EcoHealth 14, 552–563, 2017
DOI: 10.1007/s10393-017-1262-9

ECOHEALTH

Original Contribution

Human Exposure to Wild Animals in the Sankuru Province of the Democratic Republic of the Congo

Anne W. Rimoin,[1] Vivian Helena Alfonso,[1] Nicole A. Hoff,[1] Reena H. Doshi,[1] Prime Mulembakani,[2] Nevile K. Kisalu,[3] Jean-Jacques Muyembe,[4] Emile W. Okitolonda,[2] and Linda L. Wright[5]

[1]*Department of Epidemiology, UCLA Fielding School of Public Health, 650 S Charles E Young Drive, Los Angeles, CA*
[2]*Kinshasa School of Public Health, Kinshasa, Democratic Republic of the Congo*
[3]*Vaccine Research Center, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, MD*
[4]*Institut National de Recherche Biomedicale, Kinshasa, Democratic Republic of the Congo*
[5]*National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, MD*

Abstract: Due to the high level of biological diversity in the Congo Basin and human population dependence on bushmeat, the DRC represents an ideal location for expanding knowledge on wild animal exposures and thus the potential for transmission of zoonotic pathogens. However, limited information exists on patterns and extent of contact with wildlife in such communities. Using a cross-sectional study, 14 villages in the Sankuru Province of the DRC were surveyed between August and September 2007. Villagers $\geq$ 1 year of age and at home of the time of the survey were eligible and enrolled to describe and assess factors associated with animal exposures (both activity and type of animal). Among respondents, 91% reported exposure to rodents, 89% to duikers, 78% to non-human primates (NHPs), and 32% reported contact with bats in the month prior to the survey. The most frequently reported activities included eating (95%), cooking (70%), and butchering or skinning of animals (55%). The activities and animals to which subjects had contact varied by sex and age. Moreover, we observed a high correlation of the same activities across animal types. In this and other populations that rely on bushmeat, there is a high frequency of exposure to multiple animal species through various modalities. In the event of future zoonotic disease outbreaks, effective public health interventions and campaigns that mitigate the risk of animal contact during outbreaks need to be broad to include various modes of contact and should be directed to both men and women across all age groups. As available information is limited, further studies are necessary to better understand the complex relationships and exposures individuals have with animals.

Keywords: Zoonosis, Wild animal exposure, Hunting, Butchering, Emerging infections, Spillover

Introduction

Emerging infectious diseases represent one of the most important threats to global public health. While 61% of the 1415 known human pathogens are considered zoonotic,

Linda L. Wright—Retired from National Institute of Child Health and Human Development, National Institutes of Health.

Electronic supplementary material: The online version of this article (doi:10.1007/s10393-017-1262-9) contains supplementary material, which is available to authorized users.

Published online: August 22, 2017

Correspondence to: Anne W. Rimoin, e-mail: arimoin@ucla.edu

75% of "emerging" pathogens are thought to have originated in animals (Taylor et al. 2001). The process of zoonotic disease emergence is complex and multifactorial, with potential factors including environmental drivers or human behaviors, such as increased travel, migration, or trade (Morens et al. 2004). These and other factors can impact the frequency, duration, and type of contact between humans, wildlife, and domestic animals, providing an opportunity for pathogens to move, or "spillover," into new host populations (Taylor et al. 2001; Daszak et al. 2001). Thus, as humans encroach upon and alter habitats, environmental barriers between humans, animals, and the pathogens they carry decrease. However, despite the growing threat of zoonotic emerging infectious diseases, our understanding of the process remains poor.

While predicting the emergence of new zoonoses may be an unattainable goal (Murphy 1998), understanding and characterizing trends in human behaviors as they relate to wildlife hosts and reservoirs of potentially zoonotic pathogens are an imperative first step. To date, a limited number of studies have characterized animal exposure among populations living within or bordering areas of high biological diversity in Africa. For example, in a small study of Nigerian males living within remote hunting communities, animal exposure was ubiquitous and the most prevalent animal contacts included primates, ungulates, and rodents, with hunters having significantly higher reports of butchering and receiving an injury from a wild animal than non-hunters (Friant et al. 2015). However, two studies on exposure to non-human primates resulted in different findings: in a large study of rural Cameroonian villages, animal exposure was significantly higher in forested regions compared to other habitats, and while hunting was a predominantly male activity, little to no difference across sex was observed for butchering of animals (Wolfe et al. 2004). On the other hand, a recent study in the Taï Region of Côte d'Ivoire observed that the type of contact with non-human primates (specifically monkeys and chimpanzees) differed according to demographic characteristics such as age and sex; older males had the highest reports of hunting and butchering, while older females were most likely to report cooking the animals (Mossoun et al. 2015). While significant, these studies provided a limited scope of population exposure to wild animals and highlighted the differences in exposures according to study location, which may reflect differences in cultural practices or gender roles.

The rainforest of the Congo Basin is the second largest in the world after the Amazon, and approximately 50% lies within the boundaries of the Democratic Republic of the Congo (DRC) in Central Africa (Observatory for the Forests of Central Africa 2010; FAO 2011). The DRC possesses the highest level of biological diversity on the African continent, home to more than 400 known mammal species, including a wide variety of non-human primates (NHP), rodents, and bats (Observatory for the Forests of Central Africa 2010). While dependent upon subsistence farming for nutritional and economic sustenance (United Nations 2014), 65% of the estimated 80 million inhabitants of the DRC live in rural, densely forested areas and rely upon hunting locally available wild animals as a source of protein (Golden et al. 2011). To rural communities across the tropical forests of West and Central Africa, bushmeat serves as a significant nutritional, economic, and cultural component of their livelihoods (Milner-Gulland and Bennett 2003) and such dependence may also result in enhanced opportunities for zoonotic disease transmission (Wolfe et al. 2005). Therefore, the DRC is an ideal location for expanding knowledge on animal exposures that may contribute to and result in the transmission of zoonotic agents (Golden et al. 2011; De Merode et al. 2004; Poulsen et al. 2009).

To add to the body of literature quantifying the frequency and type of animal contact, we conducted a population-based survey of all inhabitants (>1 year of age) living in remote, rural villages of the Sankuru Province (formerly Sankuru district) of the DRC to assess the extent of exposure to wild animals. The objective of this study is to identify and describe human–animal contact in the region and to assess sociodemographic risk factors associated with different types of animal contact, particularly to species that are suspected or known reservoirs or hosts of important zoonotic pathogens.

