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Prevalence of ticks infesting grasscutters (*Thryonomys swinderianus* Temminck, 1827) in the south of Côte d'Ivoire

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ABSTRACT

Objective: Ticks play a significant role in the transmission of pathogenic agents to animals. In Côte d'Ivoire, there is little information on those of grasscutters.

Methodology and results: Thus, 150 wild grasscutters and 150 farm grasscutters from the south of Côte d'Ivoire were examined from April 2010 to October 2012. Ticks collected were identified under binocular lens and optical microscope. The species identified were confirmed at CIRDES (Bobo-Dioulasso). The species *Ixodes aulacodi*, *Rhipicephalus simpsoni*, *Rhipicephalus* (*Boophilus*) *microplus*, *Amblyomma compressum* and *Haemaphysalis leachi* were identified. Farm grasscutters were not infested but 92% of wild grasscutters were infested by at least one species. The most abundant species was *I. aulacodi*. It accounted for 67.98% of all collected ticks and was encountered on 75.33% of wild grasscutters. The second one was *R. simpsoni* (29.11% of collected ticks and collected on 54% of wild grasscutters). The three other species were slightly encountered (less than 2% of collected ticks and prevalence lower than 7%). Similarly, the infestation level by *I. aulacodi* was the highest (7±5 ticks per grasscutter against less than 4 for the others).Ticks belonging to *R. simpsoni*, *R. microplus* and *Ha. leachi* were adults. On the other hand, 9.16% of *I. aulacodi* and 25% of *A. compressum* were nymphs.

Conclusion and application of results: Farm grasscutters in Côte d'Ivoire do not carry some ticks. But five species were found on wild grasscutters. Ticks and pathogenic agents that they transmit and for which some are responsible for zoonosis, could constitute a major obstacle for the development of grasscutters' farming and constitute a problem of public health. This study gives an update on diversity and database information for surveillance and prevention of zoonosis and their vectors.

Keywords: grasscutters, Ticks, Prevalence, Côte d'Ivoire.

RESUME

Objectif: Les tiques jouent un rôle très important dans la transmission d'agents pathogènes aux animaux. En Côte d'Ivoire, il existe peu d'information sur celles rencontrées chez les aulacodes.

Méthodologie et résultats: Ainsi, 150 aulacodes sauvages et 150 aulacodes d'élevage provenant du sud de ce pays ont été examinés d'avril 2010 à octobre 2012. Les tiques récoltées ont été identifiées à la loupe binoculaire et au microscope optique. L'identification a été confirmée au CIRDES (Bobo-Dioulasso). Les espèces *Ixodes aulacodi*, *Rhipicephalus simpsoni*, *Rhipicephalus* (*Boophilus*) *microplus*, *Amblyomma compressum* et *Haemaphysalis leachi* ont été identifiées. Aucun aulacode d'élevage n'en portait. Par contre 92% des aulacodes sauvages étaient infestés par au moins une espèce. L'espèce la plus abondante était *I. aulacodi* (67,98% de l'effectif total des tiques récoltées et rencontrée sur 75,33% des aulacodes sauvages). La deuxième espèce était *R. simpsoni* (29,11% de l'effectif total et récoltée sur 54% des aulacodes sauvages). Les autres espèces ont été faiblement rencontrées (moins de 2% de l'effectif total et prévalences inférieures à 7%). De même, le degré d'infestation par *I. aulacodi* était plus élevé (7±5 tiques par aulacode contre 4 pour les autres espèces). Toutes les tiques des espèces *R. simpsoni*, *R. microplus* et *Ha. leachi* étaient adultes. Par contre, 9,19% des *I. aulacodi* et 25% des *A. compressum* étaient des nymphes.

Conclusion et application des résultats: Les aulacodes d'élevage en Côte d'Ivoire ne portent pas de tiques. Cependant, 5 espèces ont été rencontrées sur les aulacodes sauvages. Les tiques et les agents pathogènes qu'elles transmettent et dont certains sont responsables de zoonoses, pourraient constituer un obstacle majeur au développement de l'aulacodiculture et constituer un problème de santé publique. Cette étude représente donc une base de l'information pour la surveillance et la prévention de zoonoses et leurs vecteurs.

