



PREVALENCE OF TICK SPECIES INFESTING CATTLE IN ITEK SUB-COUNTY, LIRA DISTRICT

BY

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19/U/0516

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A SPECIAL RESEARCH PROJECT REPORT SUBMITTED TO THE COLLEGE OF VETERINARY MEDICINE, ANIMAL RESOURCES, AND BIOSECURITY IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF A DEGREE OF BACHELOR OF VETERINARY MEDICINE OF MAKERERE UNIVERSITY

SEPTEMBER, 2024

DECLARATION

I **AJULA EMMANUEL**, declare that this report is an original copy of the work done by myself and has not been submitted to any other institution for the award of any degree, diploma or academic certificate.

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APPROVAL

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DEDICATION

To my mother Mrs. Molly Adongo & father Mr. Felix Ajula, and my siblings whose love and sacrifices allowed me to pursue my dreams, even when the path was uncertain. This report is a testament to the power of collaboration, perseverance and dedication. May it inspire future generations of researchers to pursue their passions with courage and curiosity.

ACKNOWLEDEMENT

The study has been out of along an academic journey and struggle filled with inspirations from many individuals. First and foremost, I would like to extend my profound appreciation to my supervisor, Dr. Nanteza Ann for without your immense efforts, nothing much would have been presented in this report. Thank you so much for the expert guidance, valuable feedbacks, and unwavering support throughout this research journey, I am forever grateful and to my brother, Mr. Etime Kizito, I am forever grateful for all the support of all kind you granted me throughout my studies, I am forever grateful for your endeavor and wish that almighty God reward you abundantly. I also want to thank the cooperation of the farmers who offered me support especially during restraint of animals and provided a wonderful environment for me during data collection, am forever grateful. Above all, to God be the glory.

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LIST OF ABBREVIATIONS AND ACROYMNS

CDC	Centers for Disease Control
CO ₂	Carbon-dioxide gas
COVAB	College of Veterinary Medicine, Animal Resources and Biosecurity
°	Degree
DVO	District Veterinary Officer
ECF	East Coast Fever
Dr.	Doctor
Km	Kilometer
NARO	National Agricultural Research Organisation
%	Percent
TBDs	Tick-Borne Diseases
TTBDs	Ticks and Tick-Borne Diseases
US\$	United states Dollars

ABSTRACT

Ticks and tick-borne diseases (TTBDs) are a significant threat to livestock production in sub-Saharan Africa. Cattle trade, communal grazing and the presence of wildlife predispose cattle to TTBDs. Tick species abundance and distribution data can be used as a tool for early disease diagnosis and inform tick control strategies. However, these data for Itek sub-county were limited. Ticks were randomly collected from 210 cattle spread across Itek sub-county from 27th March, 2024 to 2nd April, 2024. The ticks were identified morphologically using a light stereo microscope. A collection of 702 ticks was examined and four tick species from two genera (three *Rhipicephalus*; one *Amblyomma* species) were identified. *Amblyomma variegatum* was the most dominant 343 (54.71%) tick species, followed by *Rhipicephalus appendiculatus* 213 (33.97%); *Rhipicephalus decoloratus* 70 (11.16%) and *Rhipicephalus evertsi* 1 (0.16%). Three of these tick species were ubiquitous in all the parishes while one, *Rhipicephalus evertsi* was only found in Olilo parish. The proportion of cattle infested with ticks in Itek sub-county was 95.7%. This study demonstrates high tick infestation rates in cattle by different tick species with potential to transmit several tick-borne diseases including zoonotic pathogens in Itek sub-county, Lira district. *Rhipicephalus decoloratus* transmit *Babesia* pathogens that cause bovine babesiosis, *Rhipicephalus appendiculatus* transmit *Theileria* that cause theileriosis, *Amblyomma variegatum* transmit *Ehrlichia ruminantium* that cause heartwater. The study provides the knowledge on tick fauna in Itek sub-county. Tick occurrence and prevalence data are useful in the design of targeted tick control strategies which are affordable and environmentally friendly. There is a need to determine the extent of spread of tick species in other regions of Uganda.

CHAPTER ONE

1.0 INTRODUCTION

1.1 BACKGROUND

Ticks are arthropod ectoparasites that exclusively feed on blood and are found worldwide (de la Fuente & Contreras, 2015). They fall into two economically significant families: Ixodidae (hard-bodied ticks) and Argasidae (soft-bodied ticks). The Ixodidae family comprises genera such as *Amblyomma*, *Dermacentor*, *Haemaphysalis*, *Rhipicephalus*, *Hyalomma* and *Ixodes*. These ticks not only cause harm to their hosts but also rank second only to mosquitoes in disease transmission (Estrada-Pena et al., 2012). They have a substantial impact on global cattle populations, affecting approximately 80% of them and resulting in numerous health and economic consequences (Jaime Betancur Hurtado & Giraldo-Ríos, 2019). Among these, the cattle tick *Rhipicephalus appendiculatus* stands out due to its extensive distribution, disease transmission capability, blood-feeding habits, and the significant proportion of cattle it affects (Muhanguzi et al., 2020). The financial losses caused by this tick alone amount to roughly USD 13.9 - 18.7 billion annually on a global scale (Jaime Betancur Hurtado & Giraldo-Ríos, 2019). Some tick species thrive in warm and humid climates, with the genus *Rhipicephalus* being particularly adapting to various geographic regions around the world. This places nearly one billion bovines at risk of tick infestations (Jaime Betancur Hurtado & Giraldo-Ríos, 2019).

Research has shown a strong link between the economic impact and the epidemiology of tick-borne diseases (de la Fuente et al., 2019). The losses attributed to ticks and TTBDs can be categorized as direct and indirect. Direct economic losses result from tick bites, which cause damage to heavily infested animals, blood loss in cases of severe infestations, anemia, and severe allergic reactions due to tick saliva toxins, chronic stress and irritation. These factors not only affect the behavior and well-being of animals but also lead to immune suppression and energy loss due to constant movement in response to infestations (Abbas et al., 2014). Indirect losses arise from tick-transmitted pathogens. Other indirect costs include the expenses for treating clinical cases, tick control measures, unrealized income, or inefficiencies in the production system, such as the maintenance of genetically tick-resistant breeds that are less productive (loss of potential),

acaricide-contaminated animal products, and trade restrictions on livestock products with potential trading partners. Therefore, ticks not only cause economic losses through the direct mortality of high-value animals but also hinder the genetic improvement (productive potential) of entire herds or regions (Kivaria, 2006).

In Africa, TBDs pose a significant challenge to animal health, especially in smallholder farms across East, Central, and Southern Africa (Jongejan & Uilenberg, 2004). In Uganda, the equatorial climate favors livestock production and sustains large tick populations, facilitating the transmission of TBDs. The TTBDs have been responsible for over 75% of cattle losses in Uganda, with control costs accounting for 86% of total animal disease control expenses (Ocaido et al., 2009). East Coast fever (ECF) alone is responsible for approximately half of all calf deaths in pastoral and agro-pastoral production systems. To mitigate the substantial losses caused by tick infestations, it is essential to control tick populations to manageable levels (Parizi et al., 2009). Data on tick species abundance and distribution could serve as a valuable tool for early disease diagnosis and inform tick control strategies. However, such data for Itek sub-county, Lira District, Uganda, are currently limited.

1.2 PROBLEM STATEMENT

The inadequate development of Uganda's animal industry, particularly in the dairy and beef sectors, especially in rural areas where animals graze, can be attributed to various factors. Among these factors, ticks and the diseases they transmit have emerged as significant concerns. The increasing tick population has raised alarms among farmers and stakeholders, prompting a need for a comprehensive study to identify tick species and assess their prevalence in the areas where animals are kept. Numerous tick species are known to exist within tropical regions, but limited resources and efforts have been allocated to conducting thorough tick studies and precise identification. Consequently, there is a lack of precise information regarding the geographical distribution of ticks in Uganda.

The shifting rainfall patterns, interactions between domestic and wild animals, and the development of tick resistance to acaricides may have contributed to the dominance of certain tick species in specific regions of the country. Therefore, it is both logical and necessary to establish

an up-to-date assessment of tick populations in cattle, differentiating between various tick species in Itek Sub-county where the information is inadequate.