Methods

Study Design and Population

From August to September 2007, we conducted a population-based survey to assess human exposure to wild animals (a proxy measure for risk of zoonotic infection) in healthy, rural village populations in the Sankuru Province of the DRC (Fig. 1). At the time of the study, the population of this province (which was a district with the same geographical boundaries at the time of data collection) was

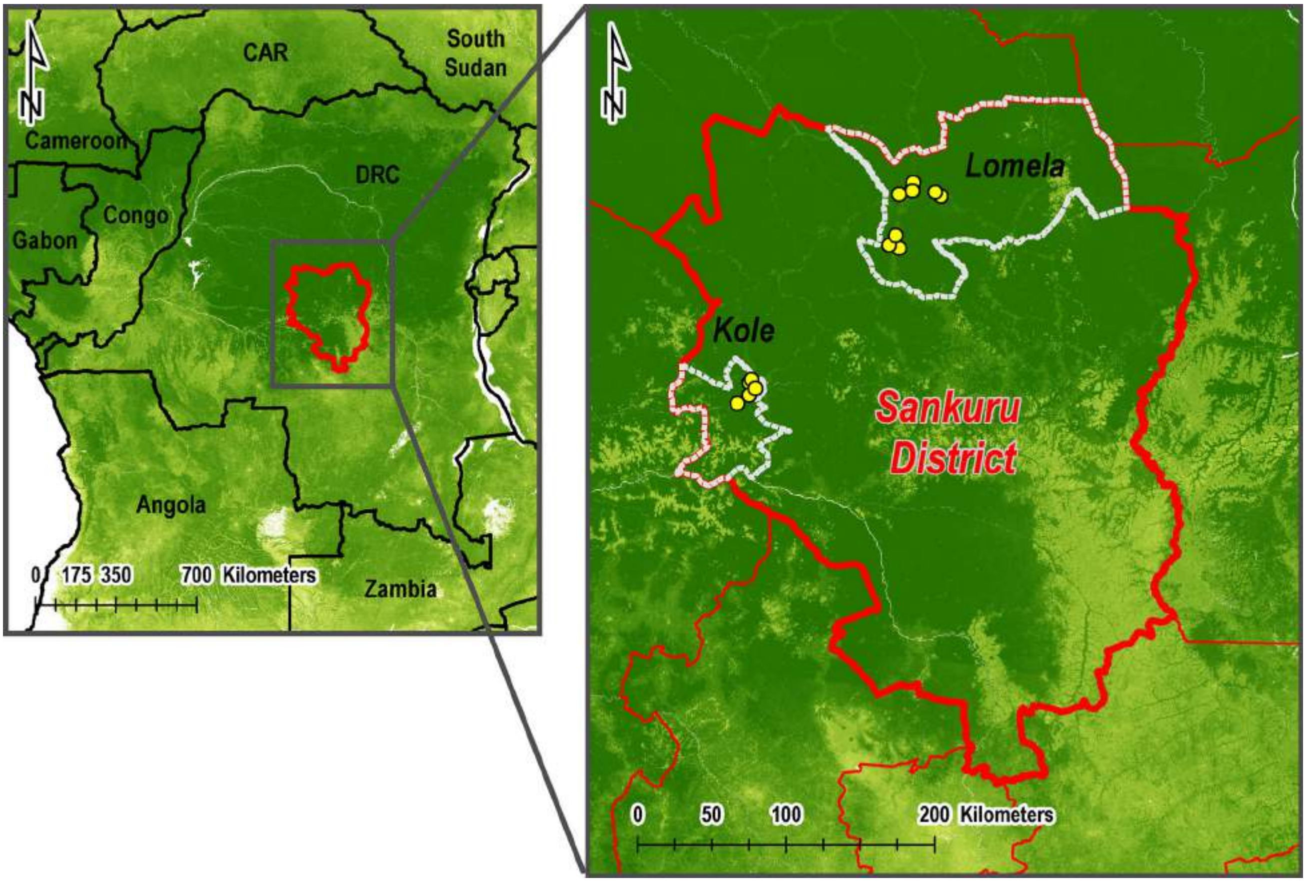

Figure 1. Map of study sites, Sankuru Province, Democratic Republic of the Congo

estimated at 676,839 with a population density of 14.28 people/km^2 (Ministry of Health DRoC 2009). The study design and population have been described elsewhere (Switzer et al. 2012). Briefly, using a village list from the DRC Ministry of Health, 14 locations were randomly selected in two monkeypox endemic health zones: Kole and Lomela. Most villages in this region are located in small clearings of tropical forest, surrounded by agricultural fields.

All healthy individuals $\geq$1 year of age residing in the selected villages were eligible for enrollment. Verbal informed consent was obtained by trained interviewers from all participating adults as well as assent from children 7–18 years with parental or guardian consent. Parents/guardians of participants $<$7 years of age answered on behalf of their children. A standard questionnaire was administered to all participants by trained health care workers in either Tetela (the local language) or French. All questionnaire data were double-entered into a Microsoft Access database for quality control purposes.

Among the 7545 individuals living within the participating villages (obtained from a door-to-door census conducted one week prior to participant enrollment), only individuals $>$1 year that resided in participating villages were eligible (n = 5687) (Fig. 2). While approximately 20% declined participation among those eligible for enrollment, 4574 were enrolled in the study. Missing information on animal exposure questionnaire resulted in the final study population of 3140 subjects. Those with incomplete survey information (n = 1434) were younger and more likely to be male and from Kole health zone (*data not shown*).

Animal Exposure Assessment

We collected information on exposure to wild animals using locally appropriate taxonomic categories derived from focus group interviews and lists of animals found in that region. Participants were shown a representative photograph or drawing of 28 animals and asked about the frequency and type of exposure, if any, to the animal in the past month. Participants were also able to specify additional animals not included on the preformed list using an "other" category. Animal categories were defined as follows: rodent included exposure to squirrel, porcupine, African dormouse, Gambian rat, rat, and mouse; eutheria included exposure to pangolin and Elephant shrew; lorisidae included exposure to potto and galago; and non-human primate included exposure to *Cercopithecus ascanius, Cercocebus chrysogaster, Colobus angolensis, Cercopithecus neglectus, Cercopithecus wolfi, Cercopithecus nicitans, Procolobus tholloni,* and monkey not identified. Exposure was defined as binary (any or no exposure) for both the animal species/categories (included in Table 2) and for the following activities: hunting, butchering/skinning, cooking, eating, playing with/getting bitten/scratched and picking up dead animal carcasses to eat.