Mots-clés : Aulacodes, Tiques, Prévalence, Côte d'Ivoire.

INTRODUCTION

In order to cater for animal proteins requirements, Côte d'Ivoire has put in place semi-intensive farming of animals with short biological cycle (poultry, pigs, rabbit). Local breeds of small ruminants and fish farming have been improved (Fantodji & Mensah, 2000). Then, farming in captivity of certain species of wild animals such as giant snails and grasscutters has been initiated. Since 1995, grasscutters farming, one of the most important bushmeat in West Africa (Abé, 2009), shows a considerable development in Côte d'Ivoire. According to Soro (2007), grasscutter represents around 60% of bushmeat consumption in Côte d'Ivoire. The current level of exploitation could lead to the decrease of the whole natural stock of grasscutter. To prevent this decrease and to reduce animals proteins imports, research has been carried out on grasscutters growth, reproduction and feeding (Soro, 2007). Research has been made done on the pathology of this animal and allowed the identification of numerous parasites namely: protozoans (Arène, 1986), gastro-intestinal parasites (Omonona, 2011 ; Zouh Bi, 2013), blood parasites (Opara & Fagbemi 2010) and ectoparasites mainly represented by ticks (Yeboah & Simpson 2004 ; Abé, 2009). As the second most important diseases vectors around the world, after mosquitoes (Parola & Didier 2001), ticks could transmit zoonotical parasites which could cause a problem of public health. So far, no investigation has been conducted in Côte d'Ivoire in order to establish their prevalence in grasscutters. Therefore, it could be useful to carry out a study on ticks infesting grascutters in Côte d'Ivoire. It is the target of this study, which aims at identifying and determining abundance and prevalence of ticks on wild and farm grascutters.

MATERIAL AND METHODS

Study areas: Côte d'Ivoire, a country located in the northern hemisphere in the humid and coastal zone of west Africa, is between the tropic of Cancer and the Equator, precisely between 4th and 10th degree of latitude north, and 2nd and 8th degree of longitude west. The study has been carried out on grasscutters selected from eight regions of the south : Districts of Abidjan and Yamoussoukro, regions of Agnéby-Tiassa, la Mé, Grands Ponts, Lôh Djiboua, Sud Comoé and Belier located in forest zone with high rainfall. These regions have been chosen because of many existing grasscutters' farms.

Animals: The study was conducted on 150 farm grasscutters from fifteen farms. One hundred and fifty (150) wild grasscutters were also submitted for investigations and were from the regions quoted above.

Tick collection: Tick collection was done from April 2010 to October 2012. The fur was explored with care and all the ticks found were collected with a pair of tongs preventing the hypostom from breaking. The ticks collected on a grasscutter were conserved in a bottle with a hermetic latch containing Ethanol 70%. Each bottle was labelled with the following : ordinal number of the sample, farm where sample was collected (name of the District or the region), date of collection, sex of the host.

Tick identification *:* Tick identification has been performed at the Veterinary Central Laboratory of Bingerville (LCVB). The content of each bottle was poured into a kneaded box, and then the parasites were separated and counted by the ornamentation of the body, by the sex and by the physiological stage. The species identification was completed under binocular lens (CETI) at 80-fold magnification and optical microscope (Leica DMLS) at 100-fold magnification. It bases on the morpho-anatomic caracteristics (Arthur, 1956; Aeschlimann, 1963; Morel & Mouchet 1965; Walker *et al*., 2003). Indeed, the female of the species *Ixodes aulacodi* Arthur, 1956 were identified by the presence of fine long hairs on the alloscutum, triangular form of basis capituli, genital opening between coxae IV and anal valves large and eccentrically placed. For the male, the scutum punctuation and genital aperture position between coxae III was searched for. Regarding the species *Rhipicephalus simpsoni* Nuttall, 1910*,* characteristics of the female observed were long scutum without interstitial punctuations and little superficial punctuations. The male were discriminated by the sickle form of adanal plates. Concerning *Rhipicephalus* (*Boophilus*) *microplus* Canestrini, 1888, we discriminated with microscope at 100-fold magnification. We highlighted on layout of dentition, the existence of ventro-internal protuberance bearing setae near the roster. As far as concerning *Amblyomma compressum* Macalister, 1872, there is no ornamentation on the scutum. The marginal furrow is short but distinct for the male and absent for the female. Teeth are distributed in three pairs of files on each hypostom and the spur of coxae IV for the male is particularly robust. The species *Haemaphysalis leachi* Audouin, 1826 has conspicuous lateral extensions to palp articles 2, forming mouthparts with a distinctive conical shape. In addition, festoons have been observed and counted. All ticks were convoyed in the acarology section of CIRDES in Bobo-Dioulasso, for confirmation. Indeed, confirmation was done morphologically and the species *Rhipicephalus* (*Boophilus*) *microplus* was confirmed by molecular biology using a PCR–RFLP test (Lempereur *et al*., 2010 ; De Clercq *et al*., 2012).