1.3 JUSTIFICATION

There is currently no information about the different tick species infesting cattle in Itek sub-county, Lira district in Lango sub-region, northern Uganda. This research is intended to find out the different tick species infesting cattle in the sub-county. This information can later be used to keep cattle keepers updated with the different tick species parasitizing cattle in the sub-county, as well as the potential tick-borne diseases they can expect as a result of ticks infesting their cattle. Additionally, veterinary drug sellers can use this information to stock acaricides and drugs necessary for effective control of ticks and the treatment of tick-borne diseases respectively.

1.4 OBJECTIVES

1.4.1 GENERAL OBJECTIVE

- To determine the diversity of tick species infesting cattle in Itek sub-county, Lira District, northern Uganda.

1.4.2 SPECIFIC OBJECTIVES

- i. To identify the different tick species present on cattle in Itek Sub-county.
- ii. To determine the prevalence of tick species infesting cattle in Itek sub-county.

1.5 RESEARCH QUESTIONS

- i. Which tick species are infesting cattle in Itek Sub-county, Lira District?
- ii. What is the prevalence of tick species infesting cattle in Itek sub-county, Lira District?

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Overview on ticks

Ticks belong to an ancient lineage, with specimens found in Burmese amber dating back to about 100 million years ago. They are a very successful group of arthropods and have adapted to feeding on practically every terrestrial mammal, bird and reptile, and have attacked humans and infested the animals that they have domesticated for thousands of years. During the past 150 years they have become an important concern of veterinary and medical research, not only because of the direct effects they have on their hosts, such as injury at their points of attachment, blood loss and paralysis caused by toxins in their saliva, but also because they are efficient vectors of a wide variety of micro-organisms (Madder et al., 2014)

Globally a total of close to 900 tick species have been described, of which slightly more than 700 species belong to the Ixodidae or hard ticks and the remainder to the Argasidae or soft ticks. Ticks are particularly abundant in the Afrotropical region with its rich animal fauna and climatic zones ranging from arid to tropical. Approximately 200 ixodid tick species (hard or shield ticks) and 40 argasid tick species (soft ticks or tampanas) are present in the Afrotropical region, but only a small number are of veterinary and medical importance. Many of the ticks and tick-borne diseases occur usually in specific geographical areas but with globalisation and climate change their range may expand and may even spread intercontinentally. Although it is common to consider domestic animals as being the preferred host of ticks, most species occur on wildlife, and several would not be able to complete their life cycles without the presence of small wild mammals or birds as hosts for their immature stages. Most importantly, at the livestock/wildlife interface transfer of tick-borne pathogens frequently occurs and poses a risk to livestock farming and development (Madder et al., 2014).

Failure to control ticks and tick-borne diseases effectively is a major factor limiting livestock production. The worldwide economic loss due to tick infestation and the additional burden of protecting livestock against ticks and tick-borne diseases is estimated to be in the billions of dollars annually (Madder et al., 2014).

2.2 Classification of ticks

Ticks are classified into three families, the hard ticks or ixodidae, the soft ticks or Argasidae, and a third family, the Nuttalliellidae, of which there is a single species of tick found only in Southern Africa, *Nuttalliella namaqua* (Johnson, 2023). Approximately 840 species of ticks are known to occur in the world and these are classified into the families Ixodidae and Argasidae (Jongejan & Uilenberg, 1994). And belong to the following taxonomic groups.

Kingdom: Animalia

Phylum: Arthropoda

Subphylum: Chelicerata

Class: Arachnida

Subclass: Acari

Order: Parasitiformes

Suborder: Ixodida (=Metastigmata)

Families: Ixodidae

Argasidae

Nuttalliellidae

Phylum Arthropoda: The distinguishing feature of arthropods is the presence of a jointed skeletal covering composed of chitin (a complex sugar) bound to protein. This nonliving exoskeleton is secreted by the underlying epidermis (which corresponds to the skin of other animals). Arthropods lack locomotory cilia, even in the larval stages, probably because of the presence of the exoskeleton. The body is usually segmented, and the segments bear paired jointed appendages, from which the name *arthropod* (“jointed feet”) (Barnes, R. D, 2023, December 12).

Subphylum Chelicerata: The body is divided into prosoma (cephalothorax) and opisthosoma (abdomen); no antennae; the first pair of appendages consist of chelicerae flanking the mouth; in most chelicerates, the other prosomal appendages are a pair of pedipalps and four pairs of legs.

Class Arachnida: They are characterized by having **two body regions, a cephalothorax and an abdomen**. They also have 6 pairs of appendages: **4 pairs of legs** and 2 pairs of mouthpart appendages, the first are called chelicerae (hence, the subphylum Chelicerata). The second pair of mouthpart appendages are called pedipalps.

Subclass Acari: exhibit extreme fusion of the body segments. The podosoma and the opisthosoma are fused, forming the idiosoma. In contrast with other arachnids, the opisthosomal region lacks distinct segmentation while the mouthparts are joined to a separate body region, the gnathosoma (= capitulum in ticks). Ticks and mites belong to this Subclass. Mites and ticks exhibit certain key characteristics that set them apart from other arachnids. Typically, they possess two body segments—the cephalothorax and the abdomen. However, in mites, these two segments often appear fused, giving them an oval or globular shape. Ticks, on the other hand, have a more distinct separation between the cephalothorax and abdomen. Adult Acari possess four pairs of legs, totaling eight legs, although they may have only six legs during the larval stage. Their mouthparts are adapted for specific feeding habits, which can range from piercing and sucking for feeding on plant fluids or blood to chewing for consuming solid food. Acari can vary widely in size, ranging from microscopic mites, which may only be a fraction of a millimeter in length, to larger ticks that can measure several millimeters to centimeters in size (“Subclass Acari – Mites & Ticks,” 2020).

The Acari are subdivided into two major orders:

Parasitiformes: these have stigmata pores (i.e., respiratory pores) on the podosome and/or opisthosomal portion of the body, and have well-developed, freely articulated coxae,

Acariformes: which lack stigmata and typically have the coxae fused with the ventral body wall.

Suborder Ixodida: Ticks belong to the suborder Ixodida (=Metastigmata), which is a suborder of the order Parasitiformes. These creatures are obligatory blood-feeding parasites characterized by a singular pair of stigmata positioned behind the third or fourth pair of coxae during post-larval stages. Furthermore, the hypostome, an organ used for feeding, is enlarged and protruding, featuring backward-facing denticles that anchor the tick into the host's tissue. Additionally, the first tarsus carries a noticeable sensory organ known as Haller's organ.

The suborder Ixodida comprises three families:

- Ixodidae or hard ticks, with 14 genera and approximately 700 species/subspecies;
- Argasidae or soft ticks, with 5 genera and approximately 190 species; and
- Nuttalliellidae with only one species *Nuttalliella namaqua* (Madder et al., 2014).

