording other select mammals and human activities (IGCP unpub. data). The field sampling effort for the second sweep in 2011 and each individual sweep in 2018 was sufficiently comparable in terms of distance walked that we assumed that detection probabilities of mammal and human-activity signs were similar in each individual sweep, and that all three sweeps could thus be compared to each other. For comparing within the same season, the second sweep of both 2011 and 2018 took place from September to December.

Sightings of black-fronted duikers (*Cephalophus nigrifrons*), bushbucks (*Tragelaphus scriptus*), and bushpigs (*Potamochoerus larvatus*) all had roughly similar encounter rates in 2011 and 2018, whereas all other mammals surveyed exhibited much higher encounter rates in 2018 than in 2011. For example, we recorded 0.615 and 0.725 encounters/km for elephant dung, in sweeps 1 and 2 of 2018 respectively, whereas 0.518 encounters/km were reported in 2011. For chimpanzee nests, we recorded 0.463 and 0.679 encounters/km in sweeps 1 and 2 of 2018, respectively, whereas 0.288 encounters/km were recorded in 2011.

While we do not infer population trends from surveys of indirect signs, these data suggest at least a relatively stable status for the other select mammals surveyed, as they provide no indications of population declines since 2011. The information collected will inform species-distribution models for a better understanding of the population ecology of several species of mammals in relation to abiotic and biotic factors, including the potential influence of human activities in shaping their spatial distributions.

Data suggest that illegal activities in the Bwindi-Sarambwe ecosystem also have not declined since 2011, despite formidable conservation efforts in both law enforcement and community engagement. For example, the survey teams destroyed 88 snares during the 2018 surveys. Snare encounter rates were roughly similar between 2011 and 2018; we recorded 0.042 and 0.055 encounters/km in sweep 1 and 2 of 2018, respectively, compared to 0.058 encounters/km reported in 2011. For comparison, snare-encounter rates in the Virunga Massif were reported as 0.15 and 0.09 snare encounters/km in 2015 and 2016, respectively (Hickey *et al* 2019), suggesting that although snare encounter rates observed in Bwindi-Sarambwe appear not to have declined since 2011, they remain substantially lower than in a similar nearby ecosystem.

The results of this collaborative survey provide conservation practitioners valuable information to help assess past, and inform future, management actions. The findings here highlight areas of conservation progress and areas where more effort appears necessary. Furthermore, the 2018 Bwindi-Sarambwe surveys generated the requisite baseline data to inform many related studies, from the potential influences of human activities on wildlife to the production of niche models based on associations between species occurrences, land cover, and other variables. Together, those future studies can also bolster the collective understanding of what influences species distributions and offer insights into ways to further support biodiversity conservation in this landscape.

Résumé Français

Le suivi à long terme des populations de faune permet d'en définir les tendances à partir d'estimations régulières et rigoureuses de leur abondance. Les tendances ainsi dérivées permettent d'évaluer le statut et les efforts de conservation de l'espèce. Grâce à des inventaires collaboratifs, les chercheurs et les autorités en charge des aires protégées ont ainsi estimé au cours des décennies l'abondance des gorilles de montagne dans le Parc national de la forêt impénétrable de Bwindi et dans le Massif des Virunga. Nous présentons ici les résultats des recensements des gorilles de montagne, des autres mammifères et des activités humaines réalisés à Bwindi en 2018. Comme lors de recensements récents (ex. Roy et al 2014, Hickey et al 2019, Granjon et al sous presse), les équipes ont parcouru, à l'aide d'un compas, des « sentiers de reconnaissance » prédéterminés dans la forêt de l'écosystème de Bwindi-Sarambwe, en deux balayages distincts pour couvrir toute la zone, recherchant les signes de présence des gorilles de montagne, d'autres mammifères sélectionnés et d'activités illégales. Lorsque ces équipes découvrent un sentier frais ou récent de gorilles, elles le suivent dans l'espoir de trouver idéalement trois sites récents de nidification d'un groupe de gorilles ou d'un individu solitaire. À chacun de ces sites, les équipes recueillent dans les nids des échantillons fécaux (Photo 1), qui sont ensuite analysés génétiquement pour distinguer les génotypes individuels et déterminer ainsi le nombre minimum des gorilles de montagnes dans l'écosystème de Bwindi-Sarambwe.



Photo 1. Préparation pour l'échantillonnage biologique par des membres d'équipe

Nous estimons le nombre minimum de gorilles à 459, sur la base du nombre de consensus de génotypes uniques de gorilles non suivis ($n=263$) trouvé lors de cet inventaire, ajouté au nombre connu de gorilles suivis ($n=196$). Ce nombre minimum n'est pas égal à l'estimation totale de la population, car ce type de recensement ne permet pas de repérer tous les gorilles. Par exemple, seuls 1 sur les 13 individus solitaires et 14 sur les 33 groupes ont été repérés lors du premier et du deuxième balayage. Les autres gorilles n'ont été repérés que lors d'un seul balayage. Par conséquent, les probabilités de détection et une estimation associée de l'abondance totale de la sous-population de gorilles de montagne de Bwindi-Sarambwe seront présentées dans un document séparé, en attendant les analyses de marquage-recapture de l'historique de détection des gorilles selon des approches similaires (Roy *et al* 2014, Granjon *et al* sous presse).

Si deux balayages ont été effectués en 2011 pour estimer l'abondance des gorilles, un seul a porté sur l'inventaire d'autres grands mammifères et d'activités humaines. Ainsi, le recensement de 2018 à Bwindi-Sarambwe représente à peu près le double de l'effort de 2011, en termes de kilomètres parcourus pour relever les signes de certains mammifères et d'activités humaines (données non publiées du PICG). L'effort d'échantillonnage du second balayage en 2011 et de chaque balayage en

2018 étant comparable en termes de distance parcourue, nous supposons que les probabilités de détection des signes de mammifères et d'activités humaines sont similaires pour chaque balayage et que, par conséquent, les trois balayages sont comparables entre eux. Concernant la comparaison saisonnière, le deuxième balayage en 2011 comme en 2018 a eu lieu entre septembre et décembre.

Les observations de céphalophes à front noir (*Cephalophus nigrifrons*), de guibs harnachés (*Tragelaphus scriptus*) et de potamochères (*Potamochoerus larvatus*) ont eu des taux de rencontre similaires en 2011 et en 2018, tandis que les taux de rencontre des autres mammifères inventoriés étaient bien plus élevés en 2018 qu'en 2011. Nous avons par exemple des taux respectifs de 0,615 et 0,725 rencontres/km d'excréments d'éléphant lors des balayages 1 et 2 en 2018, par rapport à 0,518 rencontres/km en 2011. Concernant les nids de chimpanzés, nous avons des taux respectifs de 0,463 et 0,679 rencontres/km lors des balayages 1 et 2 en 2018, par rapport à 0,288 rencontres/km en 2011.

Bien que nous ne déduisons pas les tendances des populations à partir des signes indirects, ces données dénotent d'un statut relativement stable des autres mammifères étudiés, car il n'y a aucune indication de déclin des populations depuis 2011. Les informations rassemblées seront utilisées dans les modèles de distribution des espèces, afin de mieux comprendre l'écologie de la population de plusieurs espèces de mammifères par rapport à des facteurs abiotiques et biotiques, y compris l'influence potentielle des activités humaines sur leur distribution spatiale.

Les données indiquent aussi que les activités illégales dans l'écosystème de Bwindi-Sarabwé n'ont pas diminué depuis 2011, malgré des efforts considérables de conservation en termes à la fois d'application des lois et d'engagement des communautés. Les équipes ont ainsi détruit 88 pièges lors des recensements de 2018. Les taux de rencontre sont à peu près similaires pour les pièges en 2011 et 2018 ; nous avons des taux respectifs de 0,042 et 0,055 rencontres/km lors des balayages 1 et 2 en 2018, par rapport à 0,058 rencontres/km en 2011. En comparaison, les taux pour les pièges dans le Massif des Virunga étaient respectivement de 0,15 et 0,09 rencontres/km en 2015 et 2016 (Hickey *et al* 2019), ce qui indique que même si les taux de rencontre des pièges à Bwindi-Sarabwé ne semblent pas avoir baissé depuis 2011, ils restent sensiblement inférieurs à ceux enregistrés dans un écosystème similaire voisin.

Les résultats de cet inventaire collaboratif apportent aux professionnels de la conservation des informations précieuses pour évaluer le passé et guider les actions de gestion futures. Ces résultats mettent en évidence les domaines où la conservation a enregistré un progrès, mais aussi ceux où plus d'efforts semblent nécessaires. Par ailleurs, les recensements effectués en 2018 à Bwindi-Sarabwé ont produit les données de base préalables à de nombreuses études associées, telles que celles sur les impacts potentiels des activités humaines sur la faune ou celles sur la production de modèles de niche basés sur les associations entre la présence des espèces, la couverture végétale et d'autres variables. Ces futures études peuvent aussi renforcer la connaissance collective des facteurs qui influencent la distribution des espèces et apporter des éléments pour appuyer la conservation de la biodiversité dans ce paysage.

Introduction

Long-term monitoring of wildlife populations enables the assessment of species status, conservation efforts, and the effects of numerous variables including potential impacts of hunting, land-use change, climate change, and other disturbances on species of interest. The Bwindi-Sarambwe ecosystem is a protected medium-to-high elevation tropical rainforest located in Uganda and the Democratic



Photo 2. Bwindi-Sarambwe is a vast rainforest ecosystem in the mountains of Uganda and the Democratic Republic of Congo (DRC). Periodic and intensive surveys as represented in this report complement finer-scale patrol efforts.

Republic of Congo (DRC). Butynski and Kalina (1993) reported extraordinary biodiversity there for numerous taxa and select mammals have been periodically surveyed since the late 1990s with ever increasing effort (McNeilage *et al* 2001, 2006; Guschanski *et al* 2009; Gray *et al* 2013; Robbins *et al* 2012, Roy *et al* 2014). Human pressures surround and impact the Bwindi-Sarambwe ecosystem and have done so for decades; in fact, deforestation in Uganda outside the parks was described as “nearly complete” as of 1993 (Butynski and Kalina 1993). Around Bwindi Impenetrable National Park, over 90% of households practice subsistence agriculture (Korbee 2007) and human population densities exceed 250 people/km² (Boffa *et al* 2005). Even within the forests now protected, natural resources were heavily extracted via hunting or vegetation-cutting prior to 1993 (Butynski and Kalina 1993; Twongyirwe *et al* 2011) and human population growth continues to exert negative effects on the forest through encroachment due to the ever-increasing need for food and wood (Twongyirwe *et al* 2011). Such historical and relatively recent habitat losses imposed on a veritable island of mountain-top refugial rainforest, combined with persistent poaching of wildlife, require vigilant wildlife monitoring, law enforcement, community engagement, and conservation action. To that extent, periodic surveys of the entire Bwindi-Sarambwe ecosystem complement routine monitoring and patrolling activities that occur at finer scales. The ecosystem-wide surveys, termed ‘sweeps’ (McNeilage *et al* 2001, 2006; Guschanski *et al* 2009; Gray *et al* 2009, 2013; Roy *et al* 2014), cover virtually the entire area including remote locations that are rarely patrolled, and therefore provide the opportunity to survey for illegal activities and destroy snares in far reaches of the ecosystem (Photo 2).

This report provides another benchmark of updated information regarding the minimum count of mountain gorillas and the relative encounter rates of other select mammals, as well as human activities, in two successive sweeps of the ecosystem within a single study. We compare these relative encounter rates from the previous survey completed in 2011 (Robbins *et al* 2012) to the recently completed survey of 2018 (this report), the latter of which accomplished approximately twice the survey effort as the former (select mammal and human-activity signs were reported only for the second sweep of the Bwindi 2011 survey). Although we expected to effectively double the

effort of the 2011 survey (IGCP unpub. data) by monitoring select mammals and human activities in two full sweeps of the ecosystem in 2018, small but relevant adjustments to our protocol may have resulted in a greater-than-intended increase in survey effort. For example, 2018 was the first full survey of Bwindi-Sarambwe to eliminate paper data entry and instead employ electronic devices for recording all data, including geo-referenced positions of every observation.

Furthermore, to reduce data-entry time, as well as to

facilitate team movement through the ecosystem, we refined the number and type of mammal and human-activity signs compared to 2006 and 2011 protocols. Hence, it is possible that the encounter rate of these signs in the current study may appear increased since 2011, simply as an artifact of observers being able to more easily focus their search for the signs of primary interest. Further, this effort provides the requisite baseline data to inform several related investigations, from the influences of human activities on wildlife to the production of niche models based on associations between land cover and species occurrences.

Methods

Study Site

Bwindi-Sarambwe Ecosystem

The Bwindi-Sarambwe ecosystem is comprised of Bwindi Impenetrable National Park (BINP) in Uganda and the Sarambwe Nature Reserve (SNR) in the DRC, together encompassing approximately 340 km² (Figure 1). A narrow corridor, locally referred to as the ‘neck’, characterizes BINP between the core southern and far northern portions of the protected area. Elevation ranges from 1160 to 2607 m above sea level (McNeilage *et al* 2001) and annual rainfall ranges from 1400 to 1900 mm (Twongyirwe *et al* 2011). Correspondingly, bamboo or mixed bamboo, bracken fern, grassland, herbaceous, meadow, *Mimulopsis*, mixed forest, *Neobutonia* trees, and swamp characterize most of the landcover in the ecosystem (this study and Nkurunungi *et al* 2004). Climate is characterized by two rainy and two dry seasons per year. Usually, the rainy seasons span March through May and September through November. For logistical efficiency, we divided this ecosystem into 40 sectors ranging in size from 4.4 to 17.4 km², the 40th being SNR (Figure 1). The surveyed area encompassed approximately 340 km² and 335 km² in sweeps 1 and 2, respectively.

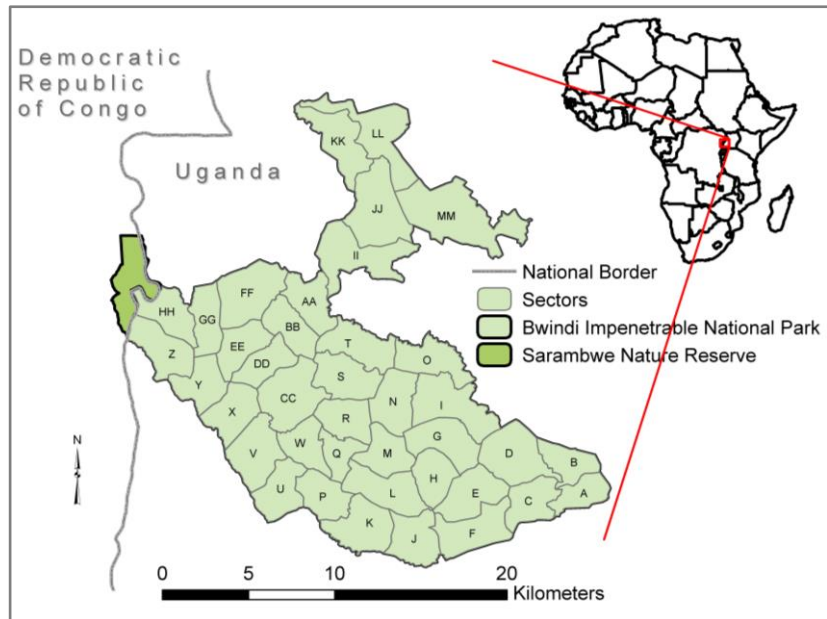


Figure 1. The Bwindi-Sarambwe ecosystem encompasses approximately 331 km² in Uganda and 9 km² in the Democratic Republic of Congo. The sectors indicated with letters helped organize the field work.

