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Ticks (Acari: Ixodidae) infesting transhumant cattle stalled in Kisangani (DR Congo): a neglected veterinary health issue

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Front cover: *Amblyomma variegatum* female before engorgement. © Anne Laudisoit.

Ticks (Acari: Ixodidae) infesting transhumant cattle stalled in Kisangani (DR Congo): a neglected veterinary health issue

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Abstract

We provide a first survey of hard ticks (Ixodidae) infesting transhumant cattle imported in Kisangani, Tshopo Province, Democratic Republic of Congo and discuss their potential implication in veterinarian health. Indeed, herds of cattle are being brought from the Central African Republic and Eastern regions to Kisangani after at least 550km journey across woodland savanna, lowland rainforest and urban centers. During this transhumance, cattle (*Bos taurus*) sleep outside potentially spreading imported tick species and are exposed to indigenous species. Between October and December 2018, we mostly collected Ixodidae on Mbororo cattle at the Mangobo slaughterhouse (Kisangani) prior to slaughtering. In total, 1,272 ticks were collected out of 83 infested bovines examined (96). The most abundant tick species were *Rhipicephalus (Boophilus) annulatus* (432; 33.9%), *Amblyomma cohaerens* (394; 30.9%) and *A. variegatum* (365; 28.7%). *Rh. (B.) decoloratus*, *Amblyomma sp.*, *Rh. appendiculatus* and *Rh. (Boophilus) sp.* represented less than 100 specimens. Tick infestation prevalence was not statistically different between bulls (89.7%) and cows (78.6%), nor between age groups; with infestation ranging between 86.2% in young cattle (<3yrs) and 87.1% in older cattle (>3yrs). Knowing that the etiology of mass cattle die-offs in the region remain mostly undetermined, the inventory of imported Ixodidae on transhumant cattle is a critical step prior to tick-borne agent molecular detection. These results call for the reinforcement of veterinary epidemio-surveillance in the country and the improvement of sanitary control policies in particular at ports of entry.

Keywords: *Amblyomma*, *Bos taurus*, bovine, DR Congo, *Rhipicephalus*, slaughterhouse, Veterinary medicine

Résumé

Nous avons étudié les tiques dures (Ixodidae) infestant le bétail transhumant, importés à Kisangani, en République Démocratique du Congo, et discutons de leur implication potentielle en santé vétérinaire. Des troupeaux de bétail sont acheminés vers Kisangani après une transhumance d'au moins 550 km traversant savane boisée, forêt pluviale de plaine et centres urbains. Durant ce voyage, le bétail (*Bos taurus*) dort à l'extérieur, propageant potentiellement

des tiques importées et les exposant à des tiques indigènes. Entre Octobre et Décembre 2018, nous avons collecté les Ixodidae principalement sur le bétail de Mbororo à l'abattoir de Mangobo avant leur abattage. Au total, 1272 tiques ont été collectées sur 83 bovins infestés examinés (96). Les espèces les plus abondantes étaient *Rhipicephalus (Boophilus) annulatus* (432; 33,9%), *Amblyomma cohaerens* (394; 30,9%), et *A. variegatum* (365; 28,7%) tandis que *Rh. (B.) decoloratus*, *Amblyomma sp.*, *Rh. appendiculatus*, et *Rh. (Boophilus) sp.* représentaient moins de 100 spécimens. L'infestation n'était pas statistiquement différente entre taureaux (89,7 %) et vaches (78,6 %), ni entre groupes d'âge, variant entre 86,2 % chez les jeunes bovins (<3 ans) et 87,1 % chez les bovins plus âgés (>3 ans). Sachant que l'étiologie des mortalités massives de bovins dans la région reste indéterminée, l'inventaire des Ixodidae importés sur les bovins transhumants est une étape critique avant la détection moléculaire des agents transmis par les tiques. Ces résultats appellent au renforcement de l'épidémio-surveillance vétérinaire dans le pays et à l'amélioration des politiques de contrôle sanitaire, en particulier aux points d'entrée.