Covariate Assessment

Basic sociodemographic information collected from participants and included in analyses was as follows: age (years), sex, health zone, ethnic group, and frequency of forest visits. Among children, school attendance was assessed, while educational attainment, socioeconomic status, martial status, and occupation (based upon primary and secondary designation) were assessed among those 15 years of age or older. A wealth index, adapted from Malleson et al. (2008), was created to assess association of socioeconomic status with animal exposure using the following reported assets: domestic animals, fields, radio, bicycle, sewing machine, and motorcycle. Each reported asset was given a value of 1 (maximum value of 6) and each extra point indicated an asset-rich household. Categories for socioeconomic status were created as follows: low (0 re-

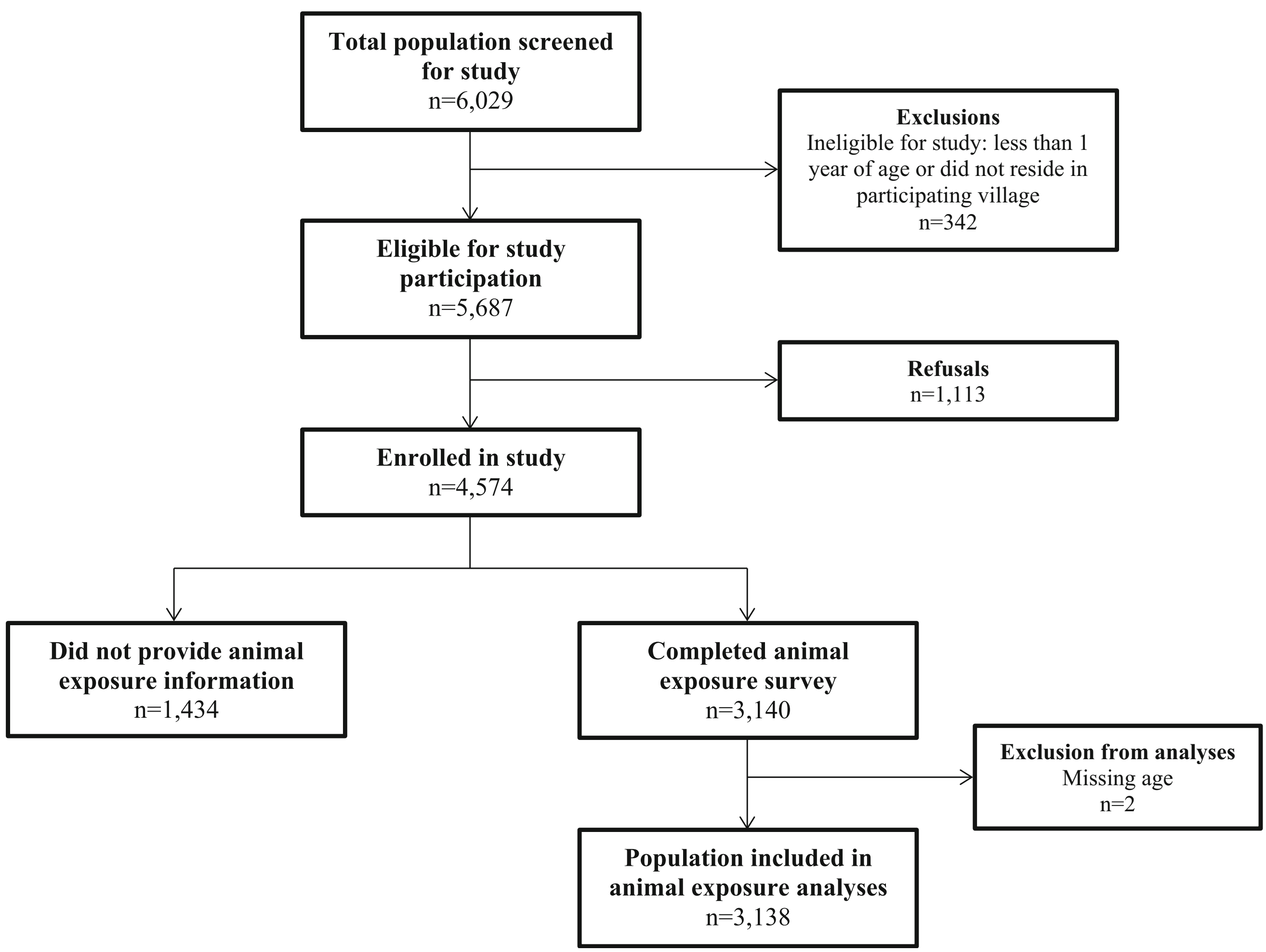

Figure 2. Flowchart of eligibility criteria and study enrollment, Sankuru Province, Democratic Republic of the Congo

ported assets), middle (1–2 reported assets), high (≥3 reported assets).

Statistical Analyses

We initially ran frequency distributions for population characteristics and wild animal exposures (both activity and taxa) to describe the population. We then assessed the relationship between sex and age with the animal exposures of interest, which included activities (eating, cooking, butchering/skinning, and hunting) and animals (rodents, duikers, non-human primates, and bats), in crude, univariate logistic regression. Models in which the Wald Chi-square p value for both age and sex was <0.05 were then assessed in multivariate logistic regression. To further describe differences across sex and age and assess for possible effect measure modification, we performed multivariate logistic models including age, sex, and age–sex interaction term for animal exposures, separately.

As approximately 30% of subjects (n = 1434) did not provide responses to the animal contact survey and thus were excluded from the final study population, in sensitivity analyses we assessed the impact of this missingness on findings using inverse probability censoring weights (Cole and Hernan 2008). All analyses were carried out using SAS software, version 9.4 (SAS Institute, Cary, NC).

Ethical Approval

Ethical approval for this study was obtained from UCLA Fielding School of Public Health and Kinshasa School of Public Health.

Results

Fifty-nine percent of the study participants were female, and the median reported age was 23 (21 and 25 years, for men and women, respectively) (Table 1). The majority of children 7–15 years reported currently attending school, and only 44% of women compared to 80% of men reported completing primary education. Among the 447 adult respondents who reported hunting as a primary or secondary source of income, 68% were male and nearly three-fourths reported visiting the forest > 4 days per week in the previous month.

Regardless of the animal, the vast majority of contact occurred by eating wildlife, ubiquitous in this study population as 95% reported this behavior, followed by cooking (70%) and butchering/skinning (55%); hunting was reported by only 19% of participants (Table 2). In response to the animals to which participants came into contact, the most commonly reported animal species were rodents (91%), mainly squirrels and porcupines, and duikers (89%). Among the key animal groups of interest including rodents, duikers, NHPs, and bats, we found extensive overlap in reporting of animal contact. Overall, 28% (n = 894) of the study population ≥5 years of age reported any exposure to all four and 42% (n = 1308) reported any combination of the contact to rodents, duikers, and NHPs; only 3% of the population reported no contact to these selected animals (Fig. 3). Moreover, we also found subjects tend to perform similar behaviors across animal types. The pair-wise Pearson's correlation between hunting for the most common reported animals (including rodents, duikers and NHPs) ranged from 0.58 to 0.74, and from 0.68 to 0.79 for butchering/skinning, and 0.62 to 0.78 for cooking (Supplemental Table 1).