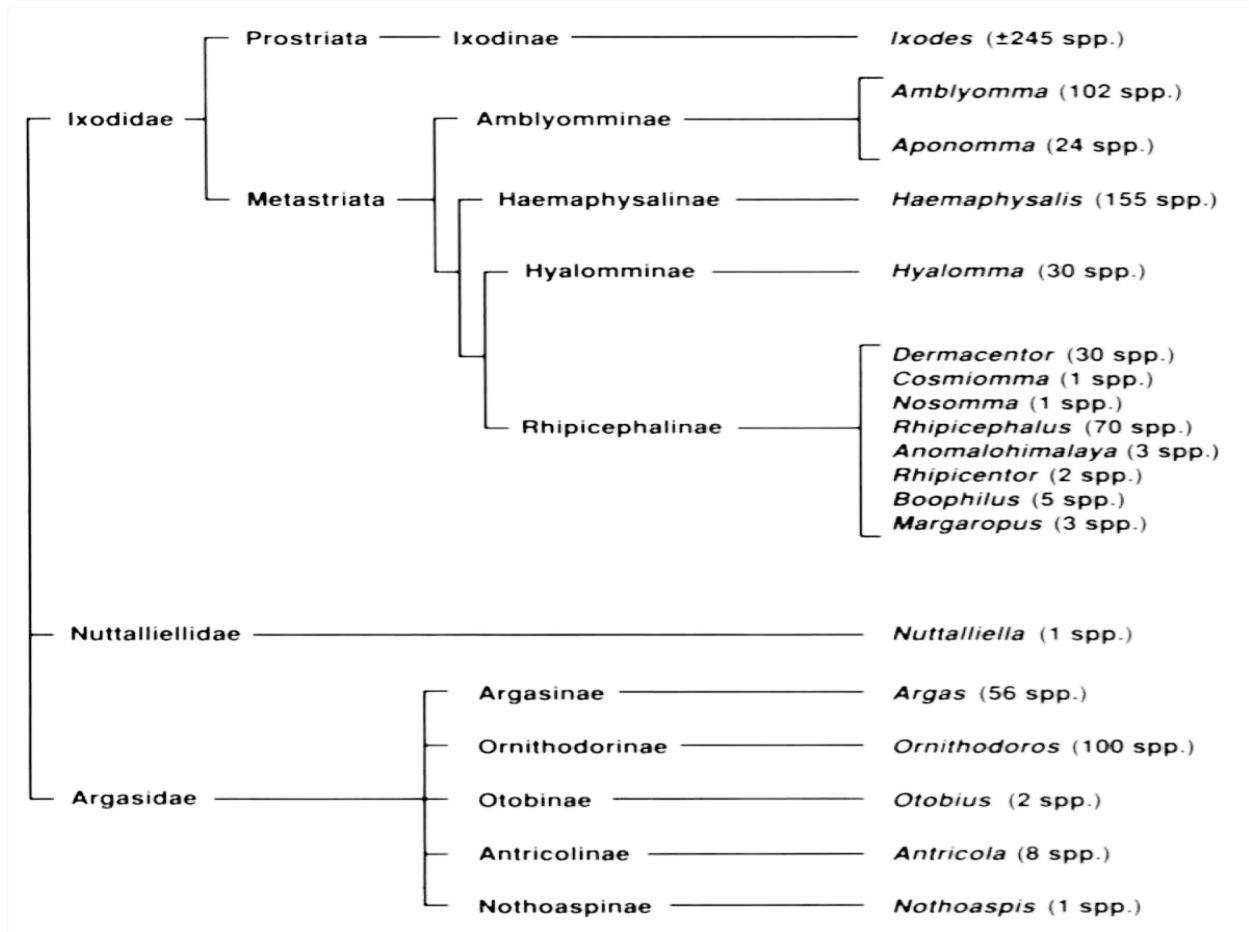


Figure 1. Genera of the three different tick families

2.3 Morphology

External Anatomy

The major external regions of ticks are the capitulum (gnathosoma), idiosoma, and the legs. The capitulum consists of the basis capituli, which articulates with the body; the segmented palps, the chelicerae, and the toothed hypostome. The capitulum of ixodid ticks is located at the anterior end of the body. Females bear paired clusters of pores, the porose areas, located dorsally on the basis capituli. The porose areas secrete antioxidants that inhibit degradation of the waxy compounds in the secretions of Gené's organ, which coat the eggs as they are laid. The chelicerae are located on the dorsal aspect of the capitulum. Their shafts, surrounded by spinose sheaths, lie between the palps and often extend even farther anteriorly than the palps. Each chelicera bears two digits distally. The larger, medial digit can be moved laterally; the smaller outer digit resides in a cavity of the medial digit and moves with it. Both digits have sharp denticles. The chelicerae are used to cut host tissues during attachment. The hypostome is a prominent, ventrally located structure that bears rows of recurved teeth on its ventral surface; teeth are absent in some non-feeding males. A narrow food canal is located on the mid-dorsal surface. The palps consist of four distinct segments. In nymphs and adults of most ixodid species, the small terminal (fourth) segment is recessed in a cavity in segment 3 and bears numerous fine setae at its tip (Nicholson et al., 2019).

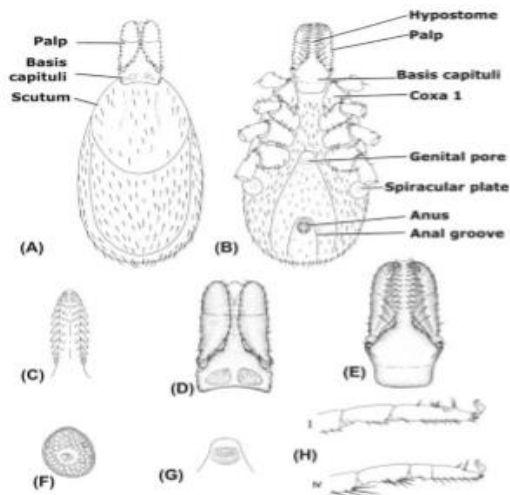


Figure 2. External morphology of representative female ixodid tick

External morphology of representative female ixodid tick (*Ixodes pacificus*). (A) Dorsal view. (B) Ventral view. (C) Hypostome. (D) Capitulum, dorsal view. (E) Capitulum, ventral view. (F)

Spiracular plate. (G) Genital pore. (H) Legs I and IV. Modified from Sonenshine (1991), with permission of Oxford University Press.

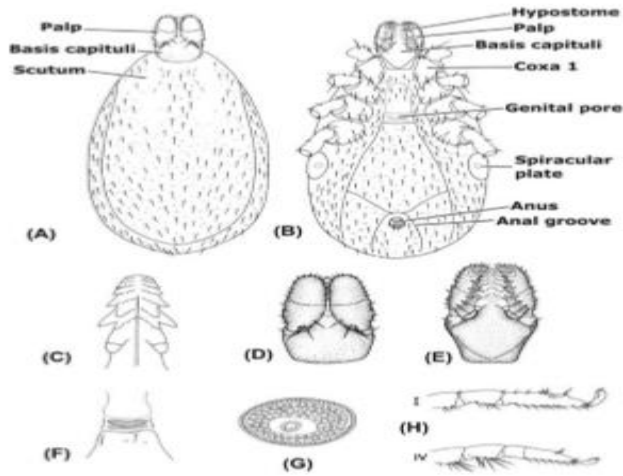


Figure 3. External morphology of representative male ixodid tick

External morphology of representative male ixodid tick (*Ixodes pacificus*). (A) Dorsal view; (B) Ventral view; (C) Hypostome; (D) Capitulum, dorsal view; (E) Capitulum, ventral view; (F) Genital pore; (G) Spiracular plate; (H) Legs I and IV. Modified from Sonenshine (1991), with permission of Oxford University Press.

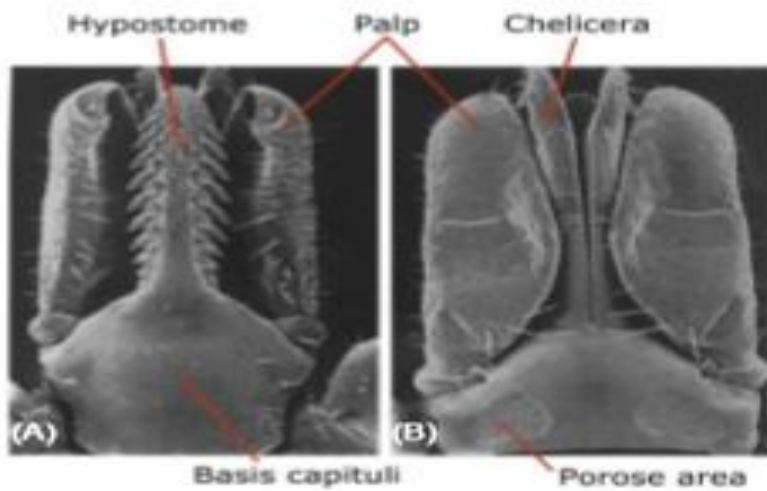


Figure 4. Capitulum of a representative ixodid tick

Scanning electron micrographs. (A) Ventral view. (B) Dorsal view. Modified from Sonenshine (1991), with permission of Oxford University Press.