Introduction

Emerging tick-borne pathogens pose a growing global health risk to both human (e.g. Lyme disease, Congo-Crimean Hemorrhagic Fever, tick-borne encephalitis) and animal (e.g. theileriosis, cowdriosis, anaplasmosis) populations besides causing important economic losses (HOUNZANGBE-ADOTE *et al.*, 2001; PAROLA & RAOULT, 2001; BOWMAN & NUTTALL, 2008; BOURNEZ, 2014; SONENSHINE & ROE, 2014). Ticks are obligate blood sucking arthropods that infest most vertebrates particularly mammals (including humans), birds and reptiles (BOWMAN & NUTTALL, 2008). So far, 989 tick species have been identified worldwide of which the hard ticks represent the majority with about 703 species (RENE, 2013; DIARRA *et al.*, 2017). In tropical countries, tick infestation represents a major constraint to the development of livestock farming, not only through the diseases they transmit, but also through collateral adverse effect (e.g. blood spoliation, lesions, bacterial superinfections) which impact the general health of the cattle (BOURNEZ, 2014). Global economic losses due to tick-borne diseases (TBDs) are estimated to be around USD 20-30 billion annually worldwide (JONGEJAN & UILENBERG, 2004; LEW-TABOR & VALLE, 2016) and the cost of theileriosis (*Theileria parva*) alone is estimated at 168 million US\$ annually in Africa (MUKHEBI *et al.*, 1992). About 80% of the world's cattle is infested with ticks. As such, they represent the most economically important ectoparasites of livestock which contribute to poor productivity and poverty among breeders (BOWMAN & NUTTALL, 2008; ABERA *et al.*, 2010). In developing countries where the livestock sector represents a significant share of the local economy, tick-borne agents commonly affect cattle in agropastoral regions (EYO *et al.*, 2014; SASSA *et al.*, 2014; DIARRA *et al.*, 2017). In the Democratic Republic of Congo (DR Congo), several cattle breeding sectors are affected by TBDs (anaplasmosis, babesiosis and theileriosis) (LESSARD *et al.*, 1990; NORVAL *et al.*, 1992) however there is a lack of centralized data and systematic reporting. Despite the high impact of TBDs on the economy, the Congolese State has been loosening its epidemio-surveillance of major enzootics for more than 40 years, so that information on the prevalence of animal diseases and their economic importance is scarce. In eastern DR Congo, it is assumed that TBDs commonly affect cattle but remain undiagnosed and hence unreported in most cases (KALUME, 2012; BISUSA *et al.*, 2014, 2016; BISUSA & KUJIRABWINJA, 2016). Similarly, the distribution range of many tick vectors is either not available or are outdated (KALUME, 2012; SILATSA *et al.*, 2019). The few authors who performed studies on tick infesting cattle in the DR Congo found a high prevalence of *Amblyomma variegatum*, *A. cohaerens*, *Rhipicephalus (Boophilus) decoloratus*, *Rh. compositus*, *Rh. appendiculatus* and *Haemaphysalis leachi* (KALUME, 2012; BISUSA *et al.*, 2014, 2016; BISUSA & KUJIRABWINJA, 2016); species known to transmit

veterinary important pathogenic agents (BOWMAN & NUTTALL, 2008). *Amblyomma variegatum* is a three host tick species known for its successful colonization of African islands through cattle trade and as major vector of heartwater disease, *Ehrlichia ruminantium* and their association with dermatophilosis, *Dermatophilus congolensis* (BARRE & UILENBERG, 2010). Moreover, in 2013, during African Swine Fever outbreak investigations, a new tick species (*Rhipicephalus congolensis*) infesting red river hogs (*Potamochoerus porcus*) and domestic pigs (*Sus domesticus*) was discovered from the DR Congo (APANASKEVICH *et al.*, 2013); it has now been found in Kisangani, on domestic pigs including on pigs imported from other provinces (NGOY, 2019). However, due to poor veterinary control measures in rural areas, most livestock mortality remain undiagnosed and the topic neglected. Such is even more the case in non-agropastoral and forested regions like the Tshopo province in general, and the city of Kisangani in particular – the third largest urbanized city in the country and the largest city lying in the lowland tropical rainforest of DR Congo. As a result, there are no recent data on cattle ticks – and even less on the pathogens they carry, despite multiple transhumances routes and intensive transregional and transboundary trade. Indeed, several studies have shown that the phenomenon of cattle transhumance is an important factor which favors the spread of exotic tick species and their pathogens into many countries (OLWOCH *et al.*, 2008; KALUME, 2012; ADINCI *et al.*, 2018; SILATSA *et al.*, 2019). Thus, we are witnessing the extension of the range of tick species and consequently the probable infestation of the local fauna (ADINCI *et al.*, 2018).