Community Outreach & Engagement

Prior to the survey, mobilization and sensitization meetings were held from 26 February to 4 March 2018 at various points around BINP to create awareness and solicit support and participation of community members in the exercise. The areas visited included Nkuringo, Rubuguri, Nyabwishenya, Kisoro district headquarters, Cyanika border, Rubanda district headquarters, Ikumba subcounty headquarters, Ndego, Ruhija, Buhoma, Bwashwa, and Kanungu district headquarters. A team composed of members from UWA, ITFC, and IGCP undertook this exercise and targeted meeting with local communities, local leaders, security personnel, immigration officers, and suppliers of goods and services in the three districts of Kanungu, Kisoro, and Rubanda.

Specifically, the audience intended for this mobilization exercise included key stakeholders and duty bearers such as District Chairmen, Resident District Commissioners, Chief Administrative Officers, District Internal Security Officers, District Police Commanders, District UPDF Commanders, Immigration Officers, Local Council Chairmen, Porter associations, and identified potential suppliers of perishable food.

The main purpose of the meetings was to let the audience know of the forthcoming survey, which would involve a large group of participants from Uganda together with partners from Rwanda and Democratic Republic of Congo with regular cross border movements between Rwanda and Uganda starting on 4 March 2018.

During these meetings local communities also were invited to participate in survey support by helping to reopen the trails, serving as porters, and supplying fresh produce to the teams.



Photo 3. Vastine Tindimwebwa (in camouflage) leads Team 2 as they record observations during the Bwindi-Sarambwe 2018 surveys

Field Methods

Sweeps

The field-survey approach was generally based on past protocols (Sholley 1991; McNeilage *et al* 2001, 2006; Gray *et al* 2009, 2013; Guschanski *et al* 2009) and modified in a similar manner as in Roy *et al* (2014) to collect two occasion histories for non-invasive genetic mark-recapture abundance estimation of gorillas (reported separately).

Starting with the eastern sectors and progressing toward the west, each sector was surveyed by two field teams that searched for both direct and indirect observations of wildlife and illegal activities (Photo 3). As described in Hickey *et al* (2019), teams typically included two trackers, an armed ranger, and one or two data recorders, for a total of four to five members. Often data recorders and rangers also had tracking skills. Once a sector was completed, teams moved to a new sector to resume surveys, until all 40 sectors were completed. Typically, six teams worked in two-week shifts and then rotated out with fresh teams replacing them for the subsequent two weeks, until a single survey of the entire ecosystem – termed a ‘sweep’ – was complete. Teams conducted two sweeps in

2018: the first occurred from March to May (62 days), and the second from October to December (60 days), both corresponding to rainy seasons.

Reconnaissance Routes
"Recces"

To survey a sector, field teams hiked through the vegetation, typically following an initial pre-determined bearing until they came within 200 m of a boundary (either of the sector or the protected area). Adjacent reconnaissance routes, termed 'recces', were spaced approximately 500 m apart. Recces departed from the bearing, becoming irregular, when teams circumnavigated obstacles such as ravines, and when teams detected fresh or recent gorilla trails. For direct and indirect observations of mountain gorillas and other select mammals, as well as human activities, teams recorded the age, species, and type of sign (e.g. track, dung, vocalization heard, sighting). Table 1 describes the complete set of species and types of mammal observations recorded. Note that some species and sign types that were included in 2006 or 2011 surveys were not incorporated into the protocol of this study. Table 2 describes the human activities and types of observations recorded. Note again that some types of human activities that were

Table 1. Species, types of observations, and manner of aging signs

Common Name	Observation Type	Age of Sign (days)	Latin Name
Black-fronted duiker			<i>Cephalophus nigrifons</i>
Yellow-backed duiker	Sighting	NA	<i>C. silvicultor</i>
Bushbuck			<i>Tragelaphus scriptus</i>
Sitatunga			<i>T. spekii</i>
Bushpig			<i>Potamochoerus larvatus</i>
Baboon			<i>Papio Anubis</i>
Black-and-white colobus	Heard	NA	<i>Colobus guereza</i>
Blue monkey			<i>Cercopithicus mitis</i>
L'Hoest's monkey	Sighting		<i>C. lhoesti</i>
Red-tailed monkey			<i>C. ascanius schmidti</i>
Elephant or Carnivore	Dung	Fresh (0-1d) Recent (2-4d) Old (>4d)	<i>Loxodonta africana,</i> <i>Canis adustus, Caracal aurata, Civettictis civetta, Leptailurus serval, or Mellivora capensis</i>
	Scraping		
	Tracks	NA	
	Heard		
Chimpanzee	Sighting	Fresh (0-1d) Recent (2-4d) Old (>4d)	<i>Pan troglodytes t.</i>
	Dung		
Mountain Gorilla	Tracks	To the date, if possible, or Fresh (0-1d) Recent (2-5d) Old (>5d)	<i>Gorilla beringei b.</i>
	Nest Sites		
	Heard	NA	

included in 2006 or 2011 surveys were not incorporated in 2018 protocols. Teams entered all data into rugged, handheld electronic devices (Toughpad FZX1, Panasonic™) equipped with Cybertracker (<http://www.cybertracker.org>) software that was customized for this survey. In addition, teams plotted their location on paper maps at ‘control points’ every 250 m to track their progress and survey coverage for coordination with other teams. These control points were also logged in the electronic devices.

Vegetation typing

In a similar protocol as used in the Virunga 2015-2016 Surveys (Hickey *et al* 2019), teams recorded the dominant vegetation type within a 10-m radius around every observation, not only where mammal signs were recorded, but also at every control point and sign of human activity. Dominant vegetation was categorized into the vegetation types described above in *Study*

Site, with the additional category of ‘former cultivation’ to record previously disturbed vegetation. Geo-referenced locations of vegetation types collected during this survey will be used as ground-truth data in a supervised landcover classification of remotely sensed imagery (WWF-Germany and IGCP *in progress*). That separate effort will eventually contribute to future studies relating species occurrences to vegetation and land-use change, as well as larger planning efforts.

Sample Collection

When teams encountered gorilla trails that were estimated (based on field evidence) to be fresh or recent (≤ 5 days old), they left the bearing of the recce to follow the trail seeking a gorilla nest site. Once at a nest site, teams assigned the gorilla group a unique identity (either an alpha-numeric code or, if a known monitored group, then the group’s name), searched for each nest (ground and arboreal), and collected fecal samples from every nest that contained ≥ 1 dung. If dung diameters were markedly different within a nest, each dung was sampled separately. Each sample collected was associated with data regarding the sector, group ID, nest site ID, nest ID, individual’s estimated sex and age class, date of collection, estimated age of the sample, and GPS coordinates. Rarely, nests were so high in vegetation as to be inaccessible. In those cases, teams recorded nests as having no dung in them, because none could be collected. Based on field evidence for each detected group, teams followed the gorilla trails and aimed to sample all nests from 3 different nest sites per group – ideally from 3 consecutive nest sites including one fresh nest site from the previous night.

Table 2. Human activities, types of observations, and manner of aging signs

Human Activity	Observation Type	Age of Sign (days)
Poaching	Snare	NA
	Poacher	
	Animal in Snare	Fresh (0-1d) Recent (2-4d) Old (>4d)
Wood cutting	Bamboo Cut	
	Firewood Cut	Fresh (0-1d) Recent (2-4d) Old (4-30d)
	Pitsaw	
	Pole Cut	
Other	Tree Cut	
	Dogs	NA
	Fire	
	Camp	



Photo 4. Field crew trainees, Donathile Mukamana and Doreen Chemayek, collect fecal sample for genetic analysis to individual

Information from genetic analyses would later help confirm or correct which gorilla group constructed each nest site.

We collected all genetic samples following the two-step procedure (Nsubuga et al 2004). In the field, we collected approximately 4 g of feces (about the size of a teaspoon) in a tube containing 99% ethanol such that the entire sample was submerged (Photo 4). After 24-30 h, the ethanol was removed, and samples were transferred into tubes filled with silica beads to complete desiccation. We then stored silica tubes at room temperature

until exportation to UC Davis. Once at UC Davis, we stored samples at room temperature until extraction, then stored DNA extracts at +4°C while awaiting genotyping.

In addition to the genetic samples, during the first sweep, teams also collected fecal samples from each nest site for viral pathogen and parasite surveillance. For each gorilla dung sampled, an approximately 9-g piece of feces was placed in a plastic specimen tube containing 10% formalin, and two additional 4-g pieces were each placed in one of two plastic specimen tubes, one containing RNALater™ and the other containing 99% ethanol. We stored specimens preserved in formalin or ethanol for parasite analysis, and specimens preserved in RNALater™ were slated for virus detection. We aliquoted formalin samples into two portions, one for Gorilla Doctors and one for Conservation Through Public Health (CTPH). We aliquoted the samples preserved in RNALater™ into four portions originally slated for (1) analysis via Gorilla Doctors by the Institute of Vertebrate Biology in the Czech Academy of Sciences, (2) storage via Gorilla Doctors in the 'biobank', (3) analysis via CTPH, and (4) potential analysis via Robert Koch Institute. We stored specimens in formalin and ethanol at room temperature. Specimens stored in RNALater™ that were slated for the 'biobank' were transferred to the Gorilla Doctors laboratory at COVAB, Makerere University, and placed in a -80°C freezer for long-term storage. During the second sweep, teams collected only the single sample per gorilla dung (for genetic analyses only), preserved in 99% ethanol, and transferred to silica.

Laboratory Methods

DNA Extraction

We extracted 1884 fecal samples via Qiagen 96-well stool kits with several modifications. Each extraction contained 100 mg of dry fecal material that was incubated overnight in Qiagen ASL buffer. On the subsequent day, all extraction methods followed the manufacturer's instructions except we used potato starch instead of InhibitEX and incubated DNA extracts on the spin column for 30 minutes prior to elution.

DNA Amplification

Our genetic marker panel included 12 loci: one sex-specific locus (Amelo) and 11 autosomal microsatellite loci (vWF, D16s2624, D7s2204, D10s1432, D14s306, D3s2459, D5s1470, D4s1627, D2s1326, D1s550, and D6s1056) and were chosen based on previous analyses (Guschanski *et al* 2009, Roy *et al* 2014). We initially amplified each extract twice in two multiplex polymerase chain reactions (PCR) with 6 loci each (Multiplex 1: Amelo, vWF, D1s550, D4s1627, D5s1470, and D7s2204; Multiplex 2: D16s2624, D3s2459, D6s1056, D14s306, D2s1326, and D10s1432). Each 11 μ L PCR reaction contained 0.5 μ L of RNase free water, 5.0 μ L of Qiagen Multiplex Mastermix, 1.0 μ L of Q-solution, 2.5 μ L of primer mix (0.08-0.8 μ M of each primer in total reaction), and 2.0 μ L of DNA extract. All PCR reactions were amplified via the following thermal protocol: initial denaturation at 95°C for 15 minutes, 33 cycles of 30 second denaturation at 94°C, 90 second annealing at 55°C, and 60 second elongation at 72°C, and a final elongation for 10 minutes at 72°C. All PCR products were electrophoresed on an ABI PRISM 3730 Genetic Analyzer and sized manually with the aid of STRand software (University of California, Davis). Replicate genotypes were merged into consensus genotypes. Samples with 10 or more loci successfully genotyped were used in analyses and those with <10 loci successfully genotyped were considered to have failed and were excluded from further consideration. Analyses showed high PCR success rates (82.2% samples had full genotypes), yielding 1548 consensus genotypes with ≥ 10 genotyped loci.

Probability of Identity and Genotyping Error

Two factors affect the accuracy of individual assignments of genotypes: resolution of the markers to differentiate close relatives and genotyping error, which can lead to false differentiation of samples from the same individual. Resolution is commonly measured in terms of the probability of two distinct individuals sharing the same genotype by chance (which is higher for siblings than random individuals in the population). Based on the marker set and dataset for the current study, we estimated the cumulative (i.e., all markers) probability of identity ($P_{ID} = 2.1 \times 10^{-9}$) and probability of identity of siblings ($P_{SIB} = 1.9 \times 10^{-4}$) to be extremely low. In general, any $P_{SIB} < 0.01$ is considered low enough to confidently distinguish individuals.

Fecal DNA samples often suffer from low quality and quantity, making them vulnerable to genotyping errors. The most common error is random allelic dropout where one of the two gene copies fails to amplify in a given PCR reaction; when this occurs in a homozygous locus, there is no error because amplification of either gene copy results in the same homozygous genotype. However, when it occurs in a heterozygous locus, allelic dropout results in a false homozygous genotype for that locus. The other type of error is less common, a false allele, whereby an allele is documented that does not in fact exist for either gene copy; this typically presents as a homozygous locus appearing as a heterozygote. In both cases, the error processes are random and their probabilities quantifiable, thereby enabling genotyping error rates to be reduced to a known level through replication of independent PCR reactions. We calculated both types of genotyping errors by replicating PCR of a subset of samples a sufficient number of times to confidently know the true genotypes and then comparing each replicate back to the presumed true genotype (the consensus genotype). Specifically, we arbitrarily selected 48 fecal samples and replicated PCR and genotyping steps 5-7 times, merged them into consensus genotypes presumed to be the true (error-free) genotypes, selected those for which ≥ 10 loci were successfully genotyped ($n = 39$), and compared each replicate to identify cases of allelic dropouts and false alleles. Genotyping errors were rare

(estimated at 2.5%) with 2.3% and 0.17% of samples exhibiting allelic dropout and false alleles, respectively. These errors were on the lower end of error rates typically reported for non-invasive fecal samples (Broquet and Petit 2004).

Individual Assignment of Gorillas

We used our quantified resolution and genotyping error rates described above to derive assignment rules that minimized misclassification probability. Specifically, we used a combined statistical and manual proofing procedure and additional replication of PCR and genotyping of ambiguous genotypes to assign fecal samples to individual gorillas, as described in detail by Lounsberry *et al* (2015). In brief, genotypes that were too similar (<2 mismatches) to be reasonably explained by shared parentage ($P < 0.005$) were considered to be from the same individual. Genotypes with number of allelic mismatches too high (>3 mismatches) to be reasonably explained by genotyping error ($P < 0.005$) were considered to represent distinct individuals. In rare cases (85 samples) where neither of these criteria was met (i.e., 2 or 3 mismatches), the samples were re-run 2 more times to verify genotypes. If mismatches occurred even after additional replication, they were considered distinct individuals.