The capitulum of adult and nymphal argasids is similar. However, it is situated just below an anteriorly protruding body extension, or hood, and is not visible dorsally in nymphs or adults

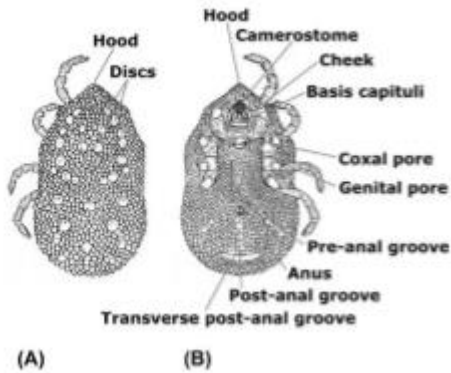


Figure 5. External morphology of a generalized argasid tick (*Ornithodoros*)

(A) Dorsal view (B) Ventral view



Figure 6. A representative female argasid tick (*Carios kelleyi*)

(A) Female, dorsal view. (B) Female, ventral view. Courtesy of Jim Gathany, Centers for Disease Control and Prevention.

The four palpal segments are about equal in size. Small flaps, the cheeks, occur alongside the capitulum in many species and can be folded to cover the delicate mouthparts. In argasid larvae, the mouthparts protrude anteriorly, as in ixodids. The body, exclusive of the capitulum, is the idiosoma. It is divided into two parts: the anterior podosoma that bears the legs and the genital pore, and the posterior opisthosoma, the region behind the coxae that bears the spiracles and the anal aperture. The cuticle is relatively tough with sclerotized plates (sclerites) in certain locations. It serves as the site of muscle attachment and protects the animal from desiccation and injury. The cuticle bears numerous sensory setae as well as various pores representing the openings of dermal glands or sensilla. The legs are jointed and articulate with the body via the coxae (Nicholson et al., 2019)

Larvae are easily recognized by the presence of only three pairs of legs, whereas nymphs and adults have four pairs of legs. The structure of the legs is similar in the Ixodidae and Argasidae. Each leg is divided into six segments: the coxa, trochanter, femur, patella ($\frac{1}{4}$ genu), tibia, and tarsus. The coxae are inserted ventrally and allow limited rotation in the anteroventral and dorsoventral planes. The other segments can be flexed, so that the legs can be either folded against the ventral body surface for protection or extended for walking. A pair of claws and a padlike pulvillus are present on each tarsus of most species. The pulvillus is absent in argasid nymphs and adults. An odor-detecting sensory apparatus, Haller's organ, is evident on the dorsal surface of the tarsus of leg I in all stages (Nicholson et al., 2019).



Figure 7. Haller's organ of the larval stage of Amblyomma rotundatum

Haller's organ of the larval stage of *Amblyomma rotundatum*, a parasite of reptiles and amphibians in the Americas; scanning electron micrograph. Courtesy of the U.S. National Tick Collection, Georgia Southern University.

This organ consists of an anterior pit and a posterior capsule. Olfactory and mechanosensory, but not gustatory functions also have been associated with this organ (Nuss et al., 2016). The tick's Haller's organ uses novel molecular processes for chemosensation different from those found in insects. Haller's organ also functions as an infrared receptor (Mitchell et al., 2017). Variations in the structure of Haller's organ are useful for distinguishing genera and species.

2.4 Life history

The life cycle includes four stages: the egg, larva, nymph, and adult. Ixodid ticks have only one nymphal instar, whereas argasid ticks have two or more nymphal instars. All ticks feed on blood during some or all stages in their life cycle; therefore, they are obligate ectoparasites. Larvae attack hosts, feed, detach, and develop in sheltered microenvironments where they molt to nymphs. Nymphs seek hosts, feed, drop, and molt to adults (except in argasid ticks, which molt into later nymphal instars). Adult ticks seek hosts, feed, and, in the case of engorged ixodid females, drop off to lay their eggs (Nicholson et al., 2019).



Figure 8. Female Lone star tick laying eggs

Lone star tick (*Amblyomma americanum*), female, that has just finished depositing an egg mass of about 4,000 eggs. Photograph by Gary R. Mullen.

In contrast to most other hematophagous arthropods, ticks can be remarkably long-lived. Many can survive for one or more years without feeding. Their life cycles vary greatly, with the greatest differences evident between the Ixodidae and Argasidae.

2.4.1 Lifecycle of hard ticks (Ixodidae)

The fully fed female detaches and drops off the host and after a few days, known as the pre-oviposition period, lays a single large batch of several thousand eggs in a sheltered spot and then dies. After weeks or even months, minute six-legged larvae hatch from these eggs. These larvae are known as "seed" or "pepper" ticks because of their similar morphology to small seeds or crunched pepper corns. The larvae of some species climb up the stems of grasses or other plants and wait for a passing host to which they attach, while the larvae of other species wait for a host on the ground and then climb on and attach. Following attachment, they engorge and a period of quiescence follows while structural changes (partial metamorphosis) take place inside the skin of each larva. The larvae then moult into nymphs that require a few days for the integument to harden before they will attach. They engorge, go through a period of quiescence and moult to adults that also require a few days for the integument to harden before they will attach. The partially engorged female is attractive to the partially engorged male that migrates to where the female is attached, they mate, and the female engorges, detaches, drops to the ground and lays eggs. The male may remain on the host for months before finally dying. The integument of the female undergoes physiological changes during the last 24 hours of engorgement. These changes make her less susceptible to desiccation and she also becomes less susceptible to the effects of acaricides (Madder et al., 2014).

2.4.1.1 One-host life cycle

Larvae hatch from eggs, climb on to a host, attach, engorge, moult on the host to nymphs, nymphs attach, engorge, moult on the host to males and females, adults attach, partially engorge, mate, females engorge fully, detach, drop to the ground, lay a single large batch of eggs in a sheltered locality and die. The next generation of larvae hatches from these eggs. The demographic structure of a parasitic population of one-host ticks is eight larvae to four nymphs to two males to one female. The population has this structure because it has been calculated that half the larvae do not successfully moult to nymphs. Some are probably lost by the host grooming itself, others during the moulting process. Similarly only half the nymphs will successfully moult to adults. The

difference in proportions between adult male and female ticks is possibly due to the larger females being more likely to be removed by host grooming, but more likely because engorged females engorge and detach from the host whereas the males can remain on the host for several weeks, resulting in a preponderance of male ticks (Madder et al., 2014).

The advantage of one- and two-host ticks is the relatively protected environment that the hosts offer to the vulnerable larval and nymphal stages. In this way, the immature stages are not exposed to hostile climatic conditions, which reduces mortality. Disease transmission of one-host ticks is limited to transovarial transmission where infection is passed from one generation to the next via eggs (Madder et al., 2014).

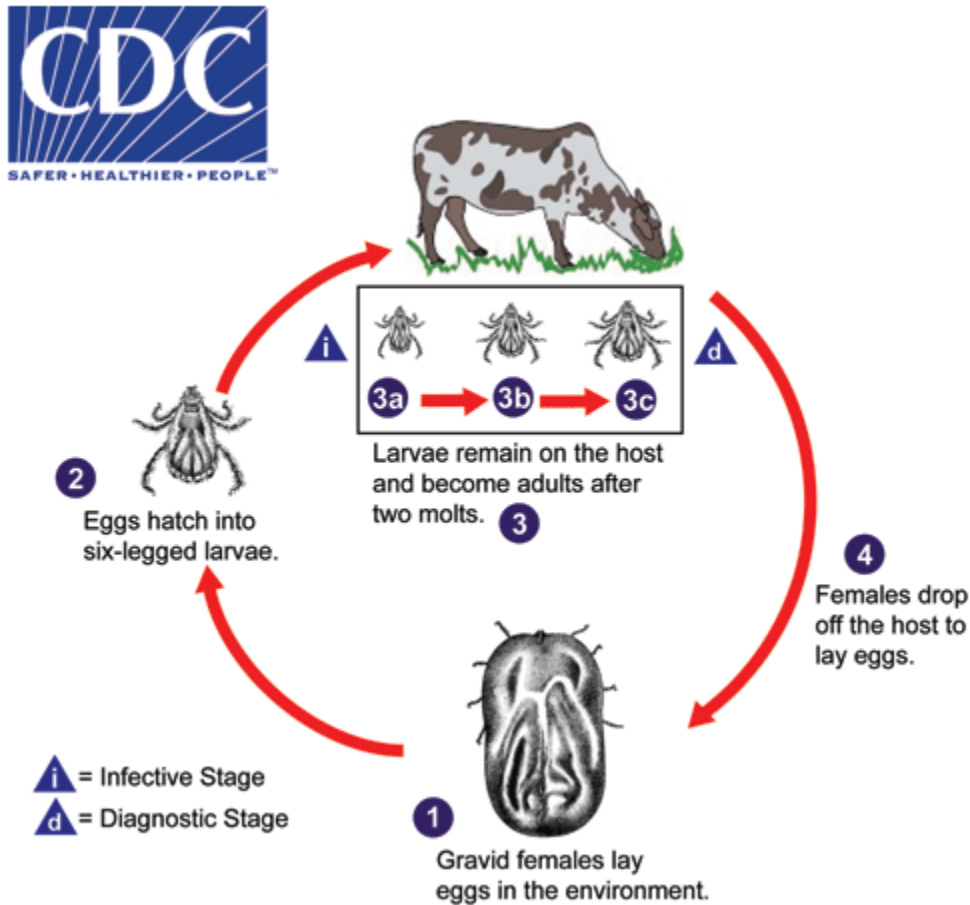


Figure 9. One-host tick lifecycle

One-host tick lifecycle (CDC - DPDx - Ticks, 2019)