Results expressing and statistical analysis: Microsoft Office Excel 2007 program was used to print data, to calculate abundance, prevalence and infestation level of each tick species. Formulas used :

Abundance of each tick species (A):

$$A = \frac{\text{Total number of a given tick species}}{\text{Total number of ticks collected}} \times 100$$

Prevalence of each tick species on grasscutters (P):

$$P = \frac{\text{Number of grasscutters carrying a given tick species}}{\text{Total number of grasscutters}} \times 100$$

Infestation level of each tick species (I) :

$$I = \frac{\text{Total number of a given tick species}}{\text{Total number of grasscutters carrying this tick species}}$$

The statistical comparisons of those abundances, prevalence and infestation levels have been done respectively by Chi-square test and Student t test. Difference was significant when p value was lower than 0.05 ($p < 0.05$).

RESULTS

A sample of 1065 ticks was considered in wild grasscutters. Ninety two per cent (92%) of wild grasscutters were carying at least one tick species. On the other side, none of the farming grasscutters examined was carying a single tick. The identification mainly based on morpho-anatomical characters has revealed the presence of five species of ticks from five genera on wild grasscutters: *Ixodes aulacodi* (Figure 1), *Rhipicephalus simpsoni* (Figure 2), *Amblyomma compressum* (Figure 3), *Rhipicephalus* (*Boophilus*) *microplus* (Figure 4) and *Haemaphysalis leachi* (Figure 5).

Figure 1 : *Ixodes aulacodi* (male)

Figure 2: *Rhipicephalus simpsoni* (female)

Figure 3 : *Amblyomma compressum* (male)

Figure 4: *Rhipicephalus* (*Boophilus*) *microplus* (male)

Figure 5: *Haemaphysalis leachi* (female)

Those ticks were collected on grasscutters from all the study areas. Indeed, the Autonomous Districts of Abidjan, the regions of Agnéby-Tiassa, la Mé, and Bélier sheltered four of the five tick species identified. It was about *I. aulacodi*, *R. simpsoni*, *A. compressum* and *R. microplus*. Concerning the Autonomous District of Yamoussoukro, three species that were *I. aulacodi*, *R. simpsoni* and *A. compressum* were met whereas grasscutters from Lôh Djiboua region lodged *I. aulacodi*, *R. simpsoni* and *Ha. leachi*. At last, only two species namely *I. aulacodi* and *R. simpsoni,* were collected on grasscutters from Grands Ponts and Sud Comoé regions.

- ***Abundance and prevalence of the different species identified:*** Among all the ticks collected, the species *I. aulacodi* was the most abundant. It represented 67.98% of ticks' total number. This value was higher than the one of *R. simpsoni* (29.11%). The three other species were very lowly represented (1.7% for *A. compressum*, 1.12% for *R. microplus* and 0.09% for *Ha. leachi*) (Figure 6).