Mbororo are nomadic pastoralists (subgroup of the Fulani) people encountered in Africa (UILENBERG *et al.*, 2012). Mbororo herders originate from the Central African Republic (CAR), Sudan and Chad and migrated with their cattle south into DR Congo beginning in the 1990s, and established in northern DR Congo in the early 2000s. Due to the environmental changes, including drought, they are driven increasingly to the south of the savannah regions (UILENBERG *et al.*, 2012; MBUYI, 2018); as such Mbororo herders are considered by ecologist as climatic refugees. They rear two major breeds of cattle, namely Hogo and CAR which they have been selling in Kisangani for several decades. Consequently, it is likely that the importation of foreign transhumant cattle may introduce blood-borne and tick-borne agents along their migration to the city of Kisangani.

In this context, we carried out a first inventory of hard ticks (Ixodidae) infesting transhumant Mbororo herders' cattle brought to the Mangobo slaughterhouse of Kisangani, Tshopo province, DR Congo. We carried out this exploratory study to (1) inventory tick species infesting imported cattle prior to slaughtering, (2) determine tick index (prevalence of infestation) of imported cattle (per age and gender), and (3) discuss their potential implications for veterinarian health.

Material and methods

TICK COLLECTION

The tick (Ixodidae) were manually collected on the cattle stalled at the slaughterhouse of Mangobo (N00°32. 75', E25°10.23'), Kisangani city (Fig. 1), Tshopo province, DR Congo. The national slaughterhouse of Mangobo was created in 1956, is built of durable materials, and consists of two sheds and two restraint iron corridors where animals are directed to be slaughtered; patches of grass (*Paspalum notatum*) grow in the premises. The slaughterhouse is mostly known for the slaughter of cows; other livestock (pigs, donkeys, goats and sheep) is seldom slaughtered.

The survey was carried out between October 22nd and December 1st 2018 which corresponds to the major rainy season (September to November) and the major dry season (December to February). In 24 visits (4 times/week; 6 weeks), we visually inspected 96 *Bos taurus* (68 bulls

and 28 cows) belonging to the local breed namely Ituri and Bunia and the imported breed Hogo and CAR. Most cattle examined belonged to Mbororo herders. The journey of the transhumant Mbororo cattle examined lasts on average 1 month before reaching Kisangani (walking a minimum of 550 km). Upon arrival, herders keep their cattle either in pasturing fenced plots, in a pen, tied to a pole or let free-grazing. Some cattle are placed in a pen next to the house during the night for an average 5 days to recover from the journey before being sold. Once within the compound of the slaughterhouse, the cattle are usually kept between 1 or 2 days before being slaughtered.