2.4.1.2 Two-host life cycle

Larvae hatch from eggs, climb on to the first host, attach, engorge, moult on the host to nymphs, nymphs attach, engorge, detach, drop to the ground, moult to males and females in a sheltered locality, adults climb on to the second host, attach, partially engorge, mate, the females fully engorge, detach, drop to the ground, lay a single large batch of eggs in a sheltered locality and die. (Madder et al., 2014).

Transmission of pathogens can be transovarial from one generation to the next via ovaries or transstadial from the nymphal to the adult stage. In some species, e.g. *Hyalomma anatolicum anatolicum* host availability can influence the life cycle; it can feed on hares as a two-host tick for its entire life cycle and on cattle as a three-host tick (Madder et al., 2014).

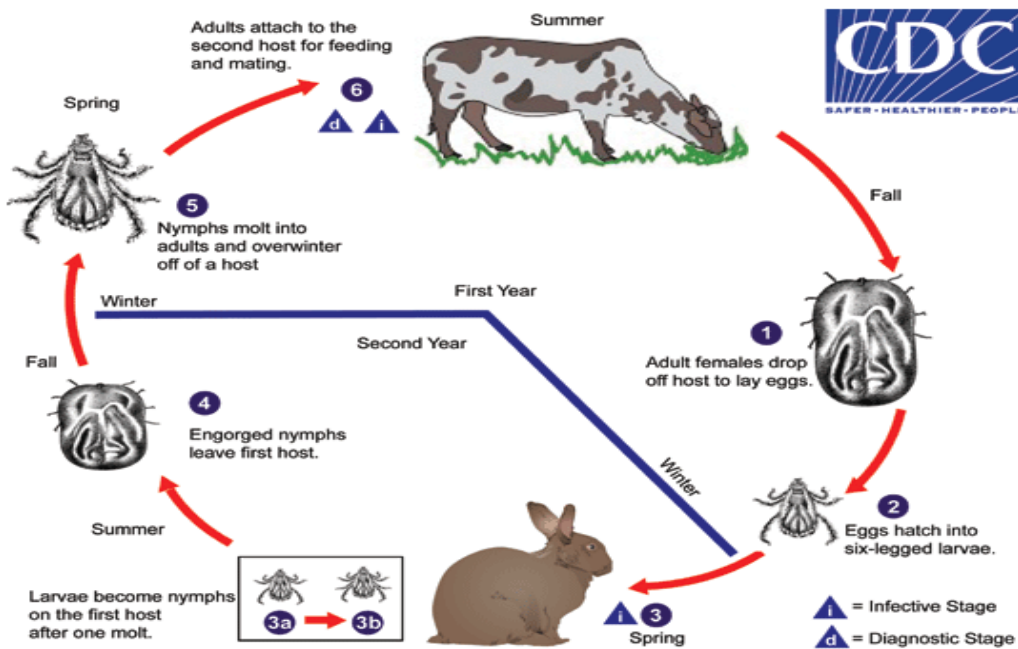


Figure 10. Two- host tick lifecycle

Two-host tick lifecycle (CDC - DPDx - Ticks, 2019)

2.4.1.3 Three-host life cycle

After oviposition, hatching of the eggs begins after a few weeks or a month depending on the temperature. The emerged larvae disperse into the vegetation or nest to seek hosts after hardening.

Once attached to a passing host, larvae feed slowly (several days) to repletion. The engorged larvae drop from their host and find a sheltered micro-environment. Ecdysis (=moulting) starts after several days. The newly emerged nymphs harden and again seek hosts (sometimes the same hosts as those fed upon by the larvae). They in turn attach, feed and drop from the host as engorged nymphs. They too try to find an appropriate niche to shelter and moult into adults. After hardening, male and female ticks start questing, attach, feed and mate after a small initial blood meal. Female ticks engorge to repletion after mating, drop off and find a suitable place to oviposit and finally die. Males on the other hand, can mate several times before they die. The engorgement weight of the females can sometimes be 100 X the unfed adult weight. About 50% of this weight is converted to egg. The largest egg mass ever recorded from a single *Amblyomma nuttalli* consisted of 22 891 eggs (Madder et al., 2014).

The three-host life cycle is the most common development pattern and is characteristic of the vast majority of the species. It is the least evolved of the various life cycle patterns and huge losses in numbers occur between the larval and nymph stages and between the nymph stage and adults. Transmission of pathogens can be transovarial, transstadial and intrastadial. (Madder et al., 2014)

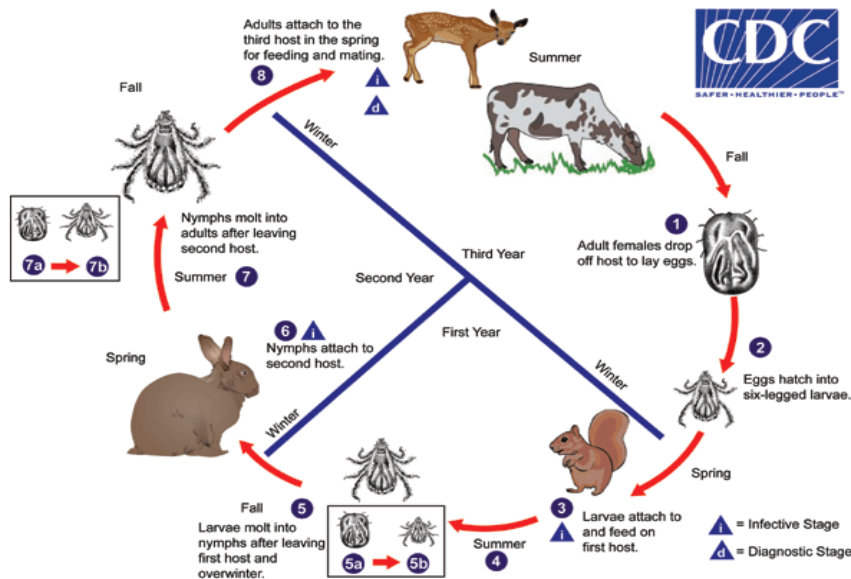


Figure 11. Three-host tick lifecycle

Three-host tick lifecycle (CDC - DPDx - Ticks, 2019)

2.4.2 Life cycle of soft ticks (Argasidae)

The life cycle of the argasid tick species is more diverse than the much more uniform pattern found in the ixodid tick species. Some soft ticks seek hosts by questing on low-lying vegetation, but the vast majority are nest parasites, residing in sheltered environments such as burrows, caves, or nests. Certain biochemicals such as carbon dioxide as well as heat and movement serve as stimuli that guide host-seeking behaviour. The feeding behaviour of many soft ticks can be compared to that of fleas or bedbugs, as once established, they reside in the nest of the host, feeding rapidly when the host returns. The outside surface, or cuticle, of soft ticks expands, but does not grow to accommodate the large volume of blood ingested, which may be anywhere from 5-10 times their unfed body weight. Argasid ticks feed rapidly, females feed and oviposit frequently (multiple gonotrophic cycles) and deposit small egg masses (< 500 eggs/cycle). There are also 2 - 7 nymphal stages (moult) in the life cycle (Madder et al., 2014).