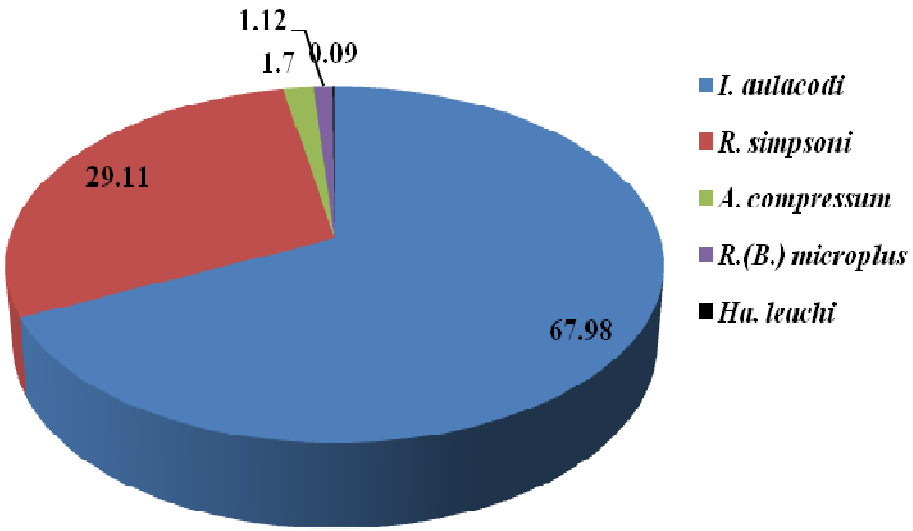

Figure 6: Abundance (%) of each tick species

As far as concerning the different species prevalence on grasscutters, the one of *I. aulacodi* (75.33%) was once again higher than the one obtained for *R. simpsoni* (54%), which also was higher than ones of *A. compressum* (6%), *R. microplus* (5.33%) and *Ha. leachi* (0.66%) (Table 1). According to regions, the prevalence obtained for *I. aulacodi* was included between 52% and 95%. The lowest value (52.94%) was recorded in the region of Grands Ponts and the highest one (95%) was observed in Agneby-Tiassa. These values of prevalence were higher than ones of *R. simpsoni*, according to all regions. Indeed, the prevalence of this species was between 38.88% (Lôh Djiboua region) and 75% (Agneby-Tiassa region) (Figure 7). As far as concerning the species *A. compressum,* its lowest prevalence (5.26%) was obtained in the Autonomous District of Yamoussoukro and the highest one (15%) in Agneby-Tiassa region. The prevalence of *R. microplus* species was 22.22%, 10%, 5.55% and 5% respectively for Agneby-Tiassa région, la Mé region, Autonomous District of Yamoussoukro and Bélier region. The species *Ha. leachi* was found only on one grasscutter in Lôh Djiboua region. Its prevalence was 5.55% in this region (Figure 7).

Table 1: Prevalence (%) and ticks' burden of wild grasscutters

Tick species	Prevalences on grasscutters	Infestation level	physiological stages	
			Adults	Nymphs
Ixodes aulacodi	75.33%[a] (113/150)	7±5[a] (1-47)	90,84%	9,16%
Rhipicephalus simpsoni	54%[b] (81/150)	4±2[b] (1-9)	100%	0%
Amblyomma compressum	6%[c] (9/150)	2±0,5[c] (1-3)	75%	25%
R.(Boophilus) microplus	5.33%[c] (8/150)	2±0,5[c] (1-2)	100%	0%
Haemaphisalis leachi	0.66%[d] (1/150)	1±0[c]	100%	0%

ab means in the same column with dissimilar superscripts are significantly different (p<0.05)