Group Assignment of Gorillas

Following individual identification, we attempted to assign gorillas to one of 17 monitored groups, an unmonitored group, or as solitary (no other nests at nest site). Although the monitored group sizes were known from the long-term demographic monitoring by UWA, it was nevertheless necessary to determine which samples (and genotypes) reflected those groups to avoid double-counting (i.e., classifying some monitored groups as unmonitored). Assignment of samples to groups required multiple steps. First, field crews designated putative groups based on evidence at each nest site, recorded if a nest site likely belonged to a monitored or unmonitored group, and designated solitary gorillas (all of which were unmonitored) based on the presence of only one nest at a nest site (Table 3). These field assignments were conservative, presuming groups were unmonitored unless it was known with certainty that the group was one of the monitored groups. In the second step, we used genetic information to identify the individuals sampled together at one or more nest site during one or both sweeps, along with known locations of monitored groups, to obtain final assignments of each genotyped individual. For 11 of the 17 monitored groups, field crews assigned group names to nest sites in at least one sweep. Six monitored groups that were not identified during the field survey were later confirmed to have been among the groups detected and sampled based on known locations of monitored groups. In 5 cases, nest sites had multiple nests with failed amplifications where only a single individual was successfully genotyped. These individuals could not be assigned confidently to a particular group, yet they clearly were not solitary based on the association with other nests. Further, based on known locations of monitored groups and the full suite of nest sites where each of these individuals was detected, we were able to categorize their groups as monitored but unclassifiable to a particular group ($n = 2$) or unmonitored but unclassifiable to a particular group ($n = 3$). These two categories also included individuals sampled in nest sites of either multiple monitored or multiple unmonitored (but not both) groups. Lastly, 13 individuals were sampled in nest sites of both monitored and unmonitored groups and, therefore, considered 'unclassifiable' and excluded from minimum counts. For monitored groups, we report the known size based on daily monitoring. For unmonitored gorillas, we report the minimum

number, which equated to all sampled gorillas in the following categories: unmonitored groups, unmonitored solitary, and unmonitored but unclassifiable.

Table 3. Definitions of Training Samples and 6 mutually exclusive and exhaustive categories of individuals detected of the 2018 Bwindi-Sarambwe field survey.

Designation	Definition
Training Samples	Fecal samples of two monitored groups (Mukiza, Bitukura) collected prior to sweeps during field-team training
Monitored Groups	Seventeen groups monitored daily by UWA or MPI staff for 3 or more years and designated in the field with their known group names
Unmonitored Groups	All groups not considered among one of the 17 Monitored groups ^a
Unmonitored solitary	Nest sites or series of nest sites that each contained only a single nest and fecal sample
Monitored but unclassifiable	Single successful genotype at nest site with multiple nests and inferred indirectly to be associated with one or more monitored groups
Unmonitored but unclassifiable	Single successful genotype at nest site with multiple nests and inferred indirectly to be associated with one or more unmonitored groups
Unclassifiable	Sampled in nest sites of both monitored and unmonitored groups

^aTwo groups that have been monitored for <3 years (may be considered monitored groups in future once enough information is gained of their membership) are included here simply as “Unmonitored”

Parasite and Viral Analyses

We analyzed formalin-preserved gorilla fecal samples (n = 329) collected during sweep 1 for intestinal helminth (worm) parasites at Conservation Through Public Health (CTPH). We used a combination of flotation for lighter parasite eggs, sedimentation for heavier parasites eggs and the McMaster method to determine the fecal parasite egg counts per taxa.

Formalin and ethanol-fixed gorilla fecal samples (n = 666) were shipped by Gorilla Doctors (Uganda Wildlife Permit No. COD/96/05 and Wildlife Export License No. 32307) to the Institute of Vertebrate Biology in the Czech Academy of Sciences (Brno, Czech Republic) for application of classical and molecular methodologies and tools to identify and quantify helminths that may be of health consequence for Bwindi-Sarambwe gorillas, with an emphasis on strongylid nematodes and tapeworms. Employment of classical coproscopical methods (Mini-FLOTAC, sedimentation) and advanced high-throughput genetic sequencing (HTS) tools will be applied to detect, quantify and uncover the diversity of helminth infections in Bwindi-Sarambwe. Egg per gram feces counts will be conducted, and worms will be morphologically identified and subsequently sequenced to confirm species.

USAID PREDICT consensus PCR protocols that combine high sensitivity with broad reactivity (i.e. detect viruses at low levels while casting a wide net) were available to be applied (pending available resources) to enable the detection of both known and novel viruses in feces from viral families of potential consequence for gorilla and human health, including influenza, corona-, paramyxo-, filo- and flavi- viruses (Goldstein *et al* 2013; Anthony *et al* 2013 a,b).

Analytical Methods

Spatial & Tabular Analyses

Spatial analyses were conducted following Hickey *et al* (2019) using ESRI ArcGIS 10.6.1. Briefly, to estimate survey effort (distance walked in km), we used all observations and control points recorded by each team in each two-week phase of work and converted those points to lines, linking points consecutively in time within each day. We then merged all lines for a sweep into a single shapefile to determine the total km per sweep. For each sweep's km layer, we conducted a neighborhood analysis (focal statistics) with windows of 1-km radius to produce a raster of km (walked) per window that depicted survey effort spatially across the Bwindi-Sarambwe ecosystem. Areas >600 m from recces were considered outside the survey effort (e.g. northern portion of SNR during sweep 2).

To compare 2018 mammal and human activity results to 2011 results, we presented tabular data in the same manner as in 2011, specifically, as encounter rates (encounters per km walked). To determine spatial encounter rates in raster calculator, we divided the number of encounters per species or human activity by km walked in that same window.

Additionally, to eliminate potentially redundant or spatially-autocorrelated records, we presented the results a second way. Specifically, we converted point observations of each mammal or human activity sign to raster, then condensed all observations per species and sign type within a single 30-m pixel to a single occurrence for that species or sign type. We conducted a neighborhood analysis, using 1-km-radius moving windows to count the number of occurrences (i.e., raster pixels containing ≥ 1 such observation) per window. Therefore, multiple encounters of the same activity or species within a single raster pixel were deemed 1 occurrence. We provided number of occurrences in 2018 for comparison to number of encounters.

Results

Reconnaissance Routes "Recces"

Effort (distance walked) in the 2018 surveys totaled 1815.6 km. Broken down by sweep, effort was 921.5 km in sweep 1 and 894.1 km in sweep 2. Maps of the spatial distribution of survey effort for the two sweeps of 2018 demonstrate thorough coverage, with only the far northern portion of SNR excluded from sweep 2 (Figure 2). While surveying for mountain gorilla fecal samples, specifically, this two-sweep effort was considerably higher (3x) than the one-sweep survey for 2006 (600 km; Guschanski *et al* 2009) and slightly higher (1.2x) than that for the gorilla portion of the survey in 2011 which included two sweeps (1562.26 km; IGCP unpub. data). Survey effort when recording select mammals and human activities was also approximately 3-times more than in 2006 but was 2.2-times more than in 2011 which limited survey of other mammal and human activity signs to one sweep (816.26 km; IGCP unpubl. data).

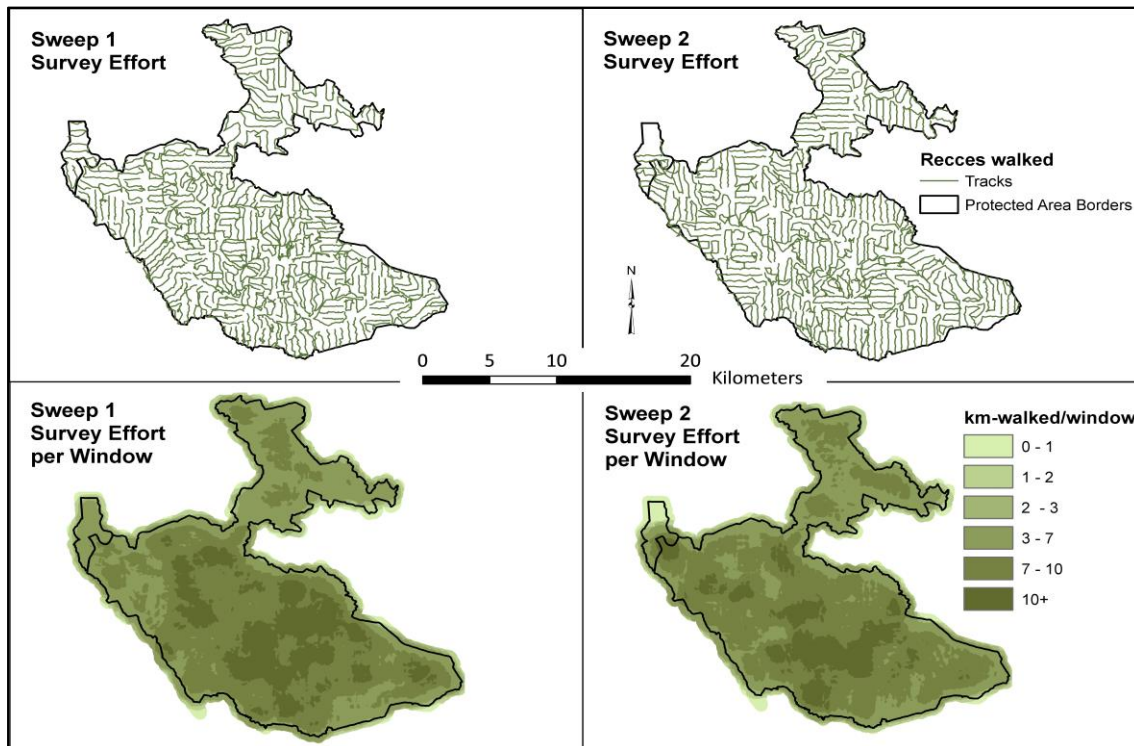


Figure 2. Survey effort walked (top panels) and distance-walked (km) per 1-km-radius moving window (bottom panels) in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 Surveys of Select Mammals and Human Activities

Mountain Gorillas

Spatial Analysis

We mapped the spatial distribution of occurrence rates (occurrences/km) of the following types of mountain gorilla signs: sightings, calls, dung, tracks, and nest sites (Figure 3). Compared to the other species mapped, mountain gorilla signs were fairly uniformly distributed within Bwindi-Sarambwe except for noticeable absence from the far eastern and far northern portions and higher concentrations in the middle and southern portions of the ecosystem. Surveys within SNR revealed signs of gorillas, but no recent or fresh nest sites. The area north of the ‘neck’ also exhibited few gorilla signs, yet we did detect some fresh nest sites there in addition to signs. As reported historically, the gorilla signs of 2018 mostly concentrated in the interior. Specifically, in sweep 1, the

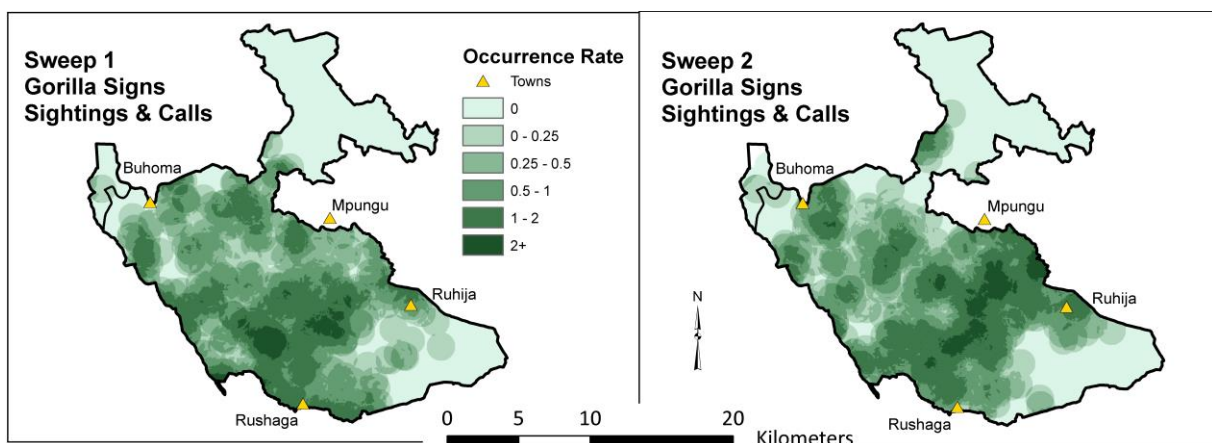


Figure 3. Spatial distribution of mountain gorillas in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 mammal surveys.

areas both northeast and northwest of Rushaga harbored the highest occurrence rates of gorilla signs, whereas in sweep 2, areas west and southwest of Ruhija harbored the most.

Field Sampling and Genetic Data Analysis

A total of 1884 fecal samples was collected during the training (n = 95), and during two sweeps: 814 in sweep 1 (9 March to 10 May 2018) and 975 in sweep 2 (5 October to 30 November 2018). Most fecal samples (82.7%) were conservatively assigned in the field as putatively unmonitored (n = 1481; Table 4). However, as detailed below, genetic analyses and comparison with known group locations later resulted in the fraction of samples attributed to unmonitored individuals being reduced to 57.5%, with a total of 42.5% determined to be from monitored gorillas; this large adjustment was expected and is similar to that observed in previous surveys.

Table 4. Summary of preliminary group membership assessments made in the field of fecal samples collected in two sweeps of the 2018 Bwindi-Sarambwe field survey. Conservatively, only those samples and nest sites that were definitely assigned a monitored group in the field were considered Putative Monitored here, but subsequent analyses of known group locations and genetic identities resulted in many samples within the Putative Unmonitored category here being corrected and switched to the Monitored category in the final analysis (see Table 5). We extracted all fecal samples (n = 1884) and considered them Failed extracts if they failed to amplify ≥ 10 microsatellite loci.

	Putative Monitored	Putative Unmonitored	Training ^a	Total
# Samples collected	308	1481	95	1884
# Failed extracts	55	255	26	336
# Fully genotyped samples	253	1226	69	1548

^aTraining samples were collected prior to the main sweeps during field-crew training and represent two monitored groups (Mukiza and Bitukura)

Gorilla Group Composition and Minimum Number Detected

We assigned the 1548 successfully genotyped samples to 451 individuals (192 males, 259 females), reflecting 50 groups (17 monitored, 33 unmonitored) and 13 solitary (unmonitored) individuals (12 male, 1 female; Table 5). Excluding training samples, the average number of genotyped samples per individual was very similar between monitored (average = 3.22 fecal extracts/individual) and unmonitored (3.25 fecal extracts/individual) gorillas. Where field teams assigned a known monitored group name to an initial nest site for a particular group (n = 11), we assumed that the group name was correct and genetics confirmed that subsequent nest sites were correctly assigned to the appropriate group. The genetic samples known to be from monitored groups (n = 633) included 175 of the 196 known monitored individuals in the 17 groups (Tables 5, 6). Additionally, 60 genetic samples identified 13 individuals that were associated with monitored groups in one of the sweeps but not in the other. Thus, in total, genetic samples included somewhere between 175 and 188 of the 196 monitored individuals.

Table 5. Summary of total number of genotypes, unique individuals, and groups in 4 mutually exclusive and exhaustive categories as determined genetically in final analyses of fecal samples collected during both sweeps of the 2018 Bwindi-Sarambwe field survey. Genotypes found in both monitored and unmonitored groups during the survey were unclassifiable.

	Monitored ^b	Unmonitored group	Unmonitored solitary	Unclassifiable	Total
# Genotyped samples ^a	633	832	23	60	1548
# Unique individuals found	175	250	13	13	451
# Groups	17	33	--	--	50

^aGenotyped samples are individual fecal samples with successful genotypes, not individual gorillas. These include multiple fecal samples for many individuals

^bMonitored groups also include samples collected during field training from nest sites of two monitored groups

Table 6. Group size and total abundance (Number of gorillas) in December 2018 as determined by daily monitoring, and field identification (ID) during the survey of fully habituated mountain gorillas residing in Bwindi Impenetrable National Park during two sweeps in 2018.