In the majority of species, larvae seek hosts, feed within 15 - 30 minutes and drop off to moult in the sand, duff or cracks and crevices of the natural habitat. *Ornithodoros* larvae don't feed and moult immediately to the nymphal stage. The first stage nymphs resemble the adults, but are smaller, lack the genital pore and any evidence of dimorphism. They in turn attack hosts, feed rapidly and moult again to another nymphal stage. The cycle of host seeking, feeding and moulting can be repeated up to 7 times in the nymphal stage. After the last nymphal moult, adult ticks become sexually active and they do not require a blood meal to initiate gametogenesis. Mating occurs before as well as after feeding, but rarely if ever on the host itself. *Otobius* adults do not feed at all (Madder et al., 2014)

Following feeding, oviposition begins and once completed, the ticks remain vigorous, seek new hosts, feed and oviposit again. This pattern of repeated gonotrophic cycles enables argasid ticks to spread their progeny gradually over time, often across a span of many years. Diapause can be a major factor regulating the time of development of many of the argasid species, which survive in empty burrows or nests for periods of many months or years until their hosts return or new hosts (Madder et al., 2014)

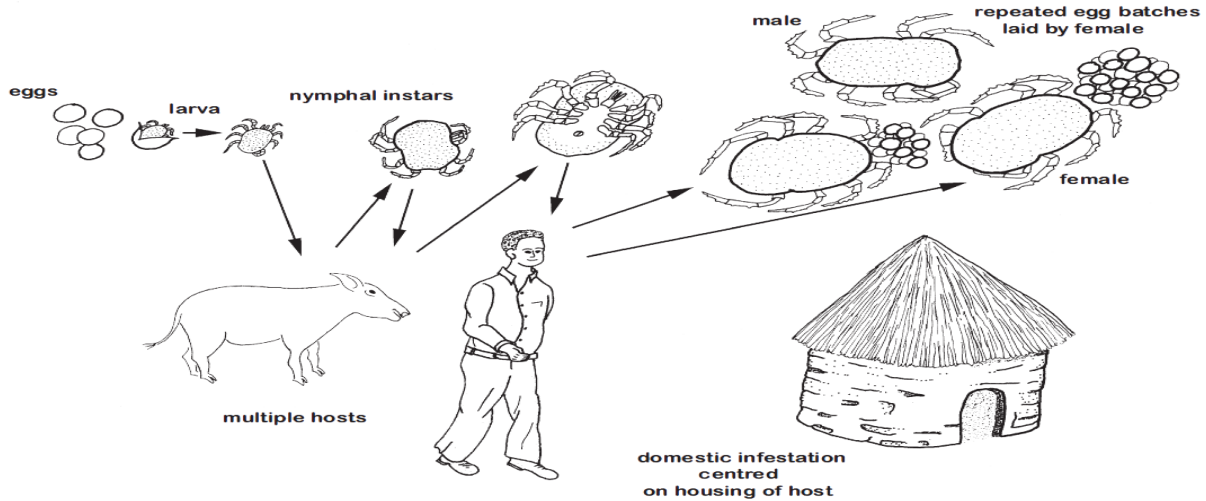


Figure 12. Argasid tick lifecycle

Argasid tick life cycle. The example is *Ornithodoros moubata* group, other argasid ticks may differ considerably. From “Ticks of Domestic Animals in Africa” (A. Walker et al., 2003).

2.5 Identification / general characteristics of tick genera (hard and soft ticks)

Although most people consider ticks to be either small and red or large and blue, on closer inspection many species are extremely colourful when examined under a stereoscopic microscope. The range of colours or ornamentation on the scutum, particularly of the males of certain species, is spectacular, from metallic mauve, shiny dark orange, bright yellow to iridescent green. The legs of certain species may also differ in colour from that of the scutum and the posterior edge of each segment of the legs may be encircled by an ivory-coloured band. Some of these features can readily be seen with the naked eye and the ticks have been given common names by farmers and researchers. Thus, we have bont (brightly coloured) ticks, bont-legged ticks with ivory-coloured bands around their legs, red-legged ticks whose legs vary from light to dark orange, yellow dog ticks, and blue ticks, the last-mentioned ticks acquiring their common name from the slaty blue colour of their engorged females (Madder et al., 2014)

The various genera of hard ticks can easily be differentiated by a set of features unique to each genus: mouthparts, basis capituli, scutum, eyes, festoons, adanal, subanal and accessory anal plates, coxae, anal groove (Madder et al., 2014)

2.5.1 Characteristics of the different genera of hard ticks

Amblyomma

Members of this genus are large, often highly ornate, ticks with long, often banded, legs. Unfed females may be up to 8mm in length and when engorged may reach 20mm in length. Eyes and festoons are present. Males lack ventral plates. They have long mouthparts with which they can inflict a deep, painful bite which may become secondarily infected. There are about 100 species of Amblyomma, largely distributed in tropical and subtropical areas of Africa (Wall & Shearer, 2001).

Rhipicephalus

The basis capituli is hexagonal and, in the male, paired adenal plates and usually also accessory adenal plates are found on each side of the anus. They are not ornate. Palps are short and eyes and festoons (Scutum usually uniformly brown, but four species have ivory-coloured ornamentation.) are usually present. Coxae of first pair of legs with long, prominent posteriorly directed spurs. Spiracular plates are comma-shaped. They infest a variety of mammals but seldom birds or reptiles. Most species are three-host ticks but some species of the genus are two-host ticks (Wall & Shearer, 2001).

Hyalomma

Species of this genus are medium-sized or large ticks, with convex eyes and long mouthparts, second segment of palps elongate. The males have ventral adenal, su-anal, and accessory anal plates. Festoons present, scutum pale to dark brown. Banded legs, Coxae of first pair of legs with long, prominent posteriorly directed spurs. Hyalomma species are usually two-host ticks, though some species may be three hosts. They are most commonly found on the legs, udder, tail or perianal region (Wall & Shearer, 2001).

Ixodes

Ixodes is the largest genus in the family Ixodidae, with about 250 species. The second segment of the palps may be restricted at the base, creating a gap between the palp and chelicerae. The mouthparts are long and are longer in the female than male. Auriculae latero-ventrally on basis capitulum. The fourth segment of the palps is greatly reduced and bears chemoreceptor sensilla. They are small, inornate ticks which do not have eyes or festoons. Males have several ventral

plates which almost cover the ventral surface. Ixodes can be distinguished from other ixodid ticks by the anterior position of the anal groove. In other genera of the Ixodidae the anal groove is either absent or is posterior to the anus (Wall & Shearer, 2001).

Haemaphysalis

Ticks of the genus *Haemaphysalis* inhabit humid, well-vegetated habitats in Eurasia and tropical Africa. They are three-host ticks, with the larvae and nymphs feeding on small mammals and birds and adults infesting larger mammals and, importantly, livestock. Most species of the genus are small, with short mouthparts and a rectangular basis capituli. Ventral plates are not present in the male. Spiracular plates are rounded or oval in females and rounded or comma-shaped in males. Like *Ixodes* spp., these ticks lack eyes, but they differ in having festoons and a posterior anal groove. There are about 150 species, found largely in the Old World, with only two species found in the New World (Wall & Shearer, 2001).

Dermacentor

Ticks of the genus *Dermacentor* are medium-sized to large ticks, usually with ornate patterning. The palps and mouthparts are short and the basis capituli is rectangular. Festoons and eyes are present. The coxa of the first pair of legs is divided into two sections in both sexes. Coxae progressively increase in size from I to IV. The males lack ventral plates and, in the adult male, the coxa of the fourth pair of legs is greatly enlarged. Most species of *Dermacentor* are three-host ticks, but a few are one-host ticks. The genus is small with about 30 species, most of which are found in the New World. Several of the species are directly associated with Rocky Mountain spotted fever, Q fever, tularaemia and Colorado tick fever. The salivary secretions of some species may produce tick paralysis (Wall & Shearer, 2001).