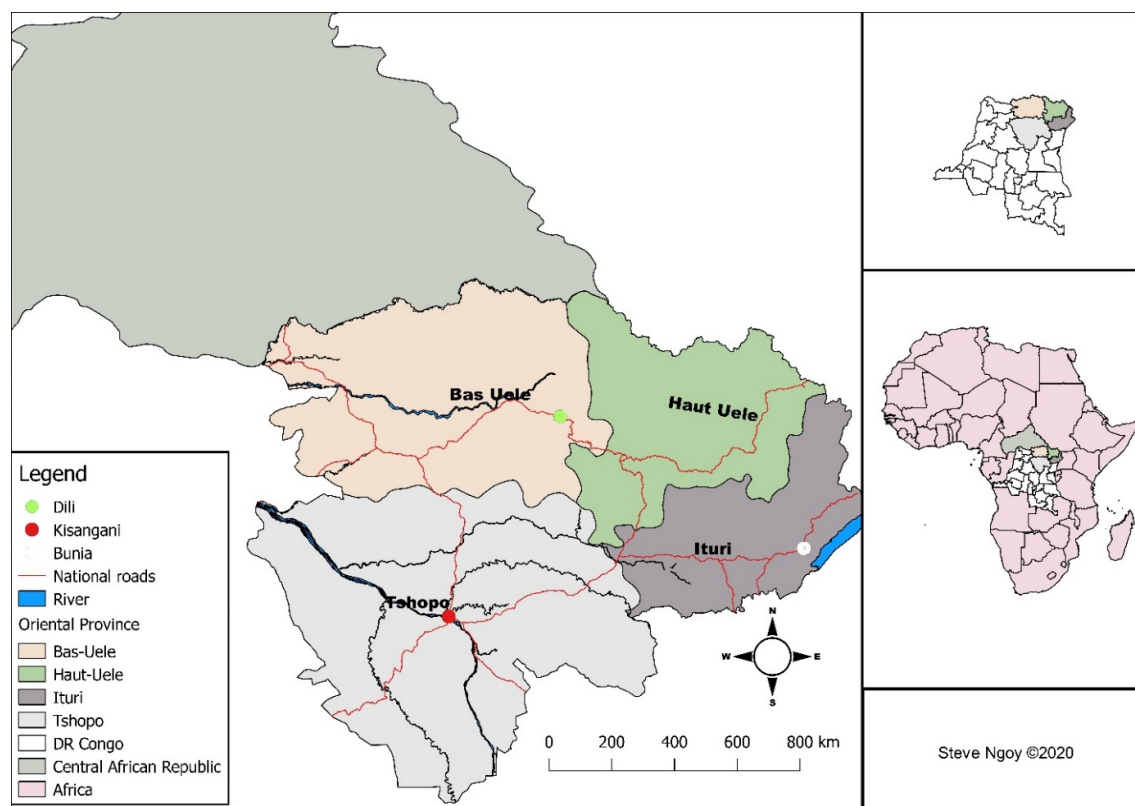


Fig. 1. Map of Kisangani and Major transhumance routes from probable departure location to Kisangani city, DR Congo.

The examined cattle were categorized into two age groups these are ≤ 3 years and > 3 years. All visible ticks were removed manually from infested bovines recently slaughtered. The body regions (abdomen, armpit, back, flank, udder, limbs, anal region, inguinal region, head and tail) on which they were found were noted according to SHICHIBI *et al.* (2017) and KEMAL *et al.* (2016). The age, sex, breed and origin of each bovine examined were also noted. After their collection, ticks were placed in labeled bags, transported to the laboratory, and transferred to labelled 2 ml vials containing 70% alcohol. We proceeded to tick morpho-anatomical identifications under a Leica EZ4 binocular magnifier using keys by WALKER *et al.* (2003), OKELLO-ONEN *et al.* (1999) and MATTHYSSE & COLBO (1987). Tick specimens were identified from the genus to the species level; 5 specimens were only identified to the genus. All specimens associated with their unique codes have been sent to the Royal Belgian Institute of Natural Sciences (RBINS) research collection after their morphological identification.

PREVALENCE INDEX AND INFESTATION LOAD

We calculated the prevalence (P) of infested cattle as the ratio of the number of infested hosts (H^{in}) to the number of examined hosts (H^{ex}), expressed as follows: $P\ (%) = (H^{in}/H^{ex}) * 100$ (LAAMRI *et al.*, 2012). We calculated the relative prevalence (PRt) of tick species as the proportion of tick of each species (nS^i) – or genera – on the total number of ticks collected (N): $PRt(\%) = (nS^i / N) * 100$ (TADDESE & MUSTEFA, 2013).