Social Unit	Number of gorillas	Field ID Sweep 1	Field ID Sweep 2
Bikingi	13	Bikingi	Not found
Bitukura	11	Bitukura	Bitukura
Bushaho	11	Bushaho	Bushaho
Busingye	12	Busingye	U2, W2
Bweza	11	J2	K2
Christmass	6	Christmass	Christmass
Habinyanja	15	Not found	Habinyanja
Kahungye	22	P1, P3	K4
Katwe	7	GG2	GG4
Kyaguliro	8	Kyaguliro	I1
Mishaya	9	K2	Mishaya
Mubare	5	FF4	GG6
Mukiza	13	I1	G1
Nkuringo	14	Y1	Nkuringo
Nshongi	7	K3	Nshongi
Oruzogo	17	Not found	O1
Rushegura	15	Rushegura	GG2
Total Monitored	196		

The remaining 855 genotypes corresponded to 263 unique gorillas in 33 unmonitored groups (n = 250 gorillas) and 13 solitary gorillas. Genotyped samples of unmonitored groups of gorillas contained 2 to 22 individuals (Table 7).

Table 7. Details of the social units of unmonitored mountain gorillas genetically detected in Bwindi Impenetrable National Park during sweeps 1 and 2 combined in 2018 (SU^a, unmonitored group; SOL, solitary individuals). For gorillas that were detected in multiple unmonitored groups across the survey, we categorized them as ‘unclassifiable to group but unmonitored’ (n=23).

Social Unit	Number of gorillas	Number of males	Number of females	Field ID Sweep 1	Field ID Sweep 2
SU-1	2	2	0	AA1	Not found
SU-2	17	3	14	BB1	GG1
SU-3	2	2	0	CC2	R2
SU-4	7	1	6	CC4	CC1
SU-5	11	3	8	Not found	DD1
SU-6	3	1	2	EE1	Not found
SU-7	2	2	0	EE5	Y2
SU-8	7	4	3	F2	Not found
SU-9	4	3	1	G1, N2	N3
SU-10	5	4	1	Not found	GG3
SU-11	8	1	7	Not found	H2, L3, M5
SU-12	17	4	13	I2, N4	I4, N2
SU-13	3	1	2	I3, G3	G2
SU-14	2	1	1	L1, L4	Not found
SU-15	4	1	3	Not found	M1
SU-16	8	4	4	M2, M4	Not found
SU-17	12	3	9	Not found	M3, N3
SU-18	2	1	1	Not found	N1
SU-19	5	1	4	N6	G3, I2
SU-20	2	1	1	N8	Not found
SU-21	2	1	1	Not found	P1
SU-22 ^b	1	1	0	P2, W2	Not found
SU-23	2	1	1	Not found	Q2
SU-24	22	8	14	Q2	L2, L6, L4
SU-25	20	9	11	R1, R2	R4, R1, S2, S1, S3
SU-26	2	1	1	T2	Not found
SU-27	17	9	8	V1, X2	V5, X1
SU-28 ^c	1	1	0	W6	Not found
SU-29	6	1	5	Z2	Z2
SU-30	9	5	4	K1	K3, P2
SU-31	7	4	3	L2, L3, L7, Q1, Q3	Not found
SU-32	12	6	6	Not found	V2
SU-33 ^d	3	1	2	I1	G1
Unclassifiable to group but unmonitored ^e	23	14	9	Various	Various
SOL	13	12	1	Various	Various
Total Genetic Count of Unmonitored	263	117	146		

^aUnmonitored groups were called GR in 2011 survey, but the group identities (numbers) from 2011 do not correspond to those in 2018. Therefore, unmonitored groups were termed SU (social unit) in 2018 to prevent erroneous time-series interpretation between the two surveys (e.g. GR-1 of 2011 does not equate to SU-1 of 2018).

^bSU-22 was named as a group rather than a solitary individual because four individuals were in SU-22 in sweep 1, but the group split in sweep 2 with 3 individuals joining other groups (2 of these individuals are accounted for as unclassifiable to group but unmonitored and the 4th was one of the 13 individuals that was unclassifiable due to associations with both unmonitored and monitored groups).

^cSU-28 was named as a group rather than a solitary individual because this and one other individual were identified nesting at the same site in sweep 1 and the other individual was found nesting by itself in sweep 2 (and therefore the second individual was categorized as unmonitored but unclassifiable)

^dThese 3 were found in an intergroup encounter with Mukiza on 16-March-2018.

^eIndividual gorillas that were detected in two groups across sweeps, and both groups were unmonitored.

Most solitary gorillas were male (12 males, 1 female; Tables 7, 8) and were detected at a single nest site. Field crews identified seven of the solitary individuals as silverbacks, four as unknown because the dung was smashed, and two as adults. Individual names were not available to confirm the identity of each solitary individual, but six of the solitary individuals were matched to 2011 genotypes. Based on the 33 unmonitored groups (Table 7), the mean minimum group size was 6.9 (SD 6.0) gorillas. The comparable estimate of group size of monitored groups based solely on the genetic sampled individuals was 9.7 (SD = 3.4), whereas the average known group size of the monitored groups was 11.5 (SD = 4.4). This difference reflects the fact that not all individuals known to be in the monitored groups were successfully genotyped. The spatial distribution of gorilla groups and solitary individuals combined for sweeps 1 and 2 demonstrate that some groups range outside the protected areas (Figure 4), and in fact, occasionally range outside of the Nkuringo Buffer Zone, which is located along the southwestern edge of BINP.

Table 8. Solitary gorillas found during two sweeps in Bwindi-Sarambwe 2018 surveys. In total, 13 solitary individuals (SOL-1-13) were detected across two sweeps (12 males, 1 female).

Individual	Sex	Field ID Sweep 1	Field ID Sweep 2
SOL-1	Male	BB2	Not found
SOL-2	Male	Not found	CC2
SOL-3	Male	E2	Not found
SOL-4	Male	EE3	Not found
SOL-5	Female	Not found	G6
SOL-6	Male	Not found	I3
SOL-7	Male	I4, N1	M2, M4
SOL-8	Male	J1	Not found
SOL-9	Male	Not found	M7
SOL-10	Male	P5	Not found
SOL-11	Male	Q4	Not found
SOL-12	Male	Not found	V3
SOL-13	Male	Not found	X2

Averaged Locations of Detected Mountain Gorilla Groups and Solitaries In Bwindi-Sarambwe 2018 Survey

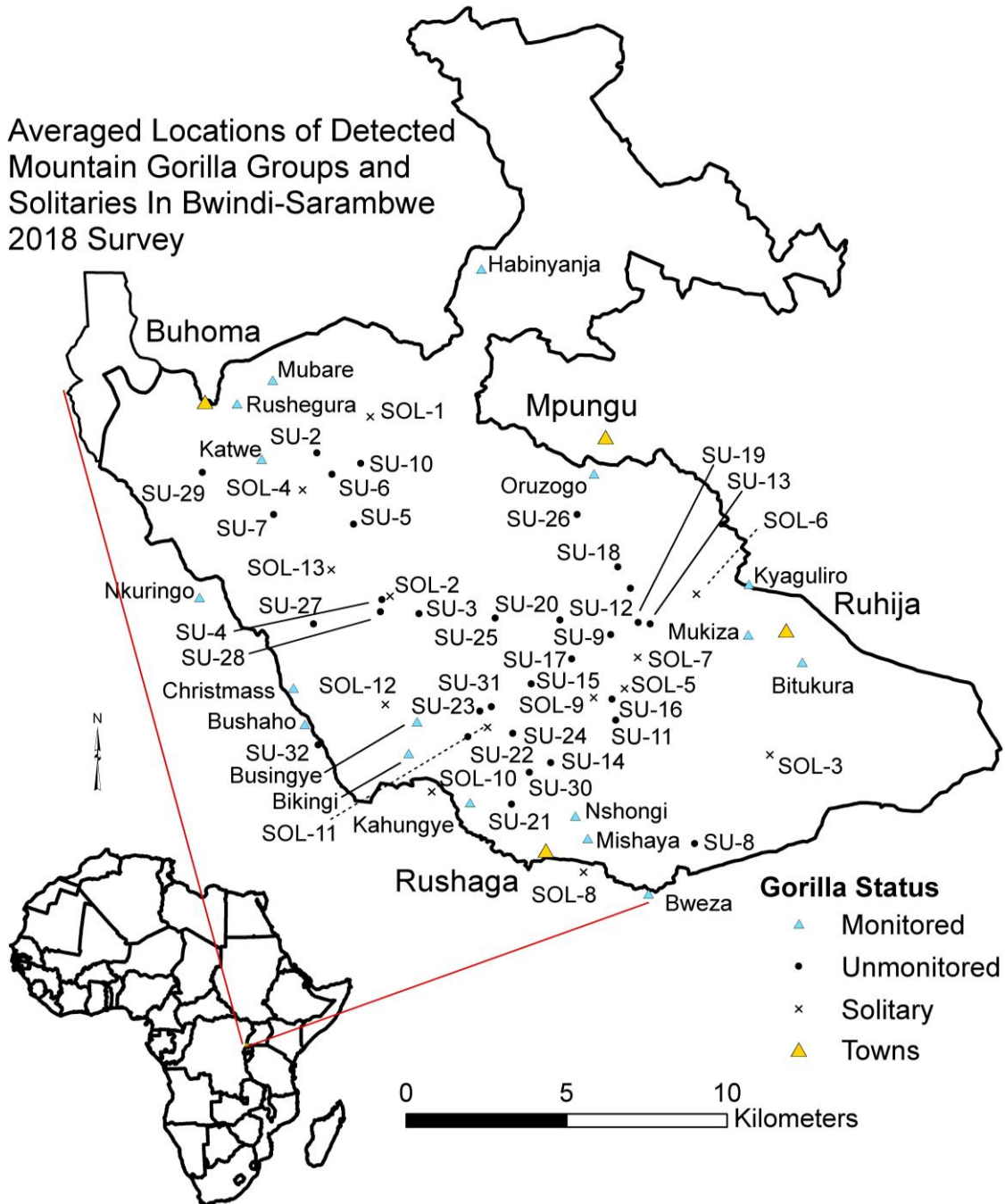


Figure 4. Averaged locations of nest sites per group or solitary gorilla detected in the Bwindi-Sarambwe 2018 survey. See Tables 6, 7, & 8 for additional details regarding each gorilla group or solitary individual

Parasites

We analyzed 329 gorilla fecal samples for intestinal helminth parasites from the first sweep, of which 206 were from field monitored gorillas and 123 from field unmonitored gorillas and graphed infection rates (Figure 5). Preliminary analyses suggest that the genus, *Ascaris*, which includes species known to infect humans, had a higher infection rate in monitored than in unmonitored gorillas. In addition, another parasite genus with species known to infect humans, *Trichuris spp.*, was only found in monitored gorillas. The third parasite, related to livestock, *Moniezia spp.*, had a higher infection rate in monitored gorilla groups than in unmonitored. *Strongyles*, which cannot be differentiated without PCR techniques or larval culture, were found at similar infection rates in both

monitored and unmonitored gorilla groups together with *Anoplocephala gorillae*, a commensal tape worm specific to gorillas. Although results are preliminary and infection rates are all relatively low, these data suggest that monitored gorillas may be infected with human and livestock parasites more than unmonitored gorillas.

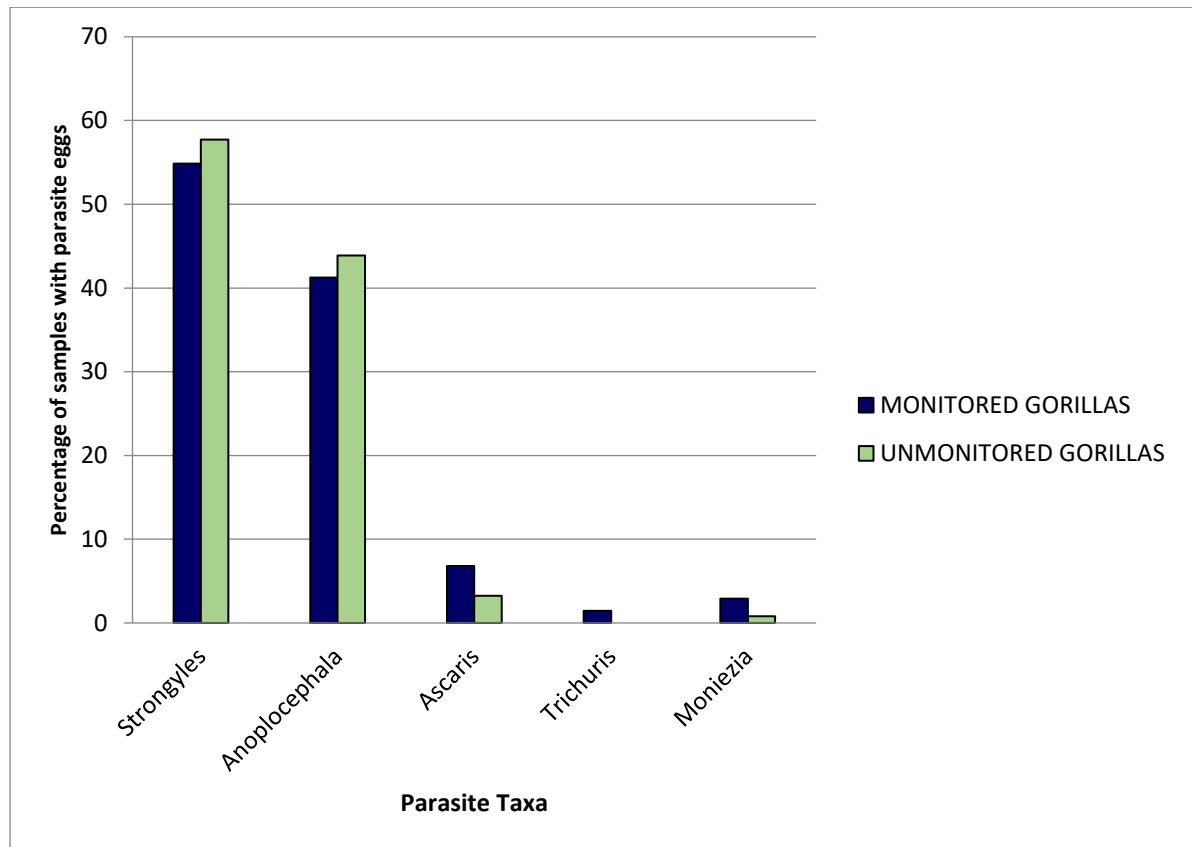


Figure 5. Preliminary results of percentage of mountain gorilla fecal samples containing parasitic helminth eggs of *Strongyles*, *Anoplocephala*, *Ascaris*, *Trichuris*, and *Moniezia* species collected during Sweep 1 of Bwindi-Sarambwe 2018

Further parasite analyses are underway at the Institute of Vertebrate Biology in the Czech Academy of Sciences (Brno, Czech Republic): results are pending, and will be preliminarily available in 2020.

Select Mammals

We mapped the spatial distribution of occurrence rates (occurrences/km) for the following types of mammal signs: sightings, calls, fresh or recent dung, and tracks of elephants (Figure 6); all dung, scrapings (digging), and sightings of carnivores (Figure 6); all dung, sightings, calls, nests, and tracks of chimpanzees (Figure 7); sightings and calls of baboons, black-and-white colobus, blue monkeys, L’Hoest’s monkeys, and red-tailed monkeys (Figures 6-8); and sightings and carcasses of black-fronted and yellow-backed duikers (Figure 8). We detected too few occurrences of bushbucks, bushpigs, and sitatunga to conduct meaningful moving-windows analyses, yet we provide a map of their locations (Figure 9).