Rhipicephalus (Boophilus)

Mouthparts very short, proximal margins of palpal segments II and III sclerotized and have the appearance of two protruding rings. Conscutum often so poorly sclerotized that the dark pattern of the caeca can be seen from above. Eyes present but not conspicuous, no festoons, adanal plates and accessory adanal plates of males well developed. Caudal process may be present in males. One-host ticks (Wall & Shearer, 2001).

In summary the most prominent features that you must focus on when deciding to which genus a particular specimen of a tick belongs to are:

- Length of mouthpart
- Eyes or eyeless
- Conscutum ornate or inornate
- Colour of legs
- Festoons or no festoons
- Anal plates or no anal plates

2.5.2 Characteristics of the different genera of soft ticks

Argas

Integument leathery, mouthparts recessed ventrally and not visible from above (except in larvae). Eyes absent, Spiracular plate postero-laterally between 3rd and 4th pair of legs. Numerous symmetrically arranged discs on dorsal side of body. Lateral margin sharp with row of quadrangular cells on both the dorsal and ventral surfaces, sexual dimorphism limited mainly to the genital aperture and several nymphal stages (Wall & Shearer, 2001)

Argas persicus

The fowl tick, *Argas persicus*, is of considerable veterinary importance as a parasite of poultry and wild birds. It originated in the Palaearctic but has been introduced with chickens into most parts of the world and is now found throughout Europe, Asia and North America. However, in North America a number of very closely related species *Argas sanches*, *Argas radiatus* and *Argas miniatus* may also be present. It is also known as 'chicken tick', 'adobe tick', 'tampan' and 'blue bug' (Wall & Shearer, 2001).

Morphology: the unfed adult is reddish brown, turning slate blue when fed. The female is about 8mm in length and the male about 5mm. The margin of the body appears to be composed of irregular quadrangular plates or cells and no scutum is present. Unlike hard ticks, the four segments of the pedipalps are equal in length. The stigmata are situated on the sides of the body above the third and fourth pairs of legs. The integument is granulated, leathery and wrinkled. The hypostome is notched at the tip and the mouthparts are not visible when the tick is viewed from above (Wall & Shearer, 2001).

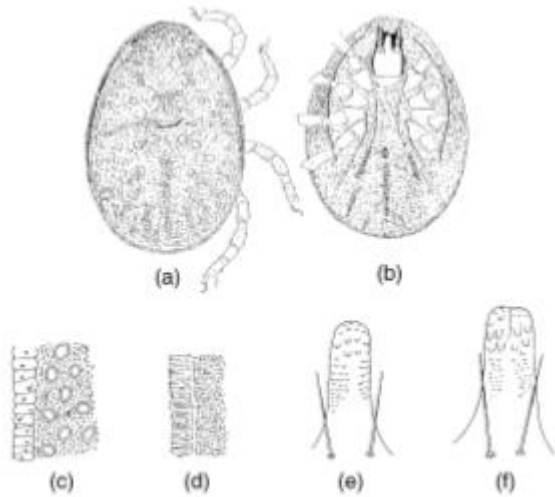


Figure 13. Female *Argas reflexus*

Female *Argas reflexus*: (a) dorsal and (b) ventral view (reproduced from Arthur, 1962). Margin of *Argas reflexus* (c) and *Argas persicus* (d). Hypostome of female *Argas reflexus* (e) and *Argas persicus* (f) (reproduced from Arthur, 1963).

Otobius

This small genus contains only two species: *Otobius megnini* and *Otobius lagophilus*. Dark, violin-shaped adults, mouthparts recessed ventrally and not visible from above (except in larvae). No lateral suture line on adults, Nymphs diamond-shaped becoming violin-shaped with numerous spines on the body. Larvae pear-shaped with clearly visibly anteriorly projecting mouthparts (Wall & Shearer, 2001).

Otobius megnini

The spinose ear tick is found through western and south-western North America and Canada, where it originated. It has subsequently been introduced to southern Africa and India. It most commonly infests wild and domestic animals, including sheep, cattle, dogs and horses. It has been found in humans (Wall & Shearer, 2001).

Morphology: the adult body is rounded posteriorly and slightly attenuated anteriorly. Adult females range in size from 5 to 8mm in length; males are slightly smaller. They have no lateral sutural line and no distinct margin to the body. Nymphs have spines. In adults the hypostome is much reduced and the integument is granular. The body has a blue-grey colouration with pale yellow legs and mouthparts (Wall & Shearer, 2001).



Figure 14. Dorsal view of nymphal *Otobius megnini* and part of the integument showing hairs and spines

(reproduced from Arthur, 1962)

Ornithodoros

Leathery-mammillated integument, mouthparts recessed ventrally and not visible from above (except in larvae). Body margin rounded. Supra-coxal fold, eyes absent or two pairs in the supra-coxal fold (Wall & Shearer, 2001).

2.6 Epidemiology

2.6.1 Host relationship

Ticks find their hosts in several ways. Some ticks live in open environments and crawl onto vegetation to wait for their hosts to pass by. This is a type of ambush and the behaviour of waiting on vegetation is called questing. Thus in genera such as *Rhipicephalus*, *Haemaphysalis* and *Ixodes* the larvae, nymphs and adults will quest on vegetation. The ticks grab on to the hosts using their front legs and then crawl over the skin to find a suitable place to attach and feed. Adult ticks of the genera *Amblyomma* and *Hyalomma* are active hunters, they run across the ground after hosts nearby. The general behaviour of seeking hosts in an open environment is described as exophilic. Ticks such as argasids and many *Ixodes* species spend their entire life cycle in their host's nest and attach to their hosts there. This is called endophilic or nidicolous behaviour. Some species such as the dog tick *Rhipicephalus sanguineus*, have adapted to living in housing. This is called domestic behavior (A. Walker et al., 2003). During questing, ticks may lose water, which they normally regain by descending at intervals to the litter zone (the layer of dead leaves, grass, twigs, etc. covering the soil) (Perret et al., 2004).

2.6.2 Attachment

The limitation of tick populations to specific attachment sites on hosts is a regulatory system that operates by restricting tick species to particular regions of the host's body. Ticks actively choose locations on the host that provide protection and favorable conditions for their development, as indicated by various studies (Huruma et al., 2015) and (Jittapalapong et al., 2004) indicated that different ticks have different predilection sites on the host's body.

Rhipicephalus decoloratus tends to prefer the lateral and ventral sides of the animal, *Amblyomma variegatum* shows a predilection for the teat and scrotum, and *Hyalomma truncatum* targets the scrotum and brisket. Meanwhile, *Hyalomma marginatum rufipes* tends to favor the udder and scrotum, *Rhipicephalus evertsi evertsi* is often found under the tail and anus, and *Rhipicephalus preaxtatus* tends to inhabit the anus and under the tail (Huruma et al., 2015). The choice of attachment site on the host by ticks is contingent upon factors such as accessibility for attachment, obtaining blood, and protection against environmental challenges that could hinder their survival. The location of ticks on the host is linked to the potential for penetration by the hypostome. Genera with short hypostomes, such as *Rhipicephalus*, *Dermacentor*, and *Haemaphysalis* species, typically attach to hairless areas like under the tail and the anovulval region (Huruma et al., 2015).

2.6.3 Feeding

Once a site has been selected, the tick cuts into the skin with its cheliceral digits and inserts its hypostome to initiate the attachment process. Shortly after they attach, most ixodid ticks secrete cement during the first 1-2 days to secure themselves at the wound site. Subsequently, the tick begins salivating into the developing hematoma and sucking blood; salivating and blood sucking alternate, often for extended periods of time for each process. The feeding lesion enlarges as the tick injects anticoagulant and antihemostatic compounds into the wound; recruitment of host leucocytes to the wound site also contributes to tissue lysis and fluid influx around the tick's mouthparts. Successful blood feeding depends upon the secretion of an extensive array of antihemostatic, anti-inflammatory, and immunomodulatory proteins and lipids in the tick saliva so as to suppress the host's ability to reject the feeding tick (Mullen & Durden, 2019).