In univariate logistic regression, we assessed the associations between age, sex, and animal exposures, independently. Overall, women were more likely to cook (OR 6.1, 95% CI (5.2, 7.3)), while men were significantly more likely to report butchering or skinning (OR 1.6), hunting (OR 20.0), picking up dead animals for later consumption (OR 2.0), and being scratched, bitten, or playing with animals (OR 3.3) (Table 3). Additionally, compared to 5–9 year olds, the older the subject, the more likely they are to report cooking, butchering or skinning, hunting, and picking up dead animals. As age may modify the relationship between sex and overall exposure activities regardless of animal (and vice versa), we then assessed the relationship between age, sex, and their interaction in multivariate logistic regression using the youngest male age group (5–9 year olds) as the referent category (Table 4). Due to limited sample size, the activities assessed included eating, cooking, butchering or skinning, and hunting. Within age strata, there was no difference in overall consumption of wildlife according to sex. Across all ages, women were more likely to cook than men (with the magnitude of the relationship increasing with increasing age) and men were more likely to butcher, skin, and hunt than women.

We then assessed multivariate logistic regression analyses between age, sex, and their interaction with animal activities within animal types individually for rodents, duikers, NHPs, and bats (Table 5). As a limited number of respondents reported picking up animal carcasses found dead in the forest or being scratched, bitten, or playing with

Table 1. Frequency Distribution of Selected Population Characteristics of Study Participants, Sankuru Province, Democratic Republic of Congo (n = 3138)

		Female (n = 1859)	Male (n = 1279)
	n (%)	n (%)	n (%)
Population characteristics*			
Age			
1–4 years	23 (<1)	10 (1)	13 (1)
5–9 years	357 (11)	181 (10)	176 (14)
10–14 years	542 (17)	271 (15)	271 (21)
15–49 years	1791 (57)	1147 (62)	644 (50)
50+ years	425 (14)	250 (13)	175 (14)
Sex			
Female	1859 (59)	1859 (100)	–
Male	1279 (41)	–	1279 (100)
Health zone			
Kole	1951 (63)	1160 (63)	791 (63)
Lomela	1154 (37)	681 (37)	473 (37)
Ethnicity			
Batetela	940 (30)	552 (30)	388 (31)
Bankutshu	78 (3)	57 (3)	21 (2)
Ohindo	1726 (5)	1027 (56)	699 (55)
Other	370 (12)	212 (11)	158 (12)
Frequency of forest visits for regular activities†			
Never	361 (12)	177 (10)	184 (15)
1–4 times per month	903 (29)	422 (23)	481 (38)
>4 times per month	1821 (59)	1229 (67)	592 (47)
Children‡ characteristics			
Attends school			
Yes	771 (89)	375 (87)	396 (92)
No	92 (11)	56 (13)	36 (8)
Adult§ characteristics			
Education			
None	285 (13)	268 (20)	17 (2)
Some primary education	638 (30)	495 (37)	143 (18)
Completed primary education	906 (42)	495 (37)	411 (51)
Completed secondary education or beyond¶	330 (15)	94 (7)	236 (29)
Socioeconomic status#			
Low	477 (22)	311 (23)	166 (20)
Middle	1,313 (60)	905 (66)	408 (50)
High	398 (18)	161 (12)	237 (29)
Marital status			
Single	642 (29)	365 (26)	277 (34)
Married	1379 (63)	861 (62)	518 (64)
Divorced/separated/widow(er)	172 (8)	157 (11)	15 (2)

Table 1. continued

		Female (n = 1859)	Male (n = 1279)
	n (%)	n (%)	n (%)
Occupation**			
Hunter	447 (21)	142 (11)	305 (39)
Other	1633 (79)	1160 (89)	473 (61)

*Population included all respondents of all ages.

†Regular activities include, but are not limited to: hunting, cultivating, searching for water or wood, foraging, and fishing.

‡Children included all respondents under the age of 15 (n = 922).

§Adults included all respondents 15 years of age or older (n = 2216).

¶Category includes completion of secondary education, apprenticeship, higher education, or university.

#SES categorization is based on calculated wealth index derived from the following reported assets: domestic animals, fields, radio, bicycle, sewing machine, and motorcycle. Each reported asset was given a value of 1 (maximum value of 6), and categories for SES are as follows: low (0 reported assets), middle (1–2 reported assets), high (≥3 reported assets).

**Occupation of hunter was assigned if participant indicated hunting or fishing as either a primary or secondary source of income.

animals, and as the vast majority of hunters were male, we were unable to assess these relationships within animal subtypes. For cooking, we found a similar pattern of association for rodents, duikers, and NHPs: females across all age groups were more likely to cook than their male counterparts and the relationship is strongest for those ≥ 15 years of age. The association between sex and cooking for bats was consistent regardless of age group as women were approximately three times as likely to report this behavior compared to their male counterparts (OR range: 2.3–4.1).

Discussion

While human exposure to wild animals was pervasive, the mode of contact varied by age group and sex. The most frequently reported animal contacts included rodents, specifically squirrels and porcupines, and duikers. The vast majority of the population reported eating animals, with cooking and butchering or skinning as the other frequent animal activities; however, only a small proportion of the population reported hunting. The activities and animals to which subjects had contact varied by sex and age: in general, males were more likely to hunt and females were more likely to cook wild animals, while there were no significant differences by sex among participants ≥ 15 years of age in butchering or skinning of NHPs and bats. Moreover, we observed a high correlation of activities across animal types—individuals who hunt, butcher and skin or cook one animal type perform the same activities with a number of other animal types and species. Therefore, we demonstrate that animal contact is not isolated to a small subgroup of the population, rather is ubiquitous across age and sex in this surveyed population from forest villages in Kole and Lomela health zones in the Sankuru Province of the DRC.

The basis for a majority of the reported animal contact in our study is bushmeat, which provides an important gateway for disease emergence. Hunting (which may include tracking, capturing, handling, and transportation) and butchering (which may include opening, cutting, and other preparations) of wild animals provide ample opportunity for transmission of infectious zoonotic agents via intimate contact with tissue, blood, viscera, feces, or other bodily fluids (De Merode et al. 2004; Wolfe et al. 2004; Gessain et al. 2013; Aghokeng et al. 2010; Calvignac-Spencer et al. 2012; Peeters et al. 2002; Locatelli and Peeters 2012; Kalish et al. 2005). As many people worldwide participate in hunting and butchering activities, a large number of people are constantly being challenged by zoonotic agents (Wolfe et al. 2004; Kalish et al. 2005; Wolfe et al. 2005; Calattini et al. 2005). For example, serosurveys of wild animals in DRC suggest that monkeypox has a number of sylvatic hosts, including rope and sun squirrels and giant pouched rats (Hutson et al. 2011; Jezek et al. 1987). Similarly, NHPs and antelopes (Lahm et al. 2007) as well as bats (Dobson 2005; Luis et al. 2013; Leroy et al. 2009) can be infected with Ebola virus and transmit the infection to humans through hunting, butchering, or preparing infected carcasses. Additionally, human immunodeficiency virus evolved from related viruses of non-human primates and likely eventually entered into human populations through

Table 2. Frequency Distribution (n (%)) of Activities Resulting in Any Wild Animal Exposure and Taxa Among Study Participants, Sankuru Province, Democratic Republic of Congo (n = 3138)