Likewise, with other select mammal species, distributions occasionally differed between sweeps 1 and 2 of 2018. For example, in sweep 1 elephants primarily concentrated in the southeastern areas, whereas in sweep 2 we detected them distributed more evenly from the southeast and northward through the central portions of BINP. Data from the current survey suggest that elephants rarely use the region north of the ‘neck’, nor do they use SNR or the area southeast of SNR. In both sweeps, we

found few signs of elephants in the far north (Figure 6). When compared to the 2011 inset map, we detected a much broader distribution of elephants in 2018 where detections expanded northward, eastward, and westward. As in 2011, carnivore signs were rarely detected in Bwindi-Sarambwe, and overall, we detected more carnivore signs in the southeast. In sweep 2, we detected more carnivore signs in the western and northern areas than in sweep 1 (Figure 6).

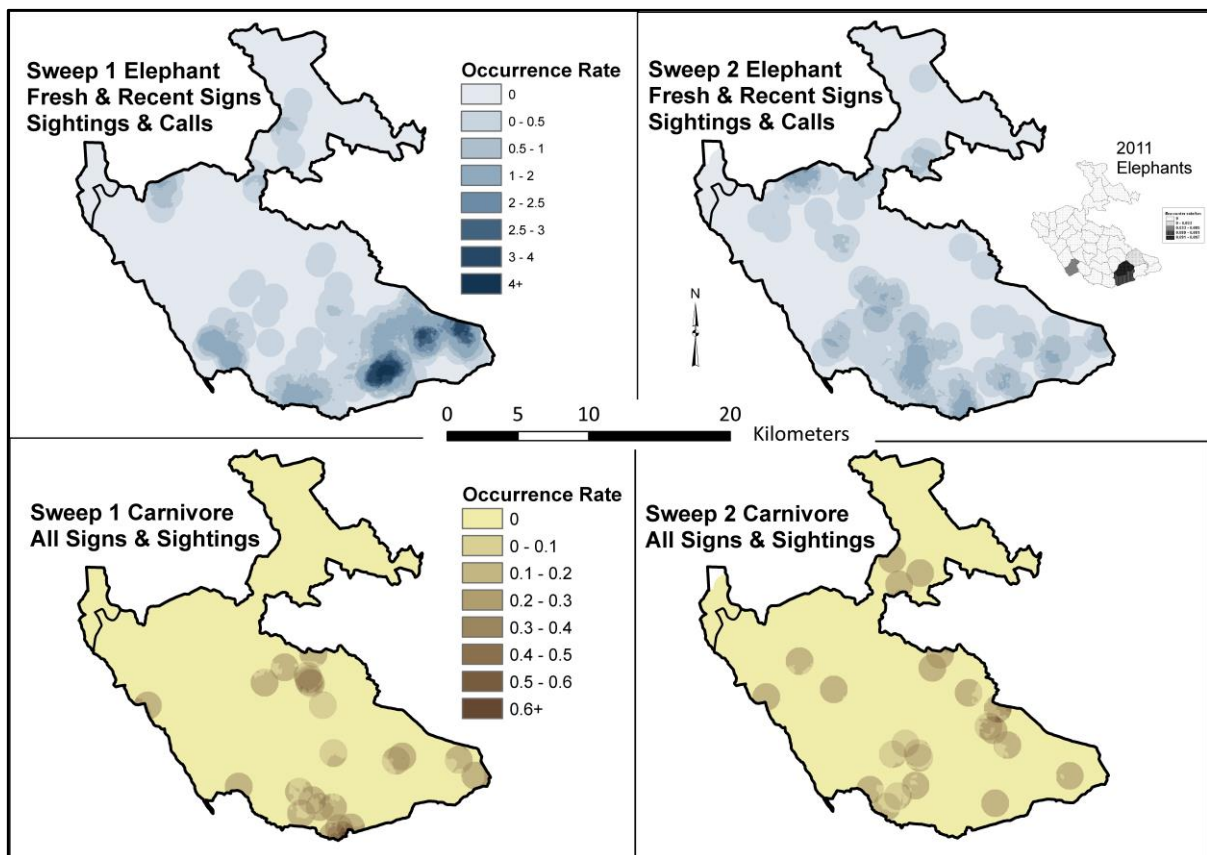


Figure 6. Spatial distribution of elephants and carnivores in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 mammal surveys. Inset map for elephants is from the 2011 survey conducted from September to November (IGCP unpub. data).

For chimpanzees in both sweeps, we detected a very wide distribution throughout Bwindi-Sarambwe except for in the extreme far east, and in sweep 2 we detected a higher concentration in the far north compared to sweep 1 (Figure 7). In both sweeps, we tended to find baboons near park edges, particularly in the area north of the 'neck'. We detected no baboons in the interior of the ecosystem (Figure 7).

Blue monkeys were well distributed throughout the ecosystem, with somewhat fewer detections in the west (Figure 8). In sweep 2, blue monkey detections concentrated more to the east, particularly around Ruhija, compared to sweep 1. Red-tailed monkeys exhibited similar distributions in sweeps 1 and 2, with most detections north of the 'neck' and in the west (Figure 8).

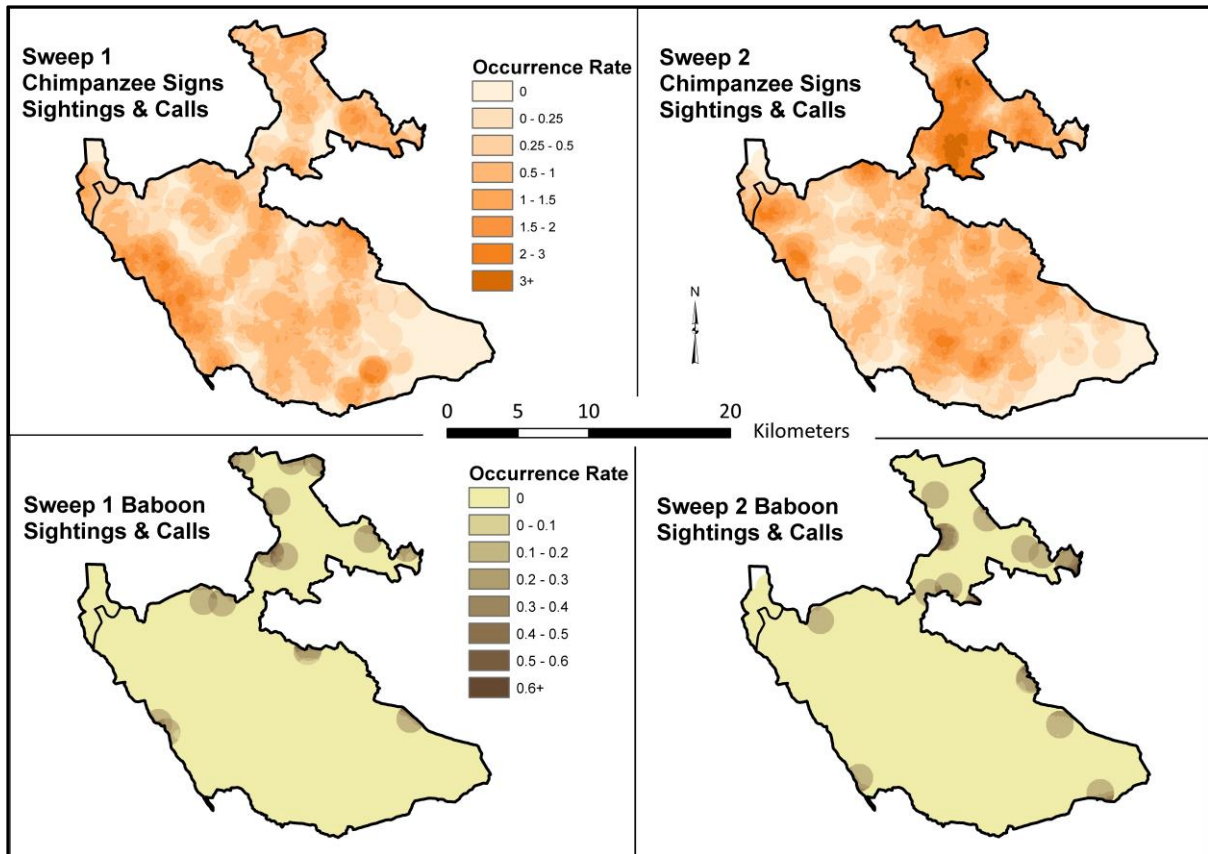


Figure 7. Spatial distribution of chimpanzees and baboons in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 mammal surveys.

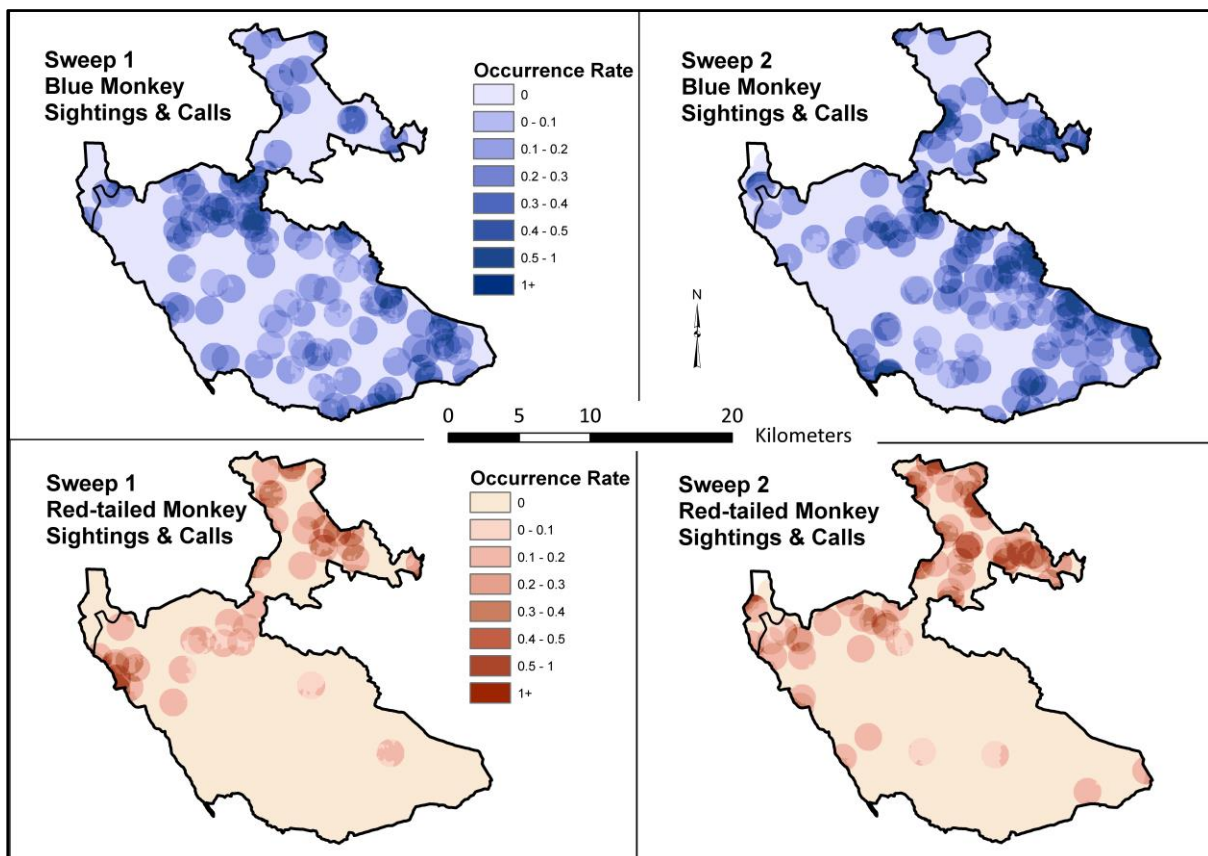


Figure 8. Spatial distribution of blue monkey and red-tailed monkey sightings and vocalizations recorded in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 mammal surveys.

We detected black-and-white colobus in roughly similar distributions for both sweeps, with most detections near park edges (Figure 9), yet black-and-white colobus occurred more frequently in the interior than did baboons (Figures 7 & 9). In both sweeps, we found l’Hoests monkeys near park edges south of the ‘neck’, with a few occurrences in the interior during sweep 2 (Figure 9). L’Hoests monkeys were occasionally detected in the ‘neck’ but not found north of the ‘neck’.

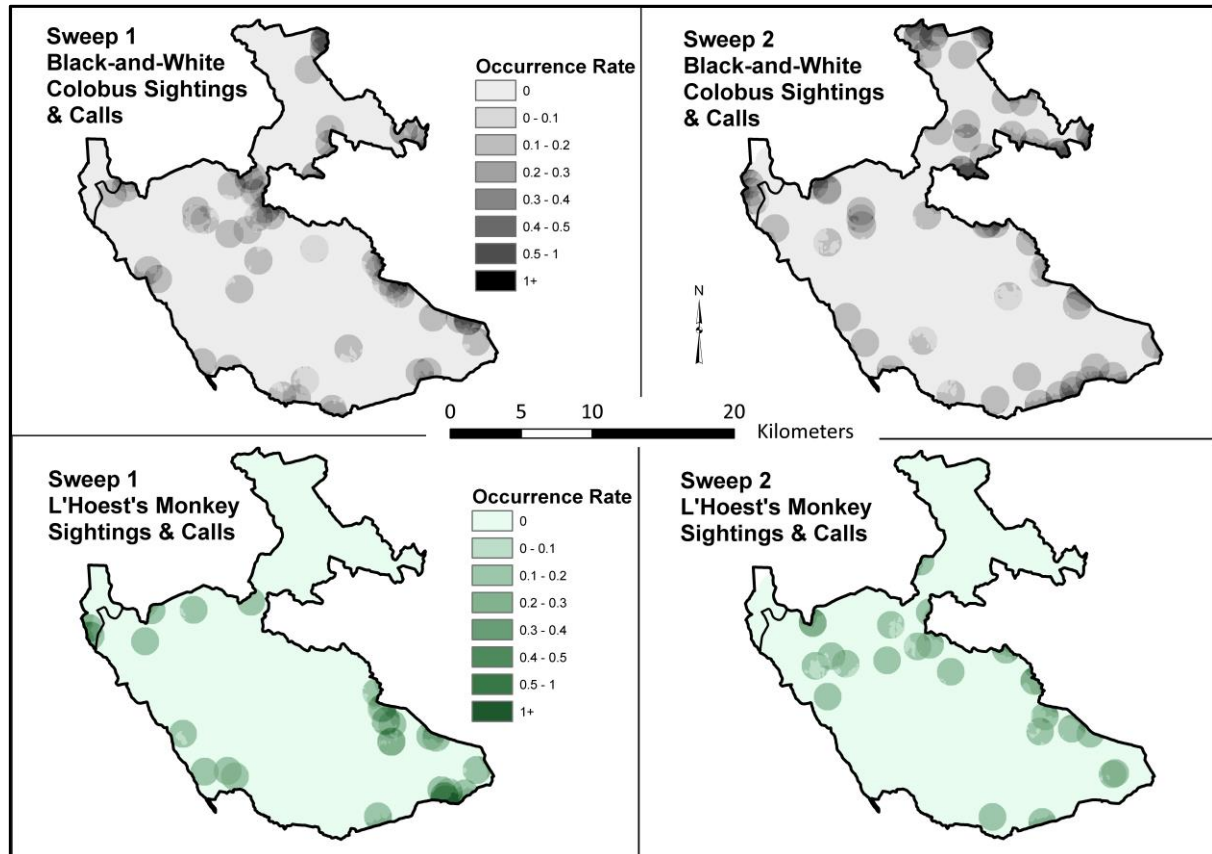


Figure 9. Spatial distribution of black-and-white colobus and l’Hoest’s monkey sightings and vocalizations recorded in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 mammal surveys.