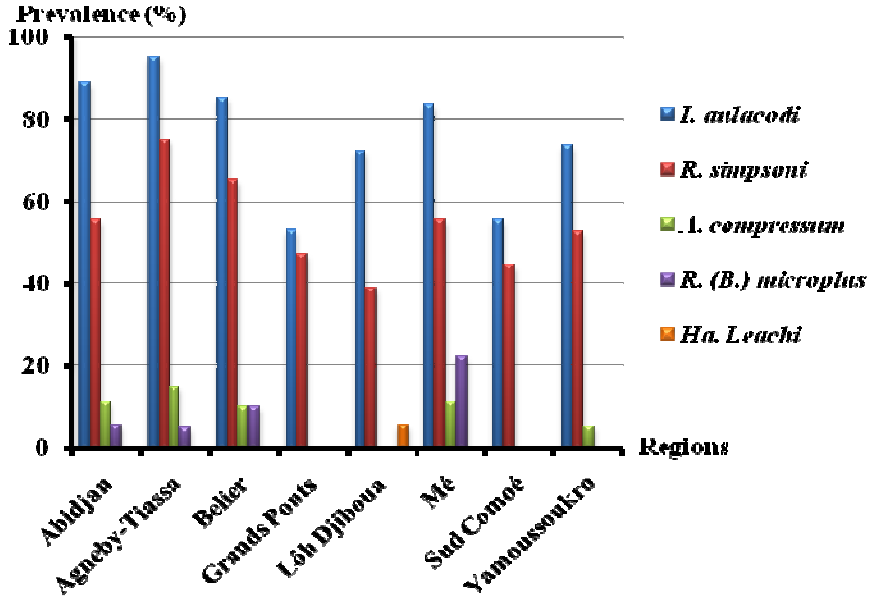

Figure 7: Prevalence of various tick species according to regions

- ***Infestation level of the different tick species collected :*** Globally, the infestation level of *I. aulacodi* was 7±5 with a minimum of 2 ticks per grasscutter and a maximum of 47 ticks per grasscutter. This number was higher than those observed with the other ticks. Indeed, the value obtained for *R. simpsoni* was about 4±2 ticks per grasscutter with a minimum number of 1 and a maximum number of 10. The one of *A. compressum* was 2±0.6 ticks per grass-cutter with a minimum number of 1 tick and a maximum number of 3. For *R. microplus,* it was 2±0.5 ticks per grasscutter with a minimum number of 1 tick and a maximum number of 2. As far as *Ha. leachi* species is concerned, only one tick was collected on average per grasscutter (Table 1). According to the regions, the number of *I. aulacodi* collected on average on a grasscutter varied from 6 to 8. Significant differences were not observed between these values. This infestation level varied from 3 to 6 for *R. simpsoni.* Those for *A. compressum* remained between 3 and 6, and then those for *R. microplus* went for 1 to 2.
- ***Physiological stage and sex of the collected ticks***All the collected ticks from *R. simpsoni*, *R. microplus* and *H. leachi* species were adults. Concerning *I. aulacodi*, 90.84% were adults and 9.16% were nymphs. For *A. compressum*, 75% were adults and 25% were nymphs. There was no larva in our collection whatever the species. The two sexes (males and females) were identified in all species with a female predominance except for *Ha. leachi* species for which a female was exclusively met (Table 1).
- **Co-infection cases:** Some of the wild grasscutters were suffering from poly-infestation. Indeed, 53% were infested by two ticks species. The combination *I. aulacodi* + *R. simpsoni* were the most encountered and accounted for 75.36% (Table 2). The other combinations such as *I. aulacodi* + *A. compressum* (12.33%), *I. aulacodi* + *R. microplus* (10.72%) then *I. aulacodi* + *Ha. leachi* (1.58%) have been examined (Table 2). Two tri-infestation cases were observed also in 6% of the wild grasscutters carrying ticks (Table 2).

Table 2: Poly-parasitism of wild grasscutters

Bi-infestations	Percentage	Tri-infestations	Percentage
I. aulacodi + *R. simpsoni*	75.36%	*I. aulacodi* + *R. simpsoni* + *A. compressum*	66.66%
I. aulacodi + *A.compressum*	12.33%	*I. aulacodi* + *R. simpsoni* + *R. microplus*	33.33%
I. aulacodi + *R. microplus*	10.72%		
I. aulacodi + *Ha. leachi*	1.58%		