	Yes	No
Activity-specific exposures		
Eat	2992 (95)	146 (5)
Cook	2203 (70)	935 (30)
Butcher/skin	1714 (55)	1424 (45)
Hunt	594 (19)	2544 (81)
Pick up dead	268 (9)	2870 (91)
Scratch/bite/play	146 (5)	2992 (95)
Animal-specific exposures		
Rodent	2855 (91)	283 (9)
Squirrel	2447 (78)	691 (22)
Porcupine	2281 (73)	857 (27)
Gambian Rat	1862 (59)	1276 (41)
Rat	1161 (37)	1977 (63)
African Doormouse	583 (19)	2555 (81)
Mouse	526 (17)	2612 (83)
Duiker	2804 (89)	334 (11)
Non-human primate	2455 (78)	683 (22)
Lophocebus aterrimus	1369 (44)	1769 (56)
Cercopithecus ascanius	1236 (39)	1902 (61)
Procolobus tholloni	1001 (32)	2137 (68)
Colobus angolensis	947 (30)	2191 (70)
Cercopithecus neglectus	946 (30)	2192 (70)
Cercopithecus wolfi	719 (23)	2419 (77)
Cercopithecus nicitans	544 (17)	2594 (83)
Cercocebus chrysogaster	532 (17)	2606 (83)
Pan paniscus	509 (16)	2629 (84)
Monkey not identified	1889 (60)	1249 (40)
Eutheria	2441 (78)	697 (22)
Pangolin	2263 (72)	875 (28)
Elephant Shrew	1631 (52)	1507 (48)
Wild Boar	1848 (59)	1290 (41)
Wild Bird	1832 (58)	1306 (42)
Bat	989 (32)	2149 (68)
Lorisidae	930 (30)	2208 (70)
Galago	808 (26)	2330 (74)
Potto	737 (23)	2401 (77)
Wild Cat	976 (31)	2162 (69)
Reptile	774 (25)	2364 (75)
Elephant	615 (20)	2523 (80)

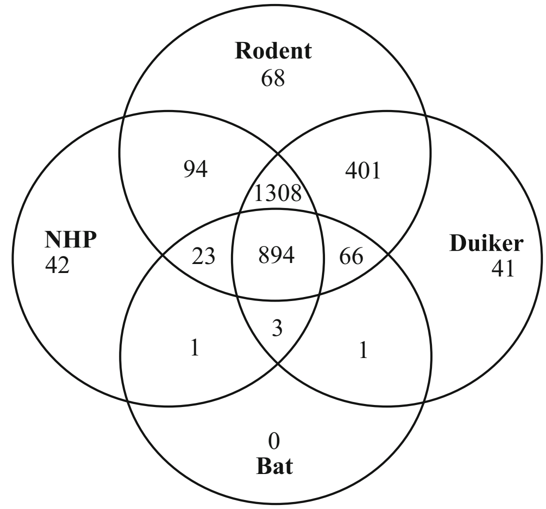

Figure 3. Overlapping exposure* to rodent, duiker, non-human primate, and bat among participants ≥ 5 years of age, Sankuru Province, Democratic Republic of the Congo. *Exposure refers to any contact to the animal types indicated

bushmeat hunting and butchering (Locatelli and Peeters 2012). However, exposure in this manner is part of a broader spectrum of activities in sub-Saharan Africa that bring people and animals into direct and potentially risky contact. Therefore, once bushmeat is caught, a cascade of high-risk exposures ensue. For example, in a study of the same population in the DRC, simply entering the forest may increase the risk of SFV infection among participants without known primate contact (Switzer et al. 2012). Moreover, previous research on exposure to NHPs indicates that simple handling of such animals promotes transmission events (Leroy et al. 2009; Wolfe et al. 2007; Davies and Pedersen 2008).

The findings from our study further expand upon and support some of the observations made in a recent western Ugandan household survey (Paige et al. 2014). While only 20% of the population reported contact with either wild or domestic animals, the animal contact survey was administered only to subjects either reporting an injury from an animal or NHP contact. Despite this limitation, the Ugandan study observed overlapping exposures to animals and the modes through which contact occurs as documented in our study. Such high correlation of animal contact within the same time period through different modes (both within and across species) and the fact that many species are able to harbor multiple pathogens (as reservoirs and intermediate or incidental hosts) make it difficult to separate the potential effect of any single behavior or species on risk of exposure to zoonotic pathogens. Furthermore, this study also observed that the type of animals for which participants reported contact varied by geographical land cover of surveyed communities, which could not be assessed in our study as all study

Table 3. Odds Ratios (OR) and 95% Confidence Intervals (95% CI) From Univariate Logistic Regression for Wild Animal-Specific Activities Among Study Participants, Sankuru Province, Democratic Republic of Congo (n = 3138)

	Eat	Cook	Butcher/Skin	Hunt	Pick up dead	Scratch/bite/play
Sex						
Male	Ref	Ref	Ref	Ref	Ref	Ref
Female	1.23 (0.88, 1.72)	6.12 (5.16, 7.25)	0.63 (0.54, 0.73)	0.05 (0.04, 0.06)	0.49 (0.38, 0.63)	0.30 (0.21, 0.43)
Age category						
5–9 years	Ref	Ref	Ref	Ref	Ref	Ref
10–14 years	1.56 (0.87, 2.79)	1.65 (2.00, 3.50)	2.38 (1.76, 3.22)	1.87 (1.18, 2.98)	4.27 (1.47, 12.37)	1.59 (0.84, 3.01)
15–49 years	1.75 (1.08, 2.81)	8.90 (6.93, 11.44)	6.49 (4.97, 8.48)	3.50 (2.33, 5.26)	10.84 (4.00, 29.38)	1.30 (0.73, 2.30)
50+ years	1.11 (0.62, 1.96)	5.87 (4.31, 8.01)	4.67 (3.41, 6.39)	3.56 (2.27, 5.61)	9.93 (3.53, 27.96)	0.47 (0.20, 1.13)

Table 4. Odds Ratios (OR) and 95% Confidence Intervals (95% CI) From Multivariate Logistic Regression* for Wild Animal-Specific Activities Among Study Participants, Sankuru Province, Democratic Republic of Congo (n = 3138)

	Age category	Eat	Cook	Butcher/Skin	Hunt
Males	5–9 years	Ref	Ref	Ref	Ref
	10–14 years	1.87 (0.66, 5.26)	2.18 (1.34, 3.57)	2.69 (1.57, 4.59)	2.16 (0.40, 11.65)
	15–49 years	1.90 (0.81, 4.47)	3.58 (2.21, 5.79)	9.79 (6.10, 15.73)	7.63 (2.20, 26.53)
	50+ years	1.17 (0.42, 3.24)	2.30 (1.15, 4.58)	6.05 (3.48, 10.51)	6.55 (1.55, 27.76)
Females	5–9 years	1.48 (0.96, 2.27)	1.68 (1.34, 2.11)	0.74 (0.58, 0.96)	0.11 (0.06, 0.20)
	10–14 years	1.87 (1.84, 1.89)	5.74 (5.58, 5.89)	1.57 (1.52, 1.61)	0.07 (0.06, 0.09)
	15–49 years	2.22 (2.07, 2.37)	50.98 (49.79, 52.20)	4.29 (4.04, 4.55)	0.27 (0.26, 0.28)
	50+ years	1.46 (1.45, 1.46)	41.78 (38.60, 45.21)	3.10 (3.01, 3.19)	0.18 (0.17, 0.20)

*Multivariate logistic regression included the following predictors: age, sex, and age–sex interaction term.

villages were in close proximity to one another and thus had similar environments. Moreover, study sites were selected to represent different primate habitats, whereas villages in our study were randomly selected to estimate overall population animal contact in forested regions of northern Sankuru Province.