Finally, in order to assess the infestation burden of each tick species, we calculated a specific tick index (S^iT) expressed as the ratio of the total number of individuals of a given tick species (nS^i) found on the examined cattle population divided by the number of tick infested cattle (H^{in}): $S^iT = nS^i / H^{in}$.

STATISTICAL DATA ANALYSIS

The collected data was grouped per cattle age class, sex, body region and the developmental instars of the ticks (adults, nymphs and larvae). We used the software PAST (PAleontological STatistics) version 3.02 for all statistical analysis with the significance threshold level set to $\alpha=0.05$. We used the χ^2 test to assess differences in tick prevalence of infestation between age classes and tick species.

Results

Of 96 examined cattle (68 bulls and 28 cows), 83 (86.5%) were found to be infested by one or more Ixodid tick. The majority of bovine slaughtered at the Mangobo slaughterhouse during the survey period (October to December) came from CAR 83 (86.5%) with 88.0% of them infested with at least one Ixodid tick (Table 1); others came from Bunia (Ituri province) 6 (6.3%), and Dili (Bas-Uélé province) 7 (7.3%) in the DR Congo. Tick infestation on bulls and cows was not significantly different ($P=0.69$; $\chi^2=0.16$), nor did we observe a difference between age classes ($P = 0.973$; $\chi^2 = 0.001$) (Table 2).

Table 1. Origin and prevalence of infestation of bovine slaughtered at the Mangobo abattoir (Kisangani, Tshopo province, DR Congo) during the study period (October – December 2018).

Origin of cattle	Observed	Infested	Prevalence (%)
Bunia	6 (6.3)	4 (4.8)	66.7
Dili	7 (7.2)	6 (7.2)	85.7
CAR	83 (86.5)	73 (88.0)	88.0
Total	96	83	86.5

We collected a total of 1,272 hard ticks of which 936 adult ticks (73.5%), 329 nymphs (25.8%) and 7 larvae (0.6%) making up the sample examined (Table 3). These Ixodid ticks belong to a single family, the Amblyommidae, two genera and at least seven species. The most abundant tick species were the East African buffalo tick *Amblyomma cohaerens* Donitz 1909, the tropical bont tick *A. variegatum* Fabricius, 1794 and the cattle tick *Rhipicephalus (Boophilus) annulatus* Neumann, 1904 (Fig. 2). Other less abundant species collected are the African blue tick *Rh. (Boophilus) decoloratus* Koch, 1844, the brown ear tick *Rh. appendiculatus* Neumann, 1901 and unidentified species in the genera *Amblyomma* sp. and *Rh. (Boophilus)* sp. (Table 3). *Rh. (B.) annulatus* was the most prevalent species with the highest tick index followed by *A. cohaerens* and *A. variegatum*. Attachment sites significantly differed between tick species with armpits selection by *A. variegatum* ($\chi^2=67.33$, $P=0.001$; 95% CI: 25.06–47.94), the

Table 2. Prevalence of tick infestation based on sex and age of host sampled at the Mangobo abattoir (Kisangani, Tshopo province, DR Congo) during the study period (October – December 2018).

Parameters	Sampled	Infestation (%)	Chi-square	P-value
Sex group				
Bull	68	61 (89.7)	0.16	0.69
Cow	28	22 (78.6)		
Age groups				
≤ 3 years	65	56 (86.2%)	0.001	0.973
> 3 years	31	27 (87.1%)		
Total	96	83 (86.5%)		

inguinal region by *A. cohaerens* ($\chi^2=140.58$, $P=0.001$; 95% CI: 38.38–40.42), and the abdomen and inguinal region for both *Rh. (B.) annulatus* ($\chi^2=74.08$, $P=0.001$; 95% CI: 42.58–43.82) and *Rh. (B.) decoloratus* ($\chi^2=27.39$, $P=0.001$; 95% CI: 7.76–8.46) (Table 4). We could not determine site preference for *Amblyomma* sp., *Rh. (Boophilus)* sp. and *Rh. appendiculatus* as for these taxa the sample size was insufficient.