We detected black-fronted duikers more frequently in the east and less frequently in the west during sweep 1 than sweep 2 (Figure 10). Black-fronted duikers appeared to avoid the ‘neck’ as well as the extreme southeastern corner of the ecosystem and rarely were found north of the ‘neck’. We primarily detected yellow-backed duikers throughout the interior south of the ‘neck’, with somewhat more frequent detections in the east during sweep 2 compared to sweep 1 (Figure 10). We found carcasses caused by poaching, as well as natural deaths, of both duiker species located near the park edge and in the interior (Figure 10).

Bushbucks and bushpigs were lightly distributed in the interior, south of the ‘neck’ with somewhat more detections of bushpigs in the south during sweep 2 compared to sweep 1 (Figure 11). Sitatunga and other duikers were not recorded during sweep 1. In sweep 2, we detected sitatunga in the eastern portion of BINP within the interior by Mubwindi swamp, which conforms to UWA ranger-based monitoring (RBM) reports (P. Ezuma, pers. comm., 7-Nov-2019). Other duikers were recorded in the interior south of the ‘neck’ (Figure 11).

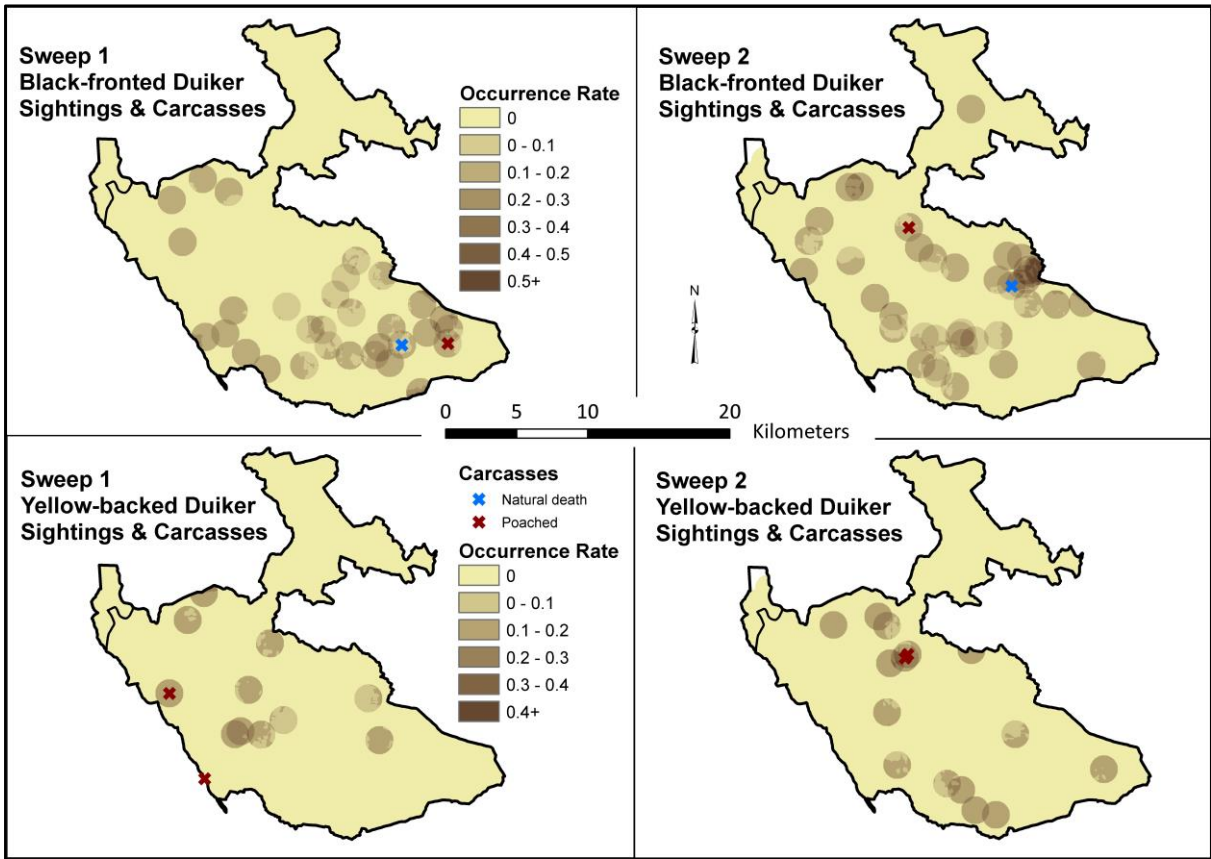


Figure 10. Spatial distribution of black-fronted and yellow-backed duiker sightings & carcasses recorded in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 surveys.

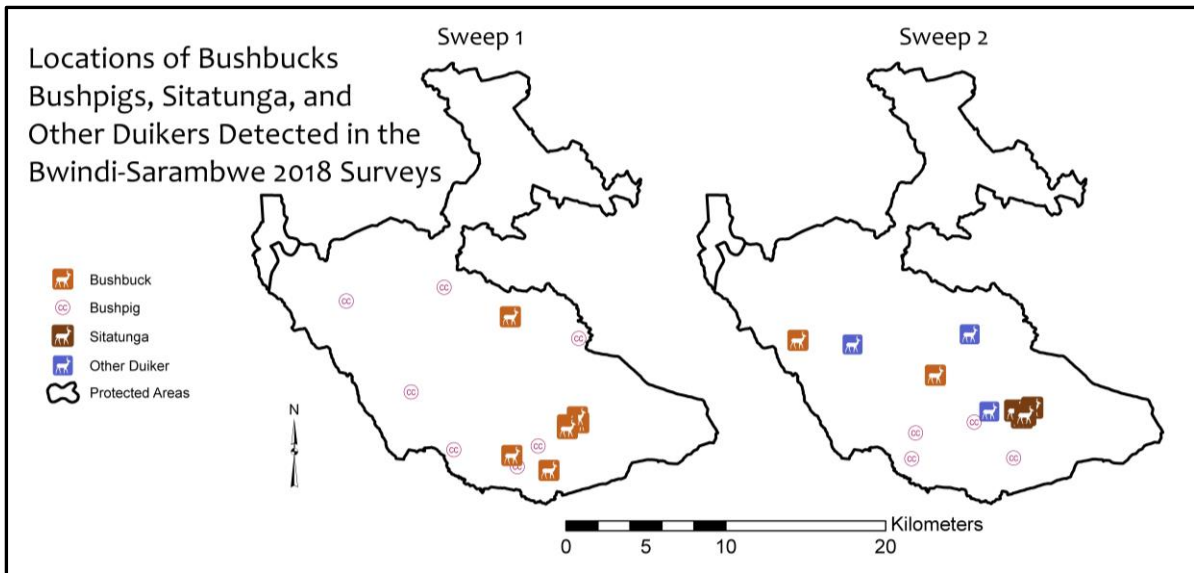


Figure 11. Locations of bushbucks, bushpigs, sitatunga, & other duikers recorded in sweeps 1 & 2 of the Bwindi-Sarambwe 2018 mammal surveys

With the exception of elephants (Figure 6), we did not compare the 2018 spatial distributions of select mammals to those of the 2011 survey, because 2011 maps were not provided (Robbins *et al* 2012). However, for a numeric comparison to the 2011 survey, we summarized the types of mammal observations that were also reported for 2011 (Table 9). These tabular data demonstrate that elephant encounter rates were somewhat higher in 2018 than in 2011 (Table 9). Consistent with past surveys, elephants were rarely actually seen; however, we recorded much more elephant dung in

this survey than in previous efforts, even after condensing encounters within 30 m of each other into single occurrences (Table 9). Similarly, we recorded substantially more detections of chimpanzees (both direct observations and signs) in 2018 than in 2011. In fact, compared to 2011, encounter rates were substantially higher in 2018 for the following species: baboons, black-and-white colobus, blue monkeys, l’Hoest’s monkeys, red-tailed monkeys, and yellow-backed duikers (Table 9). In contrast, 2011 and 2018 encounter rates were roughly similar for black-fronted duikers, bushbucks, and bushpigs (Table 9). To focus the survey effort on the primary objective – gorilla detection – and help teams move more efficiently through the forest, the 2018 protocols did not record dung of bushpigs, nor several types of human activities (e.g. human tracks or paths and collection of bark, honey, or water). By reducing the reporting burden (number and type of signs recorded), observers may have more thoroughly documented those observations of prime interest or the results may reflect true increases, there is no way to verify which.

Table 9. Total number of encounters, occurrences, and encounter rates (encounters/km-walked) of other select mammals in the 2011 and 2018 surveys. Because animals may occur in groups, encounter rates do not reflect total counts of individuals seen [displayed in brackets], but rather, 1 or more individual per encounter. Observations are animals seen or vocalizations heard, unless otherwise noted.

Large mammal observation / sign	2011 (2nd Sweep only)		2018 (Sweep 1)			2018 (Sweep 2)		
	Total encounters [individuals]	Encounter rate per km walked	Total encounters [individuals]	Total number of occurrences	Encounter rate per km walked	Total number of encounters	Total number of occurrences	Encounter rate per km walked
Baboon	3 [19]	0.004	15 [114]	15	0.016	17 [55]	16 [†]	0.019
Black & White colobus	20 [75]	0.025	50 [194]	50	0.054	60 [237]	59 [†]	0.067
Black-fronted duiker	26 [28]	0.032	31* [32]	31*	0.034	41* [47]	41*	0.046
Blue monkey	47 [193]	0.058	113 [261]	113	0.123	134 [310]	134	0.150
Bushbuck	4 [4]	0.005	7 [10]	7	0.008	2 [2]	2	0.002
Bushpig (2018 seen only)	5 [11]	0.006	7* [13]	7*	0.008	4* [6]	4*	0.004
Carnivore dung	9	0.011	24	23 [†]	0.026	22	19 [†]	0.025
Chimpanzee (2018 seen/heard)	4 [18]	0.005	25 [25]	25	0.027	33 [40]	32 [†]	0.037
Chimpanzee nest (2018 nest sites only)	235 [826]	0.288	427	418 [†]	0.463	607	599 [†]	0.679
Elephant (2018 seen/heard)	6 [51]	0.007	18 [46]	18	0.020	16* [34]	16*	0.018
Elephant dung	423 [1555]	0.518	567	560 [†]	0.615	648	637 [†]	0.725
Jackal	-	-	0	0	0	1 [1]	1	0.001
L’Hoest monkey	8	0.010	30 [89]	28 [†]	0.033	25* [66]	25*	0.028
Other duiker	-	-	-	-	-	3	3	0.004
Red-tail monkey	26 [151]	0.032	39 [276]	39	0.042	62 [294]	62	0.069
Sitatunga	-	-	-	-	-	4* [7]	4*	0.003
Yellow-backed duiker	3 [3]	0.004	12* [13]	12*	0.013	15* [16]	15*	0.017

*2 or less of the encounters or occurrences were carcasses

[†]number of occurrences were fewer than number of encounters due to condensation of like encounters within 30m distance

Human Activities

The map of all detected human activities (Figure 12) demonstrates that poaching activity continues in Bwindi-Sarambwe and appears to be at similar levels compared to 2011 (Table 10), yet remains relatively low compared to nearby locations, such as the Virunga Massif (0.09 to 0.15 encounters/km, Hickey *et al* 2019). Illegal activities, particularly snares, were prevalent near park edges, as well as in the interior both north and south of the 'neck' (Figures 12 & 13). We detected very little illegal activity in the interior south and southwest of Ruhija, although we did find snares in the vicinity of Mpungu, closer to park edge. All signs deduced in the field to be dogs were near park edges, either in the far east or the extreme far north. We detected freshly cut full-sized trees both near park edges and in the interior, concentrated in the far east and the area north of the 'neck' with a few detections in SNR (Figure 12). Camps tended to be in the core interior of BINP, well south of the 'neck', with two exceptions: one camp was detected in the interior north of the 'neck' and another was detected in the interior east of SNR. We discovered evidence of pit sawing both north of the 'neck' and within SNR. Of the five locations with evidence of burned vegetation, all five were found during sweep 1 and appeared to have burned about 1 to 5 months previous to detection (Figure 12); three were associated with honey collection and two were of unknown cause. Signs of firewood or pole cutting were rare (Figure 12).

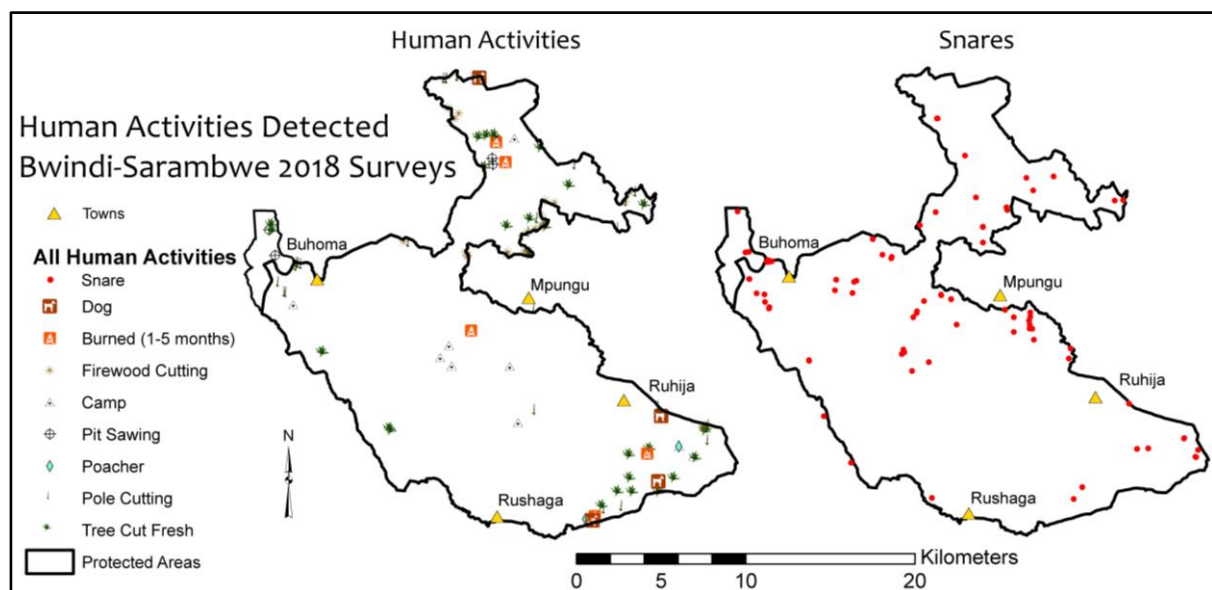


Figure 12. Spatial distribution of all human activities, with snares shown on the right, detected in both sweeps of the Bwindi-Sarambwe 2018 Surveys

We mapped occurrences of snares per km (including snares with animals still caught in them), as well as observations of all signs of human activity combined (poachers, snares, animals in snares, poached carcasses, camps) to reveal hotspots of poaching and human disturbance (Figure 13). We detected more snares and human activities in the far east and the southwest during sweep 1 than in sweep 2. In both sweeps, detections of snares and human activities demonstrated that the areas near Mpungu, the 'neck', north of the 'neck', and SNR all harbor some amount of human disturbance. Inset maps (Figure 13) broadly reflect the distribution of snares and human activities recorded in the second sweep of 2011. Comparing those 2011 insets to the 2018 distributions demonstrates that the vicinities north and south of the 'neck' continue to be problem areas for poaching. We detected snares in the far east during sweep 1 of 2018 but did not detect any there in sweep 2. Therefore, evidence supports the fact that some poaching continued since 2011 in that

area as well. Encouragingly, few illegal activities were recorded in the vicinity of Rushaga, an area where snares were historically found (Robbins *et al* 2012) and this change may indicate that conservation activities there have had positive effects. For comparison to the past survey, Table 10 summarizes those types of illegal activities that were reported in 2011 (Robbins *et al* 2012) and that levels of human activities and poaching remain similar to those of 2011.