Little is known about the complexity of the emergence or re-emergence process, but with approximately 75% of human emerging infectious diseases classified as zoonoses (Taylor et al. 2001), simply understanding the extent and type of animal contact experienced by individuals is critical. As limited information exists on patterns of contact with wildlife in communities that rely heavily on bushmeat, this study fills a knowledge gap by exploring various animals and the mode of contact experienced by rural villagers from the Congo Basin. A strength of this study is that data collection did not take place in the context of an outbreak; thus, findings are not influenced by the urgency of an investigation nor are biased due to the need to find information on specific implicated species (Wolfe et al. 2004; Paige et al. 2014). While this large study provided population estimates of wild animal contact in remote forest villages of the northern Sankuru Province, our study was limited in both the number of villages surveyed (only within Kole and Lomela health zones) and in geographical and ecological variation as all villages were located in forested areas. Reports of animal exposures varied from one location to another (Friant et al. 2015; Wolfe et al. 2004; Mossoun et al. 2015; Paige et al. 2014); thus, caution should be taken in extrapolating such findings to other locations.

Moreover, while a comprehensive list of locally available wild animals was attempted, limitations in time and resources resulted in the inclusion of only the most common in this region. While local names and photographs were used to minimize exposure misclassification, it is possible that participants may have mistaken one animal

Table 5. Odds Ratios (OR) and 95% Confidence Intervals (95% CI) From Multivariate Logistic Regression* For Wild Animal-Specific Activities Among Study Participants, Sankuru Province, Democratic Republic of Congo (n = 3138)

		Rodent†			Duiker		
	Age	Eat	Cook	Butcher/skin	Eat	Cook	Butcher/skin
Male	5–9 years	Ref	Ref	Ref	Ref	Ref	Ref
	10–14 years	1.12 (0.56, 2.24)	1.75 (1.07, 2.87)	2.51 (1.42, 4.43)	1.11 (0.57, 2.14)	2.12 (1.26, 3.58)	1.77 (0.98, 3.17)
	15–49 years	1.61 (0.87, 2.99)	3.09 (1.98, 4.84)	8.52 (5.18, 14.02)	1.47 (0.84, 2.58)	4.56 (2.85, 7.30)	8.59 (5.15, 14.31)
	50+ years	1.04 (0.49, 2.20)	2.16 (1.22, 3.84)	5.15 (2.90, 9.13)	1.13 (0.57, 2.26)	2.78 (1.54, 5.01)	7.25 (4.04, 13.01)
Female	5–9 years	1.33 (0.97, 1.84)	1.57 (1.24, 1.99)	0.70 (0.53, 0.91)	1.13 (0.97, 1.51)	2.16 (1.66, 2.80)	1.01 (0.78, 1.34)
	10–14 years	0.93 (0.90, 0.96)	4.46 (4.30, 4.62)	1.34 (1.31, 1.38)	1.38 (1.36, 1.39)	7.53 (7.12, 7.97)	2.12 (2.03, 2.22)
	15–49 years	1.51 (1.42, 1.61)	23.38 (22.16, 24.67)	4.00 (3.77, 4.26)	1.72 (1.62, 1.83)	34.48 (31.98, 37.17)	5.20 (4.82, 5.62)
	50+ years	1.31 (1.30, 1.33)	18.39 (18.32, 18.45)	2.76 (2.67, 2.85)	1.39 (1.38, 1.40)	27.42 (26.83, 28.03)	3.98 (3.80, 4.18)
		Non-human primate‡			Bat		
Male	5–9 years	Ref	Ref	Ref	Ref	Ref	Ref
	10–14 years	1.08 (0.62, 1.86)	1.78 (1.05, 3.03)	2.03 (1.09, 3.80)	0.89 (0.53, 1.47)	1.11 (0.51, 2.41)	1.90 (0.62, 5.82)
	15–49 years	1.71 (1.07, 2.73)	3.59 (2.25, 5.72)	8.56 (4.95, 14.81)	1.25 (0.82, 1.90)	2.63 (1.37, 5.06)	7.30 (2.75, 19.40)
	50+ years	1.23 (0.70, 2.17)	2.31 (1.31, 4.06)	5.85 (3.14, 10.89)	1.19 (0.71, 1.99)	1.79 (0.84, 3.81)	6.10 (2.13, 17.49)
Female	5–9 years	1.91 (1.50, 2.44)	2.45 (1.88, 3.20)	1.37 (1.00, 1.88)	1.38 (1.10, 1.74)	2.30 (1.54, 3.42)	1.14 (0.65, 2.01)
	10–14 years	1.62 (1.61, 1.64)	6.25 (5.86, 6.67)	2.88 (2.69, 3.07)	1.03 (1.01, 1.05)	3.48 (3.09, 3.92)	2.02 (1.80, 2.25)
	15–49 years	1.87 (1.78, 1.96)	20.38 (18.62, 22.30)	6.27 (5.68, 6.92)	1.18 (1.11, 1.25)	7.14 (6.08, 8.39)	4.30 (3.62, 5.10)
	50+ years	1.59 (1.58, 1.60)	17.49 (16.65, 18.36)	4.39 (4.09, 4.70)	1.27 (1.24, 1.29)	7.25 (6.36, 8.27)	4.61 (4.01, 5.31)

*Multivariate logistic regression included the following predictors: age, sex, and age*sex interaction term.

†Includes the following animals: squirrel, porcupine, gambian rat, rat, African dormouse, and mouse.