DISCUSSION

This study represents the first inventory of grasscutters ticks in the south of Côte d'Ivoire. During the investigation, no tick was collected on the farm grasscutters. This could be explained by the care hygiene given by the grasscutter farmers in the management of theirs farms and by the method of drying fodder before they distribute it to animals. In fact, enclosures of grasscutters are cleaned every day and sanitized monthly. The fodder is dried up in the sun 24 hours before distribution in order to eliminate ectoparasites and endoparasites possibly present (Soro, 2007). Schrage & Yewadan (1995) also did not meet any tick on the farm grasscutters in Benin. On wild grasscutters, five ticks species from five genera have been collected. The species *I. aulacodi* and *R. simpsoni* have been the major species encountered with respective prevalences of 75.33% and 54%. Hence, they represent specific ticks to grasscutters (Morel, 1963; Morel & Mouchet, 1965) and they had been recorded since 1963 on grasscutters in Côte d'Ivoire, in the departments of Tiassalé and Toumodi (Morel, 1963). Furthermore, Yeboah & Simpson (2004) had also found on grasscutters of Ghana the following species: *I. aulacodi, Ixodes oldi* and *R. simpsoni*. The three other species in this study (*A. compressum*, *R. microplus* and *Ha. leachi*) were revealed at a proportion lower than 7%. Indeed, *A. compressum* is known to be specific tick of *Manis tricupis* and *Manis tetradactyla* (Aeschlimann, 1963). The species *R. microplus* has been introduced in Côte d'Ivoire during importation of Girolando bovines from Brasil (Madder *et al*., 2007). Moreover, this species has been collected in Côte

d'Ivoire southeast on cattle (Madder *et al*., 2011) and in many regions (center, north, east and west) (Touré *et al*., 2014). It is known for its invasive character and fast displacement of other species of the same subgenus (Touré *et al*., 2012; Adakal *et al*., 2013). Grasscutter would be one of its new hosts. As for *Ha. leachi*, this species is never in a large number. The immature ticks engorge themselves on micro-mammals belonging to *Leggada*, *Praomys* and *Hymolyscus* (Graf *et al*., 1981). Among the others adults hosts namely sheep, goat and dog, grasscutter constitute the new host for adults of this tick species in Côte d'Ivoire, as well as human (Morel & Mouchet, 1965). In addition, Yeboah & Simpson (2004) have collected another species of *Haemaphysalis* named *Haemaphysalis parmata* on wild grasscutters in Ghana. The prevalence of the four tick species *I. aulacodi, R. simpsoni*, *A. compressum* and *R. microplus* obtained in Agneby-Tiassa region were higher than ones of the other regions. Therefore, grasscutters of this region are the most infested. Indeed, it is noteworthy that the first country in west Africa were *R. microplus* has been discovered was Côte d'Ivoire, exactly in Azaguié, a village of Ageby-Tiassa region (Madder *et al.,* 2007). All of the collected ticks identified as *R. simpsoni*, *R. microplus* and *Ha. leachi* species were adults. On the other hand, some nymphs were collected for the species *I. aulacodi* and *A. compressum* in addition to the adults. Grasscutters could be listed among final hosts of *I. aulacodi*, *R. simpsoni*, *R. microplus* and *Ha. leachi*. The immature of those tick species would attach on other animals. Further studies are worthwhile to confirm this trend. Mixed infestations characterized by bi-infestations and tri-infestations have been experienced. This poly-parasitism was dominated both by the combination *I. aulacodi* + *R. simpsoni* for bi-infestations and the combination *I. aulacod* + *R. simpsoni* + *A. compressum* for tri-infestations. That should probably be because *I. aulacodi* and *R. simpsoni* are the most frequent ticks on grasscutters in Côte d'Ivoire.

CONCLUSION

In the foreground, this study highlighted that farm grasscutters in Côte d'Ivoire do not generally carry any kind of ticks. On the other hand, wild grasscutters carry some, at least five species belonging to five genera. They are *I. aulacodi*, *R. simpsoni*, *R. microplus*, *A. compressum* and *Ha. leachi.* The species *I. aulacodi* and *R. simpsoni* are more abundant, more prevalent and are considered specific to grasscutters. Nymphs were met only in *I. aulacodi* and *A. compressum* species. This study represents the first inventory of grasscutter ticks in the south of Côte d'Ivoire. Ticks are known to be vectors of many pathogens including zoonosis causative agents. Therefore, this work will continue in terms of diagnosis of blood parasites in the sampled ticks.

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