Table 3. Species of ticks collected from slaughtered cattle at the Mangobo abattoir (Kisangani, Tshopo province, DR Congo) by sex, developmental stage, relative prevalence and specific tick index during the study period (October – December 2018).

Tick species	Ad.♂	Ad.♀	Ny.	La	Total	PRt	S ^t T
<i>A. cohaerens</i>	190	104	99	1	394	0.3	0.05
<i>A. variegatum</i>	135	57	168	5	365	30. 9	4.75
<i>Amblyomma</i> sp.	-	-	3	1	4	28. 7	4.40
<i>Rh. (B.) annulatus</i>	27	350	55	-	432	33. 9	5.20
<i>Rh. (B.) decoloratus</i>	51	18	4	-	73	5.7	0.88
<i>Rh. (Boophilus.) sp.</i>	1	-	-	-	1	0.1	0.01
<i>Rh. appendiculatus</i>	1	2	-	-	3	0.2	0.04
Total	405	531	329	7	1272		

Discussion

This original study is one of the few carried out on ticks in the DR Congo, and the only one addressing the transhumant cattle veterinary importance in Kisangani. Although this study was carried out over a short time span (6 weeks) with limited sample size of cattle (N=96), it provides preliminary information on the hard ticks (Ixodidae) infesting cattle mostly sold by Mbororo herders in a slaughterhouse of Kisangani, DR Congo. The low number of cattle examined reflects the low number of cattle brought for slaughtering on the premises. Indeed, some herders prefer to slaughter their cattle in the forest or in makeshift slaughterhouses to avoid paying taxes and escape veterinary inspection, an attitude which poses a serious veterinary and public health risk.

Table 4. Attachment site of ticks on the cattle examined and the number of tick species per attachment site

	Abd	Arp	B	Flk	Udr	Lmb	Ar	IngR	Hd	T		
Tick species	N (%)	N (%)	N (%)	N (%)	N (%)	N (%)	N (%)	N (%)	N (%)	N (%)	χ^2	p-value
<i>A. variegatum</i>	36 (2.8)	77 (6.1)	11 (0.9)	19 (1.5)	40 (3.1)	42 (3.3)	33 (2.6)	53 (4.2)	52 (4.1)	2 (0.2)	67. 3	0.000 1
<i>A. cohaerens</i>	18 (1.4)	28 (2.2)	9 (0.7)	4 (0.3)	82 (6.5)	43 (3.4)	132 (10.4)	29 (2.3)	25 (1.9)	24 (1.9)	14 0.6	0.000 1
<i>Amblyomma</i> sp.	-	1 (0.8)	-	-	-	2 (0.2)	-	1 (0.8)	-	-	8.9	0.45
<i>Rh. (B.) annulatus</i>	79 (6.2)	59 (4.6)	10 (0.8)	44 (3.5)	46 (3.6)	48 (3.8)	36 (2.8)	78 (6.1)	12 (0.9)	20 (1.6)	74. 1	0.000 1
<i>Rh. (B.) decoloratus</i>	6 (0.5)	11 (0.9)	4 (0.3)	6 (0.5)	10 (0.8)	1 (0.1)	9 (0.7)	20 (1.2)	-	6 (0.5)	27. 4	0.001
<i>Rh. (Boophilus)</i> sp.	-	-	-	-	-	-	1 (0.1)	-	-	-	-	-
<i>Rh. appendiculatus</i>	-	1 (0.1)	-	-	-	-	1 (0.1)	-	1 (0.1)	-	-	-
Total	139 (10.9)	177 (13.9)	34 (2.6)	73 (5.7)	178 (13.9)	136 (10.7)	212 (16.7)	181 (14.2)	90 (7.1)	52 (4.1)		

Legend:

Abd: Abdomen; Arp: Armpit; B: Back; Flk: Flank; Udr: Udder; Lmb: Limbs; AnR: anal region; IngR: Inguinal region; Hd: Head and T: tail