‡Includes the following species: *Lophocebus aterrimus*, *Cercopithecus ascanius*, *Procolobus tholloni*, *Colobus angolensis*, *Cercopithecus neglectus*, *Cercopithecus wolfi*, *Cercopithecus nicitans*, *Cercocebus chrysogaster*, *Pan paniscus*, and monkey not identified.

for another. Furthermore, while exposure information was collected on a wide variety of wild animals, in-depth analyses were limited to species most likely to carry pathogenic zoonotic agents. NHPs, bats, and rodents (including rats, mice, squirrels, and porcupines) are known or suspected reservoirs of zoonotic pathogens and have caused important outbreaks of human disease (Wolfe et al. 2004; Jezek et al. 1986; Hutson et al. 2011; Lahm et al. 2007; Dobson 2005; Luis et al. 2013; Leroy et al. 2009; Khodakevich et al. 1987; Hayman et al. 2013; Organization 1996). Duikers were included in species-specific analyses because they are ubiquitous in Congolese diets and have been identified as potential intermediate hosts for Ebola and other emerging infectious diseases (Locatelli and Peeters 2012; Dobson 2005; Luis et al. 2013; Khodakevich et al. 1987; Hayman et al. 2013; Meerburg et al. 2009; Khodakevich et al. 1987; Parker et al. 2007). Lastly, limitations in memory recall may have also affected the accuracy of reported exposures to wild animals. Given the large range of biodiversity and high frequency of exposure to animals, remembering the specifics of contacts within the previous month may have been difficult for participants. However, as limitations in recall likely impacted all subjects, the bias would be non-differential and therefore result in an underestimate of true associations. As this was a cross-sectional study and exposure was assessed only for the previous month, we were unable to capture how seasonality may impact exposure in this population.

Estimates from the Congo Basin suggest that over 282 grams of bushmeat is consumed per person per day, with nearly 5 million tons of bushmeat extracted annually (Fa et al. 2002). The intensity and extent of the bushmeat trade in Central Africa have increased over the past two decades (Karesh and Noble 2009); therefore, as populations grow and the demand for bushmeat increase, this will lead to increases in the exposure of humans to potentially zoonotic agents. Due to the observed widespread animal contact in our study population, these data are useful in the creation of targeted public health interventions directed at the human–animal interface. We have characterized patterns of exposure (both animal type and mode of contact) across age and sex, which provides a basis for prevention efforts to mitigate risk or contact with animals suspected to be involved in disease outbreaks. For example, in the Republic of the Congo, targeted Monkeypox outreach through film-based educational activities was provided to members of both sexes $\geq$ 13 years of age of communities at risk of Monkeypox with significant gains reported in disease recognition, transmission, and mitigation of risk (Roess et al. 2011). Therefore, such data should be taken into consideration when developing educational campaigns and other strategies to reduce risk of zoonotic transmission during outbreak settings in rural villages with such high biodiversity. While little is known about the complexity of the emergence or re-emergence process, further studies are needed to further characterize and describe wild and domestic animal contact in areas of extensive bushmeat consumption and high biological diversity.

Acknowledgements

Many individuals contributed to the design of this manuscript and data collection in the field. We thank the health care workers in the Sankuru Province and the DRC Ministry of Health for their surveillance efforts and assistance with the implementation of this study. We also thank Cyrus Sinai for assisting with Fig. 1 (DRC map) and Haroutune Armenian for comments and suggestions on the manuscript.

Funding

This work was supported by: the Eunice Kennedy Shriver National Institute of Child Health and Human Development, Global Network for Women's and Children's Health Research; National Institute of Allergy and Infectious Diseases, Division of Infectious Diseases and Microbiology (5K01AI074810-05); Fogarty International Center, Research and Policy for Infectious Disease Dynamics (RAPIDD) program of the Science and Technology Directorate, Department of Homeland Security; and the Faucett Catalyst Fund.

Author Contribution

AWR designed the study, supervised all aspects of study implementation, collected the data, guided data analysis; VHA conducted data analysis, developed figures and tables; AWR and VHA wrote and edited the manuscript; NAH and RHD assisted with data cleaning and manuscript editing; PMM, NKK, EOW and JJM participated in study design and implementation; LLW participated in study design, implementation, and editing the manuscript.

Compliance with Ethical Standards

Conflict of interest The authors report no conflict of interest with regard to this publication.

References

Aghokeng AF, Ayouba A, Mpoudi-Ngole E, Loul S, Liegeois F, Delaporte E, et al. (2010) Extensive survey on the prevalence and genetic diversity of SIVs in primate bushmeat provides insights into risks for potential new cross-species transmissions. *Infection, Genetics and Evolution* 10(3):386–396

Calattini S, Chevalier SA, Duprez R, Bassot S, Froment A, Mahieux R, et al. (2005) Discovery of a new human T-cell lymphotropic virus HTLV-3 in Central Africa. *Retrovirology* 9:30

Calvignac-Spencer S, Adjogoua EV, Akoua-Koffi C, Hedemann C, Schubert G, Ellerbrok H, et al. (2012) Origin of human T-lymphotropic virus type 1 in rural Cote d'Ivoire. *Emerg Infect Dis* 18(5):830–833

Cole SR, Hernan MA (2008) Constructing inverse probability censoring weight for marginal structural models. *American Journal of Epidemiology* 168(6):656–664

Daszak P, Cunningham AA, Hyatt AD (2001) Anthropogenic environmental change and the emergence of infectious diseases in wildlife. *Acta Tropica* 78(2):103–116

Davies TJ, Pedersen AB (2008) Phylogeny and geography predict pathogen community similarity in wild primates and humans. *Proceedings of the Royal Society of London B: Biological Sciences* 275(1643):1695–1701

De Merode E, Homewood K, Cowlishaw G (2004) The value of bushmeat and other wild foods to rural households living in extreme poverty in Democratic Republic of Congo. *Biological conservation* 118(5):573–581

Dobson AP (2005) What links bats to emerging infectious diseases? *Science* 310(5748):628–629

Fa JE, Peres CA, Meeuwig J (2002) Bushmeat exploitation in tropical forests: an intercontinental comparison. *Conserv Biol* 16:232–237

FAO (2011) The State of Forests in the Amazon Basin, Congo Basin and Southeast Asia: A report prepared for the Summit of the Three Rainforest Basins, Brazzaville, Republic of Congo. Report. Brazzaville, Republic of Congo: Food and Agriculture Organization of the United Nation; 2011 31 May-3 June, 2011. Report No.: ISBN 978-92-5-106888-5

Friant S, Paige SB, Goldberg TL (2015) Drivers of bushmeat hunting and perceptions of zoonoses in Nigerian hunting communities. *PLOS Neglected Tropical Diseases* 9(5):e0003792

Gessain A, Rua R, Betsem E, Turpin J, Mahieux R (2013) HTLV-3/4 and simian foamy retroviruses in humans: discovery, epidemiology, cross-species transmission and molecular virology. *Virology* 435(1):187–199

Golden CD, Fernald LCH, Brashares JS, Rasolofoniaina BJR, Kremen C (2011) Benefits of wildlife consumption to child nutrition in a biodiversity hotspot. *Proceedings of the National*

Academy of Sciences of the United States of America 108(49):19653–19656

Hayman DT, Bowen RA, Cryan PM, McCracken GF, O'Shea TJ, Peel AJ, et al. (2013) Ecology of zoonotic infectious diseases in bats: current knowledge and future directions. *Zoonoses Public Health* 60(1):2–21