Table 10. Total number of encounters and encounter rates (encounter/km-walked) of human activities in the 2nd sweep of 2011 and both sweeps of 2018. Note that 2018 was the first Bwindi-Sarambwe survey to incorporate two full sweeps of the study area that reported human activities. The encounter rate indicated for each sweep equals the number of encounters divided by km-walked for the respective sweep.

Human sign	2011 (2 nd Sweep only)		2018 (Sweep 1)		2018 (Sweep 2)	
	Total number of encounters	Encounter rate per km walked	Total number of encounters	Encounter rate per km walked	Total number of encounters	Encounter rate per km walked
Snares	47	0.058	39	0.042	49	0.055
Wood cutting	56	0.069	34	0.037	51	0.057
Camps	10	0.012	7	0.008	3	0.003
Poachers	1	0.001	1	0.001	1	0.001
Dogs	-	-	1	0.001	3	0.003
Fires	2	0.002	5	0.005	0	0.000

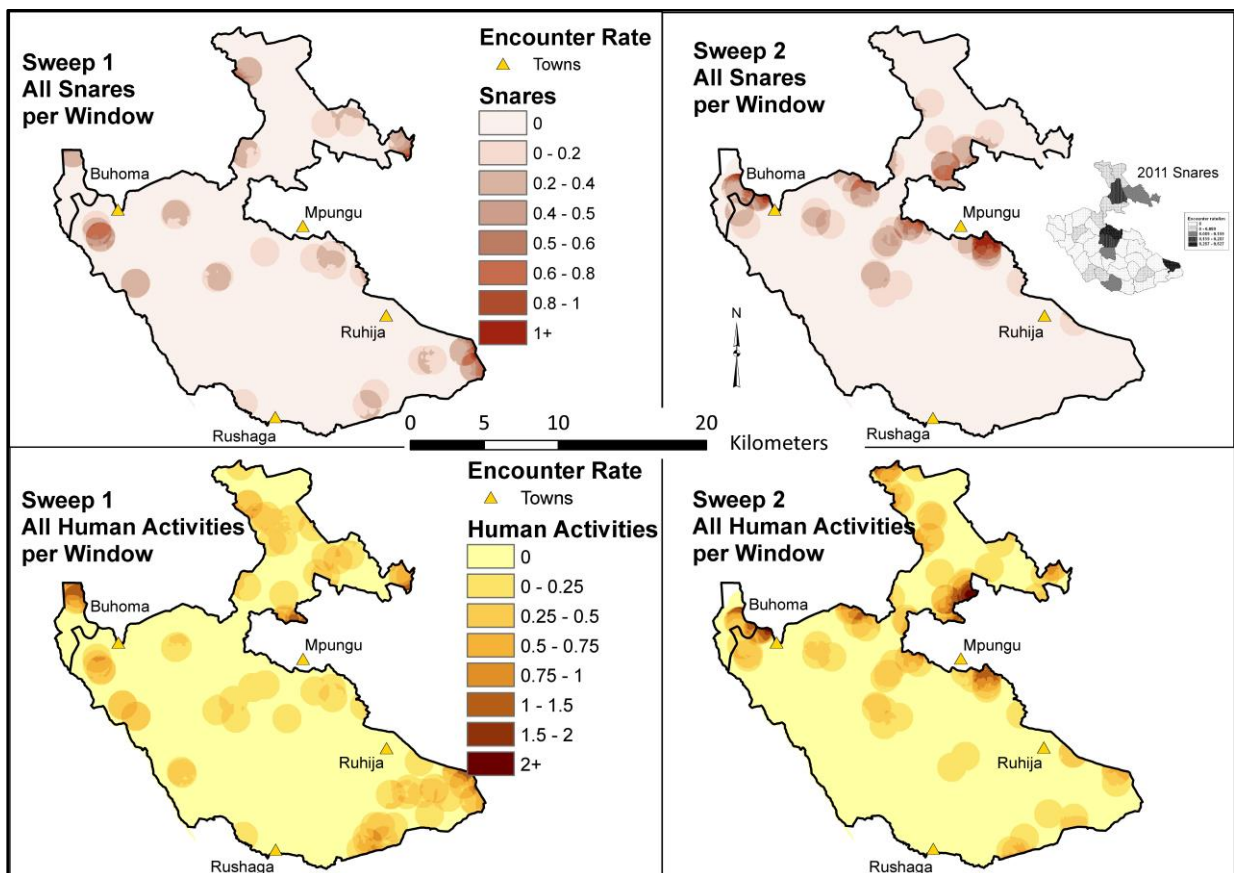


Figure 13. Occurrence rates of snares and all human activities per moving window (1-km radius) by sweep. Inset maps of snares and human activities are from the 2011 survey (Robbins *et al* 2012).

Discussion

Mountain Gorillas

We arrived at a total minimum count of 459 gorillas inhabiting the Bwindi-Sarambwe ecosystem as of December 2018 by adding the known number of gorillas ($n = 196$) from the monitored groups to the minimum count of unmonitored gorillas detected genetically ($n = 263$). Compared to the 2011 survey estimate of 400 individual gorillas (an estimate that included correction factors for 37 potentially undetected infants and/or individuals that failed to successfully genotype; Robbins *et al* 2013, Roy *et al* 2014), the 2018 minimum count of 459 gorillas (that included no correction factors and is a true minimum) confirms that the Bwindi-Sarambwe mountain gorilla population grew during the intervening period. Based solely on the number of unique consensus genotypes, a minimum of 451 individual mountain gorillas (192 males, 259 females) was detected genetically in the Bwindi-Sarambwe 2018 survey. The difference in the minimum count and the number detected genetically derives from the fact that not all monitored gorillas were sampled or successfully genotyped.

Importantly, a minimum count does not reflect the total number of mountain gorillas, as in any survey, some individuals and groups are not be detected, as evidenced by the fact that only 1 solitary individual ($n=13$) was detected in both sweeps, and 14 of 33 (42%) unmonitored groups were detected in both sweep 1 and sweep 2. The remaining individuals or groups were detected in only one of the two sweeps. Therefore, similar to previous such approaches (e.g. Roy *et al* 2014, Granjon *et al* in press), probabilities of detection will be calculated through mark-recapture analyses to estimate the total abundance of the population and a derived growth rate, which is anticipated in a manuscript in 2020.

Numbers of groups also appear to have increased between 2011 and 2018. The 459 gorillas documented in 2018 were found in 50 gorilla groups, including 17 monitored and 33 unmonitored groups, and 13 solitary individuals (12 male, 1 female). By comparison, there were 36 groups and 16 solitary individuals detected in 2011. Although the difference in numbers of groups estimated between 2011 and 2018 surveys likely reflects a real increase, the particular numbers of should be interpreted cautiously, as they represent only estimates subject to uncertainty. Whereas group identity and membership are known in the monitored portion of the population (and can be differentiated from temporary associations), assessments of group identity and membership associated with the unmonitored portion of the population represent only snapshots of associations, which could reflect any number of social dynamics not necessarily indicative of stable groups. Indeed, this dynamic situation was evident in the 23 individuals assigned to unmonitored groups but unclassifiable to a particular group (~9% of the unmonitored gorillas).

Parasite and Viral Analyses

Preliminary analyses suggested higher rates of infection from human- and livestock-associated intestinal worm taxa in fecal samples from monitored mountain gorillas than from unmonitored gorillas. Although further analyses will be done on potentially pathogenic parasites, a cautionary management approach would include greater efforts to minimize cross-species disease transmission. For example, timely and safe herding of gorillas back to the national park by trained personnel – such as the Human and Gorilla (HuGo) Conflict Resolution teams of community volunteers – when gorillas range onto community land. Longer-term measures such as encouraging community members to plant crops that are unpalatable to gorillas and installing physical barriers should also be established or reinforced.

Bwindi-Sarambwe mountain gorilla fecal samples were not screened for viruses by Gorilla Doctors as part of its implementation of the USAID Emerging Pandemic Threats PREDICT project, in part due to the necessity to prioritize testing of wildlife and human samples collected concurrently in space and time, but also because screening was not expected to generate a large quantity of positive test results from environmental fecal samples collected from presumably healthy gorillas. PREDICT's overall results in testing many tens of thousands of wildlife specimens from around the world have generally resulted in PCR-positive results for a very small fraction of samples (<1%) (PREDICT 2014).

Select Mammals

A chief motivation for monitoring species over the long-term is to ascertain if the species are still present and whether signs of their presence are decreasing, steady, or increasing (McNeilage *et al* 2001, 2006; Guschanski *et al* 2009; Gray *et al* 2013; Robbins *et al* 2012, Roy *et al* 2014, Hickey *et al* 2019). However, as several previous studies have highlighted (Gibbs 2000, Anderson 2001), counts of indirect signs such as tracks or dung are not reliable measures of abundance, particularly without robust estimates of dung-production and dung-decay rates (Barnes 2001, Laing *et al* 2003) or when effort varies between surveys. In addition to potential bias introduced from inconsistent survey effort, observer ability can also introduce bias, since some observers may be able to detect more signs than other observers (Fitzpatrick *et al* 2009). Therefore, we interpret the mammal survey results reported here as an indication of species occurrence, and that none of the species surveyed show a dramatic decline or absence. However, we do not try to discern trends by comparing encounter rates observed here to past surveys.

Regarding potential changes in spatial distributions over time, it is important to note that since animal movements are spatially and temporally dynamic, surveys such as these – which pass through any given area very quickly – can only provide approximate distributions because they essentially are snapshots in time rather than comprehensive accounts of mammal occurrence in a given year. Conceivably then, some species may have been found in other locations if the surveys had run continuously throughout the year. That said, presence-absence interpretations suggest that elephants were present only in the extreme southern and the southeastern portions of the ecosystem in 2011, whereas the 2018 data provide evidence of a much broader distribution of elephants that fanned out in all directions within BINP.

As mentioned previously, we caution that the protocols in the present survey involved a lower reporting burden than the 2011 surveys, therefore the distribution of signs of species such as elephant, and the tabular counts of signs, may appear increased simply due to changed protocols. Similarly, encounter rates of many mammal species revealed apparent increases since 2011, except for bushbucks, bush pigs, jackals, and sitatungas which were rarely detected in either 2011 or 2018 (yet a comparison of their relative detection rates provides no evidence of declines). Therefore, there is no evidence of declining populations in the select mammals surveyed in 2018; and yet, such speculation requires verification with separate species-specific surveys that produce confidence intervals around the abundance estimates.

In fact, if population abundance estimates are desired for species other than mountain gorillas, then future work will need to focus on either mark-recapture (of genotypes, unique markings, actual tagged animals, or vocalizations; Seber 1982, Barnes 2001, Marques *et al* 2013), or distance approaches (Plumptre 2000; Buckland *et al* 2005), depending on the species. All these approaches take considerably more field and laboratory time than simply recording dung observations and

would slow the process of the primary objective – to detect mountain gorilla nest sites and collect fecal samples with a sufficiently short time interval between sampling occasions (sweeps) to consider the subpopulation closed (negligible births or deaths). Although we recommend independent projects to ascertain population abundances of other species, we did initiate a pilot project in the 2nd sweep of 2018 where we collected samples of fresh (<24h) elephant dung for subsequent genetic analysis to individual. That pilot project may provide the initial ground work for future non-invasive genetic capture-mark-recapture abundance estimates of elephants within the Bwindi-Sarambwe ecosystem.

Human Activities

As in previous similar efforts, the thorough sweep approach of these surveys provided benefits beyond monitoring select mammals and allowed us to detect illegal activities in remote areas of the Bwindi-Sarambwe ecosystem that are rarely patrolled by law enforcement. Whereas monitoring of illegal activities through daily ranger-based-monitoring is opportunistic, the ecosystem-wide survey provides a more systematic and comprehensive picture of the occurrence of illegal activities. Furthermore, as with select mammal distributions, it is important to note that since human activities are spatially and temporally dynamic, surveys such as these – which pass through any given area very quickly – can only provide approximate distributions because they essentially are snapshots in time rather than complete accounts of occurrence of human activities throughout a given year. Conceivably then, some human activities might have been detected in other locations if the surveys had run continuously throughout the year.

That said, from a tabular perspective, snare encounter rates did not differ notably between 2011 and 2018, indicating little or no reduction in poaching activity despite considerable conservation efforts during that period. For example, UWA increased patrol staff by over 100 new recruits to increase the patrol efforts and introduced the Spatial Monitoring and Reporting Tool (SMART) to facilitate data collection during patrols. Moreover, two groups of poachers located in Nteko (n= 53) and Mpungu (n=13) renounced poaching in 2013 and 2016, respectively. In addition, in 2014 and 2015 UWA signed collaborative boundary management agreements with the local communities where locals harvested and sold mature boundary trees thereby earning 146,000,000 UGX. The communities then planted the length of the harvested boundary area with trees that they will harvest again once mature. This arrangement enhanced community sentiment toward the Protected Area because community members felt they were contributing to management (P. Ezuma, pers. comm. 1-Dec-2019). Further, tourism revenue sharing funds continued to be disbursed to local communities, so that the benefits of the Protected Area were shared with surrounding neighbors. The very few illegal activities recorded around Rushaga may indicate some positive conservation results from management activities in that particular area, where historically snares were found (Robbins *et al* 2012), whereas the region around Mpungu continues to harbor illegal activities as it has done historically. The presence of snares and other illegal activities documented throughout Bwindi-Sarambwe, both on the edges and in some interior regions, while fewer than in the nearby Virunga Massif, demonstrates that enhanced law enforcement and new techniques to detect and prevent illegal activities should be explored to further reduce poaching within the protected areas. Moreover, enhanced and normalized coordinated efforts between Uganda and DRC would further improve the security within SNR. The status quo is insufficient to stop bushmeat hunting and other illegal activities in Bwindi-Sarambwe.

Further, future socio-economic surveys of bushmeat consumption and extraction of timber resources among communities neighboring the ecosystem – and any links to more distant markets – could help elucidate some of the causal relationships behind patterns of illegal activities documented in this report. The conservation community would do well to increase incentives designed to further reduce the dependence of local peoples on park resources, including but not limited to more transparent and equitable access to benefits from conservation and tourism.

Future Work

Ultimately, in addition to the manuscript described above, which will result in a forthcoming mark-recapture abundance estimate of the population and a derived estimate of the growth rate, several end products will arise from this single collaborative effort. Vegetation-type data from this study are being combined with another broad-scale land-cover classification (WWF-Germany and IGCP 2017) and will inform a new vegetation-type map of the Bwindi-Virunga Landscape (BVL). As those land-cover classification products become available, they will inform numerous future studies related to habitat for local species, land-cover change, landscape planning, and population viability analyses. Likewise, location data of the select mammal species included in these surveys, in combination with the new land-cover data, will allow the development of species distribution models (SDMs), as well as explorations of ecological relationships among species occurrences and various vegetation types, distance from roads, distance from trails, occurrence rates of human activities, associations with other species occurrences, and abiotic factors such as elevation, precipitation, soil type, slope, and aspect, to name a few. IGCP plans to be forthcoming with such products (e.g. niche models) in 2020.