N: number

 χ^2 : chi-square

All Ixodidae collected belonged to a single family (Amblyommidae), two genera and seven species with decreasing number *Rh. (B.) annulatus*, *A. cohaerens*, *A. variegatum* (Fig. 3), *Rh. (B.) decoloratus*, *Rh. appendiculatus* and two unidentified species namely *Amblyomma* sp. and *Rh. (Boophilus)* sp. The low number of larvae recorded could be due to the method of visual inspection used and their small size (François, 2008). It could also relate to the seasonality and species-specific ecological preferences of pre-imaginal stages of multi-host ticks in the landscapes crossed by the transhumant cattle (mostly lowland rainforest for up to 600 km before reaching Kisangani). We mainly sampled during the great rainy season (September to November) and the major dry season (December to February) or on cattle that has moved during the rains, and ended at the beginning of the dry season (December). The studies of KALUME *et al.* (2013) and BISUSA *et al.* (2014) have also shown that *Rh. (B.) decoloratus*, *A. variegatum* and *Rh. appendiculatus* are common tick species collected on cattle in Nord-Kivu, living in pasture, and sampled during the short rainy season (February–April).



Fig. 2. A picture of four male - female pairs of *Amblyomma variegatum* attached on a cow examined during this survey. © Anne Laudisoit.

The male to female sex ratio in *A. variegatum*, *A. cohaerens* and *Rh. (B.) decoloratus* showed that males were present in greater number than females, except for *Rh. (B.) annulatus*. These results are consistent with previous reports DIOLI *et al.* (2001), TAMIRU *et al.* (2010), KASSA & YALEW (2012) and SILATSA *et al.* (2019). This either suggests a seasonal component (e.g. males remain on the host for an extended period of time; females drop off to lay eggs) or a behavioral component (e.g. females of *A. variegatum* generally do not attach to the hosts until males have been present for at least three days) (BARRE & GARRIS, 1990).

The massive infestation of the Mbororos' cattle by *Rh. (B.) annulatus* and *A. variegatum* is expected as both species are well established in Africa and are widespread nuisance of cattle just as *Rh. (B.) microplus*, *Rh. appendiculatus* and *Rh. (B.) decoloratus* (WALKER & OLWAGE, 1987; WALKER *et al.*, 2003; MADDER *et al.*, 2014; D'AMICO *et al.*, 2017). *A. variegatum* and *A. cohaerens* are three-host ticks which opportunistic immature stages feed on reptiles, mammals and birds (BARRE, 1989; GOMES, 1993; WALKER *et al.*, 2003; FRANÇOIS, 2008) abundant in the habitats crossed by the cattle. On the other hand, *Rh. (B.) annulatus* is a one-host tick that infest cattle with ecological preference for humid areas with important annual

rainfall (800–5000 mm), and luxuriant vegetation throughout the year (OKELLO-ONEN *et al.*, 1999; KALUME *et al.*, 2013); conditions prevailing in the transhumant cattle catchment area.

We found that the majority of the cattle slaughtered in Mangobo are sold by Mbororo herders coming from CAR (N=83; 86.5%) and that a large proportion (88.0%) was infested with at least one Ixodid tick. Some imported cattle were subjected to acaricide treatment upon departure according to the declarations of 13 herders; the use of antibiotic and anthelmintic treatments was also mentioned. Although it is possible that cattle infected by tick-borne blood pathogens cross the border while being free of ectoparasites, some ticks may survive acaricide treatment due to acaricide resistance (MADDER *et al.*, 2011). In any case, during their one-month journey across different agro-ecological zones to the slaughterhouse (at least 550 km), the transhumant cattle are exposed to ticks and become infested *de novo* (AYALEW *et al.*, 2013). They may thus introduce pathogenic agents they carry in the local tick populations as observed elsewhere in the country and in Africa in general (MADDER *et al.*, 2011; KALUME *et al.*, 2013) and transfer ticks regionally. The introduction of cattle (heavily infested by ticks) from CAR to DR Congo could potentially lead to the introduction of ticks and TBDs as observed with cattle imported from CAR to Cameroon by nomadic pastoralists (ADINCI *et al.*, 2018; SILATSA *et al.*, 2019).