Hutson CL, Carroll DS, Gallardo-Romero N, Weiss S, Clemmons C, Hughes CM, et al. (2011) Monkeypox disease transmission in an experimental setting: prairie dog animal model. *PLoS One* 6(12):e28295

Jezek Z, Arita I, Mutombo M, Dunn C, Nakano J, Szczeniowski M (1986) Four generations of probable person-to-person transmission of human monkeypox. *American journal of epidemiology* 123(6):1004–1012

Jezek Z, Nakano JH, Arita I, Mutombo M, Szczeniowski M, Dunn C (1987) Serological survey for human monkeypox infections in a selected population in Zaire. *J Trop Med Hyg* 90(1):31–38

Kalish ML, Wolfe ND, Ndongmo CB, McNicholl J, Robbins KE, Aidoo M, et al. (2005) Central African hunters exposed to simian immunodeficiency virus. *Emerging Infectious Disease* 11:1928–1930

Karesh WB, Noble E (2009) The bushmeat trade: increased opportunities for transmission of zoonotic disease. *Mt Sinai J Med* 76:429–434

Khodakevich L, Szczeniowski M, Manbuma D, Jeze Z, Marennikova S, Nakano J, et al. (1987) The role of squirrels in sustaining monkeypox virus transmission. *Trop Geogr Med* 39(2):115–122

Khodakevich L, Szczeniowski M, Manbuma D, Jezek Z, Marennikova S, Nakano J, et al. (1987) Monkeypox virus in relation to the ecological features surrounding human settlements in Bumba zone. *Zaire. Trop Geogr Med* 39(1):56–63

Lahm SA, Kombila M, Swanepoel R, Barnes RF (2007) Morbidity and mortality of wild animals in relation to outbreaks of Ebola haemorrhagic fever in Gabon, 1994-2003. *Trans R Soc Trop Med Hyg* 101(1):64–78

Leroy EM, Epelboin A, Mondonge V, Pourrut X, Gonzalez J-P, Muyembe-Tamfum J-J, et al. (2009) Human Ebola outbreak resulting from direct exposure to fruit bats in Luebo, Democratic Republic of Congo, 2007. *Vector-borne and zoonotic diseases* 9(6):723–728

Locatelli S, Peeters M (2012) Cross-species transmission of simian retroviruses: how and why they could lead to the emergence of new diseases in the human population. *AIDS* 26(6):659–673

Luis AD, Hayman DT, O'Shea TJ, Cryan PM, Gilbert AT, Pulliam JR, et al. (2013) A comparison of bats and rodents as reservoirs of zoonotic viruses: are bats special? *Proc Biol Sci* 280(1756):20122753

Malleson R, Asha S, Sunderland T, Burnham P, Egot M, Obeng-Okrah K, et al. (2008) A methodology for assessing rural livelihood strategies in West/Central Africa: lessons from the field. *Ecological and Environmental Anthropology*

Meerburg BG, Singleton GR, Kijlstra A (2009) Rodent-borne diseases and their risks for public health. *Critical Reviews in Microbiology* 35(3):221–270

Milner-Gulland EJ, Bennett EL (2003) Wild meat: the bigger picture. *Trends Ecol Evol* 18:351–357

Ministry of Health DRoC (2009) Recensement, Bureau Central du Zone de Sante. In: Direction i, (editors), Kinshasa: Ministry of Health

Morens DM, Folkers GK, Fauci AS (2004) The challenge of emerging and re-emerging infectious diseases. *Nature* 430:242–249

Mossoun A, Pauly M, Akoua-Koffi C, Couacy-Hymann E, Leendertz SAJ, Anoh AE, et al. (2015) Contact to non-human primates and risk factors for zoonotic disease emergence in the Taï region. *Côte d'Ivoire. EcoHealth* 12(4):580–591

Murphy FA (1998) Emerging zoonoses. *Emerging Infectious Diseases* 4:429–435

Observatory for the Forests of Central Africa (2010) Congo basin forest partnership. The forests of the Congo basin: state of the forest. In: Luxembourg: Publications Office of the European Union. p. v

Organization WH (1996) Outbreak of Ebola haemorrhagic fever in Gabon officially declared over. *Wkly Epidemiol Rec* 71(17):125–126

Paige SB, Frost SD, Gibson MA, Jones JH, Shankar A, Switzer WM, et al. (2014) Beyond bushmeat: animal contact, injury, and zoonotic disease risk in Western Uganda. *EcoHealth* 11(4):534–543

Parker S, Nuara A, Buller RM, Schultz DA (2007) Human monkeypox: an emerging zoonotic disease. *Future Microbiol* 2(1):17–34

Peeters M, Courgnaud V, Abela B, Auzel P, Pourrut X, Bibollet-Ruche F, et al. (2002) Risk to human health from a plethora of simian immunodeficiency viruses in primate bushmeat. *Emerg Infect Dis* [serial on the Internet]

Poulsen J, Clark C, Mavah G, Elkan P (2009) Bushmeat supply and consumption in a tropical logging concession in northern Congo. *Conservation Biology* 23(6):1597–1608

Roess AA, Monroe BP, Kinzoni EA, Gallagher S (2011) Ibata SR, al e. Assessing the effectiveness of a community intervention for monkeypox prevention in the Congo Basin. *PLOS Neglected Tropical Diseases* 5(10):e1356

Switzer WM, Tang S, Ahuka-Mundeke S, Shankar A, Hanson DL, Zheng H, et al. (2012) Novel simian foamy virus infections from multiple monkey species in women from the Democratic Republic of Congo. *Retrovirology* 9:100

Taylor LH, Latham SM, Woolhouse ME (2001) Risk factors for human disease emergence. *Philos Trans R Soc Lond B Biol Sci* 356(1411):983–989

United Nations (2014) Department of Economic and Social Affairs., United Nations. Statistical Division. World Statistics Pocketbook. New York: United Nations

Wolfe ND, Prosser TA, Carr JK, Tamoufe U, Mpoudi-Ngole E, Torimiro JN, et al. (2004) Exposure to nonhuman primates in rural Cameroon. *Emerg Infect Dis* 10(12):2094–2099

Wolfe ND, Switzer WM, Carr JK, Bhullar VB, Shanmugam V, Tamoufe U, et al. (2004) Naturally acquired simian retrovirus infections in central African hunters. *The Lancet* 363(9413):932–937

Wolfe ND, Daszak P, Kilpatrick AM, Burke DS (2005) Bushmeat hunting, deforestation, and prediction of zoonotic disease. *Emerging Infectious Diseases* 11:1822–1827

Wolfe ND, Switzer WM, Carr JK, Bhullar VB, Shanmugam V, Tamoufe U, et al. (2005) Emergence of unique primate T-lymphotropic viruses among central African bushmeat hunters. *Proc Natl Acad Sci* 102:7994–7999

Wolfe ND, Dunavan CP, Diamond J (2007) Origins of major human infectious diseases. *Nature* 447(7142):279–283