Conclusions

The 2018 surveys reported the largest count of mountain gorillas, as well as elephant and chimpanzee detections, ever recorded for the Bwindi-Sarambwe ecosystem. Evidence from this survey suggests substantial growth for this subpopulation of mountain gorillas since 2011. And although indirect signs are not definitive indicators of increasing trends, they do suggest that elephants and chimpanzees are not declining in this ecosystem. The results, even accounting for the increased effort in this most recent survey, represent a remarkable conservation achievement. While exercising caution due to the limitations of surveys of indirect signs which do not translate directly to abundances, there were no indications of population declines since 2011 for any of the mammal species surveyed, and encounter rates for most of the mammal species increased substantially since 2011.

Nonetheless, protected area authorities and conservation groups must remain engaged, as the Bwindi-Sarambwe ecosystem is still vulnerable to human disturbance due to factors such as its relatively small area, limited core interior, climate change, dependency of surrounding human community on park resources, and other human-wildlife conflicts. Additionally, except for the area around Rushaga, it appears that the density of snares in the Bwindi-Sarambwe ecosystem has not declined since 2011, suggesting that additional efforts need to be made to reduce poaching, because snares remain a notable threat to wildlife. Despite those human activities, the apparent increased spatial distribution of elephants and chimpanzees inhabiting Bwindi-Sarambwe validates, to some degree, the conservation policies and strategies in the region including intensive law enforcement, community conservation projects, and transboundary collaboration.

Recommendations

Based on the results and conclusions from this survey, the following recommendations are offered:

- 1) Continue to monitor and assess the population of mountain gorillas in Bwindi-Sarambwe and its associated implications on park management and prepare to adapt management to a dynamic and growing population of mountain gorillas.
- 2) Ensure best practice standards for tourism are in force and followed to mitigate risks of disease transmission to, and behavior change in, mountain gorillas.
- 3) Continue to monitor the health of habituated gorillas, conduct veterinary interventions to remove snares and treat life-threatening injuries and illness, and continue surveillance and research on diseases impacting the mountain gorilla population.
- 4) Continue to implement initiatives that improve the quality of life and conservation attitudes of surrounding human populations including conservation education, improved health services and increased alternative livelihood options which have potential to reduce human related threats to the mountain gorillas and other wildlife.
- 5) Conduct non-invasive mark-recapture or line-distance surveys if actual population abundances and trends are desired for other species.
- 6) Conduct socio-economic assessments of bushmeat hunting and consumption, as well as timber harvesting, among communities neighboring the Bwindi-Sarambwe ecosystem; develop effective interventions, with specific focus on Mpungu.
- 7) Re-establish transboundary law-enforcement monitoring and anti-poaching efforts, including regional meetings as well as joint and coordinated patrols in the Bwindi-Sarambwe ecosystem.
- 8) Invest further in ranger-based monitoring (RBM) and SMART database management: data collection, data management, and data sharing among Uganda and neighboring countries would improve the ease of use, interpretability, and understanding of dynamics in mammal populations throughout the Bwindi-Virunga Landscape.
- 9) Routinely collect fresh elephant fecal samples for genetic analysis to achieve more regular tracking of the population dynamics of elephants.
- 10) Evaluate acoustic monitoring to individual, as well as nest counts that use line-distance approaches, as alternatives to non-invasive genetic mark-recapture for estimating chimpanzee abundance and population monitoring, because consistent detection of fresh fecal samples is challenging for arboreal species such as chimpanzees.
- 11) Explore the potential responses of species distributions to roads, expansions or upgrades of the road network, and associated traffic and accessibility afforded by roads.

Acknowledgements

The 2018 Bwindi-Sarambwe population surveys of mountain gorillas, known as census, were conducted by the Protected Area Authorities in the Democratic Republic of Congo and Uganda (l'Institut Congolais pour la Conservation de la Nature [ICCN] and the Uganda Wildlife Authority [UWA]) under the transboundary framework of the Greater Virunga Transboundary Collaboration. The census was supported by the Rwanda Development Board, International Gorilla Conservation Programme (IGCP; a coalition of Conservation International, Fauna & Flora International and WWF), Mammalian Ecology and Conservation Unit (MECU) of the UC Davis Veterinary Genetics Laboratory, Max Planck Institute for Evolutionary Anthropology, The Dian Fossey Gorilla Fund, Institute of Tropical Forest Conservation (ITFC), Gorilla Doctors, Conservation Through Public Health,

Wildlife Conservation Society Uganda Country Office, WWF Uganda Country Office, and Bwindi Mgahinga Conservation Trust. The census was funded by Fauna & Flora International, WWF, and Partners in Conservation at the Columbus Zoo & Aquarium.

We would like to thank the immigration, customs, and security agencies in the three countries for their cooperation and assistance. Special thanks also to Pontious Ezuma, Christopher Masaba, Emmanuel de Merode, Désiré Sekibibi Bareke, Innocent Mburanumwe, Prosper Uwingeli, Simon Nampindo, Mike Cranfield, Ricky Okello, James Byamukama, Robert Bitariho, Dennis Babaasa, Anna Behm Masozera, Benjamin Mugabukomeye*, Altor Musema*, Henry Mutabaazi, Charles Kayijamahe*, Eustrate Uzabaho*, Richard Chota, Desi Tibamanya, Clemencia Akankwasa, Florence Tukamushaba, Gamariel Ndabananiye, Jerome Baguma, Jean Diogene Komezusenge, Jean Jacques Ngenzi, Wilbur Kaiire, Richard Ntegyereize, Kabale Aureliano, Gordon Barisigara, and Moses Ahimbisibwe for their cooperation and assistance.

This study built upon the following researchers' previous genetic analyses of mountain gorillas: Katarina Guschanski, Justin Roy, Anne-Celine Granjon and Linda Vigilant. We would also like to thank: the ITFC for the use of their facilities as a training base and for storage of equipment, supplies, and field samples; Gorilla Doctors, Conservation Through Public Health, and the University of Oregon for the provision of supplies and management of samples; and anyone else who participated in any way in this survey effort.

This effort would not have been possible without the arduous work and dedication of all the participants, through many long, wet days in steep, rugged terrain. We are extremely grateful to all the team leaders and team members: Akampulira Levios, Akantorana Moses*, Arinaitwe Joseph*, Asesa Lusambya Nicolas, Asiimwe Medard, Assu Juma, Atuzarirwe Wence, Bakebwa Ismael*, Barahebuza Lucky, Bararuha Ntuyenabo Aimé, Byarufu Katchiribindi Faustin, Byaruhanga Charles, Byaruhanga Christopher, Byomugabe Edmond, Chance Joseph, Chemayek Doreen, Gizamba Jacob, Hakuzimana Ernest, Hatangimana Ignace, Hodali Olivier, Irakoze Emmanuel, Kaberabose Prosper, Kahanga Saul, Kambale Chiyiro Jean Paul, Kandji Amisi Bernard, Katungi Gerald, Katutu Jacques, Kendelu Raymond Nestor, Kibakuli Yosia, Kirunda Ben, Kyamujara Winnie, Manizabayo Katoto David, Mbabazi Simon, Mboijana James, Mugiraneza Léonard, Muhereza Raymond, Muhindo Ngarwe Alain, Muhwezi Alex Amos, Mukamana Donathile, Mushimiyimana Straton, Musiimenta Moses, Mutale Julius, Muvunyi Emmanuel, Nahabwe Enos, Nahabwe Job, Ndashibu Dunia Philippe, Ngaruye Innocent, Ngiuwonsanga Alphonse, Nibishaka Faustin, Niyonzima Mathias, Nizeyimana Fidèle, Nkunzingoma Phocas, Nkurunziza Bichabire Timothée, Nkwasiabwe Aventino, Nsabimana Fred, Nsabimana Peter, Nsekanyarenze Christophe, Ntirumenyerwa Ezira, Owoyesigyire Narcisio, Ruranga Samson, Rwambibi Léonard, Safari Gabriel, Strutt Leo, Tibanyendera James, Tibenda Emmanuel, Tindimwebwa Vastine, Tinkasimire Rogers, Tugumisirize Alex, Tugumisirize Deus, Tukamuhabwa Damiano, Tumugabirwe Lawrence*, Tumwesigye Bernard, Tumwesigye Ignatius, Tumwiine Bruno, Turyamubona Clovis, Turyananuka Beda, Tuyishime Kagaba Clément, Twagirayezu Constantin, Twesigye Isaac, Twinomuhangi Anaclet, Uzabaho Eustrate*, Wanderu Mathe, Watuwa James, and Zirimwabagabo Jean Marie Vianney.

Additional thanks goes to: Akampurila Prudence, Arinaitwe Emmanuel, Atusasire Gad, Byamukama Innocent, Karuru Alex, Katushabe Psatel, Kyarisma Romina, Mbonimpa Didas, Mwesigire Dismus, Nturanabo Cypriano, Sigirenda Valentino, Sostine Wednesday, Tumwesigye Peter, Tushemereirwe Stephen, Ziryaharugo Silver, Zoreka Damazo, and all other chefs, camp keepers, and equipment cleaners, and to field-team instructors and scientists not listed elsewhere: Jena Hickey and Winnie Eckardt, all other instructors are marked above with an asterisk (*).

Literature Cited

- Anderson, D.R. 2001. The need to get the basics right in wildlife field studies. *Wildlife Society Bulletin*. 29:1294-1297. URL: <http://www.jstor.org/stable/3784156>. Accessed 15-Dec-2017
- Barnes, R.E.W. 2001. How reliable are dung counts for estimating elephant numbers? *African Journal of Ecology* 39:1-9
- Boffa, J-M., Turyomurugyendo, L., Barnekow-Lilleso, J-P., and Kindt, R. 2005. Enhancing farm tree diversity as a means of conserving landscape-based biodiversity. *Mountain research and development: Volume 25, Issue 3*
- Broquet, T. and Petit, E., 2004. Quantifying genotyping errors in noninvasive population genetics. *Molecular Ecology*, 13(11), pp.3601-3608
- Buckland, S.T., Anderson, D.R., Burnham, K.P., and Laake, J.L., 2005. *Distance sampling*. John Wiley & Sons, Ltd.
- Butynski and Kalina 1993. Three new mountain national parks for Uganda. *Oryx*. 27(4):214-224
- Fitzpatrick, M.C., Preisser, E.L., Ellison, A.M., and Elkinton, J.S. 2009. Observer bias and the detection of low density populations. *Ecological Applications*, 19(7): 1673-1679
- Gibbs, J.P. 2000. Monitoring Populations. In Boitani, L. and Fuller T.K (Eds.) *Research Techniques in Animal Ecology: Controversies and consequences*. Columbia University Press, New York. p213-250
- Granjon, A.C., Robbins, M.M., Arinaitwe, J., Cranfield, M.R., Eckardt, W., Mburanumwe, I., Musana, A., Robbins, A.M., Roy, J., Vigilant, L., and Hickey, J.R. In press. Estimating abundance and growth rates in a wild mountain gorilla population. *Animal Conservation*.
- Gray, M., McNeilage, A., Fawcett, K.T., Robbins, M.M., Ssebide, J.B., Mbula, D., and Uwingeli, P. 2009. Census of the Virunga mountain gorillas: complete sweep method versus monitoring. *African Journal of Ecology* 48:588-599
- Gray, M., Roy, J., Vigilant, L., Fawcett, K., Basabose, A., Cranfield, M., Uwingeli, P., Mburanumwe, I., Kagoda, E., and Robbins, M. 2013. Genetic census reveals increased but uneven growth of a critically endangered mountain gorilla population. *Biol Conserv* 158: 230–238
- Guschanski, K., Vigilant, L., McNeilage, A., Gray, M., Kagoda, E., and Robbins, M.M. 2009. Counting elusive animals: comparing field and genetic census of the entire mountain gorilla population of Bwindi Impenetrable National Park, Uganda. *Biological Conservation* 142: 290-300
- Hickey, J.R., Granjon, A.C., Vigilant, L., Eckardt, W., Gilardi, K.V., Cranfield, M., Musana, A., Masozera, A.B., Babaasa, D., Ruzigandekwe, F., & Robbins, M.M. 2019. Virunga 2015–2016 surveys: monitoring mountain gorillas, other select mammals, and illegal activities. GVTC, IGCP & partners, Kigali, Rwanda
- Korbee, D. 2007. *Environmental Security in Bwindi a focus on farmers*. Institute for Environmental Security. The Hague. 30pp
- Laing, S.E., Buckland, S.T., Burn, R.W., Lambie, D., and Amphlett, A. 2003. Dung and nest surveys: Estimating decay rates. *British Ecological Society*. 40:1102-1111

- Lounsberry, Z.T., Forrester, T.D., Olegario, M.T., Brazeal, J.L., Wittmer, H.U., Sacks, B.N. 2015. Estimating Sex-Specific Abundance in Fawning Areas of a High-Density Columbian Black-Tailed Deer Population Using Fecal DNA. *The Journal of Wildlife Management*. 79(1):39–49
- Marques, T. A., Thomas, L., Martin, S. W., Mellinger, D. K., Ward, J. A., Moretti, D. J., Harris, D., & Tyack, P. L. (2013). Estimating animal population density using passive acoustics. *Biological Reviews*. 88:287–309
- McNeilage, A., Plumptre, A.J., Brock-Doyle, A., Vedder, A., 2001. Bwindi Impenetrable National Park, Uganda: gorilla census. 1997. *Oryx* 35:39–47
- McNeilage, A., Robbins, M.M., Gray, M., Olupot, W., Babaasa, D., Bitariho, R., Kasangaki, A., Rainer, H., Asuma, S., Mugiri, G., Baker, J., 2006. Census of the mountain gorilla *Gorilla beringei beringei* population in Bwindi Impenetrable National Park, Uganda. *Oryx* 40:419–427
- Nkurunungi, J.B., Ganas, J., Robbins, M.M., & Stanford C.B. 2004. A comparison of two mountain gorilla habitats in Bwindi Impenetrable National Park, Uganda. *African Journal of Ecology*. 42:289-297
- Plumptre, A.J., 2000. Monitoring mammal populations with line transect techniques in African forests. *Journal of Applied Ecology* 37(2):356-368
- Robbins, M.M., Roy, J., Wright, E., Kato, R., Kabano, P., Basabose, A., Tibenda, E., Vigilant, L., and Gray, M. 2012. Bwindi Mountain Gorilla Census 2011 – Summary of Results
- Roy, J., Vigilant, L., Gray, M. Wright, E., Kato, R., Kabano, P., Basabose. A., Tibendad, E., Kühl, H., and Robbins, M. 2014. Challenges in the use of genetic mark-recapture to estimate the population size of Bwindi mountain gorillas (*Gorilla beringei beringei*). *Biological Conservation* 180:249-261
- Seber, G. A. F. 1982. *The Estimation of Animal Abundance and Related Parameters*, 2nd ed. London: Griffin
- Sholley, C. 1991. Conserving gorillas in the midst of guerrillas. African Wildlife Foundation. In American Association of Zoological Parks and Aquariums, Annual Conference Proceedings, pp. 30-37
- Twongyirwe, R. Majaliwa, J.G.M. Ebanyat, P., Tenywa, M.M., Sheil, D., Heist, M.V., Oluka, M., and Kumar, L. 2011. Dynamics of forest cover conversion in and around Bwindi Impenetrable Forest, Southwestern Uganda. *J. Appl. Sci. Environ. Manage.* 15(1)
- WWF-Germany and IGCP. 2017. Report: Bwindi-Virunga Landscape Vegetation Mapping. <https://panda.maps.arcgis.com/apps/webappviewer/index.html?id=7e9d539a045e44be9b28237f3d056411>