Fig. 3. *Amblyomma variegatum* female before engorgement

An unknown proportion of the recorded ticks may be acquired while the cattle were allowed to recover near Kisangani from their journey (6 to 7 days prior to slaughtering). Indeed, the short time spent feeding on the host – a maximum of 15 days for each stage of development (YONOW, 1995) – the presence of larvae, and the equal number of nymphs and adults of *A. variegatum* on the cattle examined suggest recent infestation events. The majority of *Rh. (B.) decoloratus* and *Rh. (B.) annulatus* – one host tick species – were adults and as such it is difficult to infer the proportion imported and the proportion acquired along the transhumance.

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The exhaustion, emaciation and lowered immunity of the transhumant cattle may enhance the vulnerability to multiple tick infestation and tick-borne pathogens (BYAVU *et al.*, 2000; MULUGETA *et al.*, 2010; KALUME *et al.*, 2013). We found that 46.9% of the cattle were co-infested with 3 ticks species offering increased opportunities to exchange blood-borne pathogens (e.g. pool-feeding).

The species inventoried in the present study are known to carry pathogenic agents. Indeed, *Rh. (B.) annulatus* is known to transmit several apicomplexa (*Babesia bigemina*, *B. bovis*, *Anaplasma marginale*) to cattle responsible of unassessed morbidities and mortalities in the DR Congo and in other African countries (OKELLO-ONEN *et al.*, 1999; WALKER *et al.*, 2003; KALUME, 2012; MADDER *et al.*, 2014; D'AMICO *et al.*, 2017). Comparatively to *A. cohaerens*, *A. variegatum* has a great economic importance. It is an efficient vector of *Ehrlichia ruminantium*, *Theileria mutans*, *T. velifera*, *E. bovis*, *Dermatophilus congolensis*, causing respectively heartwater disease or cowdriose, benign bovine theilerioses, bovine ehrlichiosis, bovine dermatophilosis and the virus of Nairobi sheep disease (WALKER *et al.*, 2003). Moreover, due to its long mouth parts, it causes important damage to the cowhide, thus reducing its market value (WALKER & OLWAGE, 1987; OKELLO-ONEN *et al.*, 1999; SOLOMON *et al.*, 2001; MADDER *et al.*, 2014).

The predilection sites of the tick species recorded on cattle are consistent with earlier reports from Africa (MUCHENJE *et al.*, 2008; ABDELA, 2016; ALEMU *et al.*, 2014; BISUSA *et al.* 2014; SHICHIBI *et al.*, 2017). These have shown that tick attachment site selection depends on site temperature, humidity, presence of thin skin and ample veins and on morphological features (e.g. mouthparts or hypostome length) (STACHURSKI, 2000).

The cross-border movement of cattle may lead to the introduction of exotic tick species but also blood borne and tick-borne pathogens into a new region where it can cause enormous economic losses and public and animal health burden (BARRE, 1989; OLWOCH *et al.*, 2008; BARRE & UILENBERG, 2010; KALUME, 2012; SILATSA *et al.*, 2019). Our study highlights the potential risk of introduction of ticks and tick-borne pathogens through the transhumant cattle trade in DR

Congo and Kisangani. This study has limitations as we cannot establish the exact origin of the ticks collected, and as none of the cattle examined nor the tick collected were subjected to molecular screening for tick borne agents. However, it brings attention to a neglected topic namely the absence of systematic cattle trade data, veterinary inspection and epidemio-surveillance in a region crisscrossed by major transborder roads (CAR, South-Sudan and Uganda). This is largely due to the vast area to cover, and the unsystematic border inspection due to the high number of official and unofficial land or river entry/exit points which encourages illegal movements. Moreover, the etiology of mass livestock die-offs in the region remains largely undetermined and their economic impact underestimated or not assessed. As such, we call for reinforcing regional capacity building of veterinarians and para-veterinarians and the establishment of a structured epidemio-surveillance network so as to improve epizootic preparedness and response and inform effective veterinary health and control policies.

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