2.6.4 Tick distribution in Uganda and factors affecting their spread

Various agro-ecological zones offer different climatic and ecological conditions that affect tick survival. In northern and western Uganda, for instance, most zones feature expansive savannah grasslands with varying tick densities. Ticks thrive best in warmer, more humid regions that have

a mix of tree cover, such as savannah woodland, and grass (Gachohi et al., 2012). In addition, low elevations (lowlands) are ecologically more suitable for tick survival than higher elevations (uplands) (Rubaire-Akiiki et al., 2006). Therefore, in Uganda the highest tick density is observed in the Lake Kyoga and Lake Victoria basins (Nchu et al., 2020). Such areas are notable for high seroprevalence and possibly endemic stability to various tick-borne pathogens (TBPs) (Gachohi et al., 2012) and northern savannah grassland are less favorable for tick survival and multiplication, hence the lower comparative tick density (Kabi et al., 2014).

The distribution of ticks varies due to several climatic factors, including rainfall, temperature, relative humidity, as well as vegetation, crops, and husbandry practices. For instance, the density of *Rhipicephalus appendiculatus* (brown ear tick) tends to increase with the extent of vegetation cover (Smith, 1969). The commonest tick species infesting livestock in Uganda are *Rhipicephalus* spp. and *Rhipicephalus appendiculatus* is generally the most abundant tick in the country (Vudriko et al., 2016). Recently, *Rhipicephalus pravus*, *Rhipicephalus praetextatus* and *Rhipicephalus turanicus* were reported for the first time in cattle in Karamoja, Uganda (Byaruhanga et al., 2021). Other tick species parasitizing cattle in Uganda include *Amblyomma lepidum*, *Amblyomma variegatum* and *R. evertsi evertsi* (Byaruhanga et al., 2021).

Among the tick species in Uganda, the most economically important ticks are *Rhipicephalus appendiculatus*, *Rhipicephalus decoloratus* and *Amblyomma variegatum*. These ticks are widespread throughout the country and lack seasonal variation in abundance (Kaiser et al., 1982). Kaiser (1982) attributes the higher density of *Rhipicephalus appendiculatus* to limited resistance of cattle to the tick. In addition, because *Rhipicephalus appendiculatus* is a three-host tick, it spends short periods on the host and stays longer in the environment, hence requiring more frequent acaricide application to significantly reduce its abundance. Individual animals exhibit variations in the degree of resistance to ticks, which could support selection of tick resistant hosts as a viable strategy for the control of different tick species.

In Uganda, indigenous cattle are mostly managed under the extensive grazing system where they commonly get infested with several overlapping generations of ticks annually, without clear seasonal abundance, but with all instars occurring simultaneously on the animal (Kaiser et al.,

1982). Ticks however have been reported to be more abundant after rain seasons (Chenyambuga et al., 2022). On the contrary, during the dry season, because of scarcity of pastures and water, livestock farmers (pastoralists and agro-pastoralists) practice mobile pastoralism, during which animals migrate and share grazing and watering points, exposing them to heavy tick infestations (Byaruhanga et al., 2018)

2.7 Major tick-borne diseases /conditions

Tick-borne diseases have particularly been responsible for maintenance of poor local cattle breeds in Uganda due to fear by farmers to adopt or cross their animals with more productive but susceptible exotic breeds, leading to a loss termed “lost potential”. Research carried out by (Chenyambuga et al., 2010) around Lake Victoria basin identified cattle diseases as the major constraint to cattle production, of which TBDs ranked as the most important cattle diseases in Uganda and Tanzania. The study also identified ECF caused by *Theileria parva* as the most relevant TBD in Uganda.

Babesiosis

Bovine babesiosis is a severe and often fatal disease of cattle caused by protozoan parasites *Babesia bigemina* and *Babesia bovis* which are transmitted by the ticks, *Rhipicephalus decoloratus* and *Rhipicephalus microplus*, respectively. The disease is characterized by fever, anemia, red urine (due to hemolysis) and death of the susceptible hosts (Tayebwa et al., 2018). The main species causing disease in Uganda is *B. bigemina* due to the widespread distribution of its tick vector, *Rhipicephalus decoloratus*. *Babesia bovis* has not been detected in Uganda, but its vector *Rhipicephalus microplus* has been recently identified, probably introduced from neighboring Kenya (Muhanguzi et al., 2020).

Theileriosis

Theileriosis is a hemoparasitic disease caused by protozoa of the genus *Theileria* (Apicomplexa) (Weiss et al., 2010). Theileriosis is caused by *Theileria* spp. in cattle, goats, sheep and wild and captive ungulates (Radostits & Done, 2007). The parasites are transmitted by tick such as *Rhipicephalus* and *Hyalomma* species. These parasites undergo repeated merogony in the lymphocytes ultimately releasing small merozoites, which invade the red cells to become piroplasms. Theileriosis have a variety of tick vectors which cause infections ranged from clinically inapparent to rapidly fatal (Taylor et al., 2007). Recently *Theileria* (*T. velifera*, *T.*

mutans, *T. ovis*, *T. separata*, *T. annulata*) infection in domestic ruminants is reported in northern Ethiopia (Langana, 2017).

Theileria parva which causes East coast fever is transstadially maintained by the three-host tick, *Rhipicephalus appendiculatus* (Norval et al., 1992). The disease results into lymphnode enlargement, fever and anorexia which cause lymphadenopathy and subsequently death if not treated within three weeks (Magona et al., 2008). In Uganda, the disease kills approximately 30% of the annual calf-crop of the indigenous cattle, and almost all infected untreated exotic cattle and their crosses succumb (Otim, 2000).

Cowdriosis (Heartwater)

Heartwater caused by rickettsia organism, *Ehrlichia ruminantium*, is a virulent but non-contagious disease that affects cattle and small ruminants. Infected animals manifest nervous signs, pericardial and pleural effusion (Musisi & Lawrence, 1995). In the affected regions of Africa, the pathogen is transmitted by the tick *Amblyomma variegatum* which is wide-spread in eastern, northern and central parts of Uganda (Otim, 2000). The disease is endemic in sub-Saharan African countries and it has a serious negative impact on livestock productivity, with high morbidity and mortality rates (up to 90%) in susceptible ruminants. European breeds are generally more susceptible than more susceptible than indigenous African breeds (Escobar et al., 2018).

Anaplasmosis (gall sickness)

Bovine anaplasmosis is a subacute to acute disease of cattle mainly caused by a rickettsial organism, *Anaplasma marginale* and occurs worldwide (Musisi & Lawrence, 1995).

Anaplasma marginale is an intra-erythrocytic bacterium transmitted biologically by ticks, but mechanical transmission is also possible by blood-sucking flies and contaminated fomites (Battilani et al., 2017). Transmission from dam to calf through the placenta can also occur (Costa et al., 2016). The disease causes a severe anemia and death if untreated. Bovine anaplasmosis is prevalent in the tropics and subtropics where it causes great economic losses due to high morbidity and mortality, reduced weight gain and milk yield, abortions, and treatment expenses especially among newly introduced highly susceptible exotic dairy cattle (Battilani et al., 2017). Recovered animals also require a long period of convalescence during which they are less productive. These animals remain reservoirs and act as a potential source of infection to newly introduced susceptible

cattle where ticks are present (Ssenyonga et al., 1992). *Anaplasma centrale*, transmitted by the African *Rhipicephalus simus* (Potgieter & Rensburg, 1987) is a subacute form of anaplasmosis and is used as a live vaccine against *Anaplasma marginale* in some countries. Besides cattle, anaplasmosis affects a wide range of hosts including sheep, goats, buffalo and some wild ruminants (Tumwebaze et al., 2020). In Uganda, the prevalence of anaplasmosis ranks second to that of ECF (Kasozi et al., 2021) and in some areas incriminated as the major killer of adult cattle (Ocaido et al., n.d.). The overall seroprevalence obtained from the central, southwestern and northeastern (58-62%) (Ssenyonga et al., P.D. Kasaija et al. Ticks and Tick-borne Diseases 12 (2021) 101756 5 1992) and northwestern (83%) (Byaruhanga et al., 2018) regions of Uganda indicate extensive exposure of indigenous cattle to anaplasmosis. However, the mild effects of the disease among cattle in the affected regions suggests a state of endemic stability for anaplasmosis. The major tick vector for anaplasmosis, *Rhipicephalus decoloratus*, is widely distributed throughout Uganda. Studies however show that bovine anaplasmosis is more prevalent during wet seasons when hematophagous insects such as *Tabanus*, *Stomoxys calcitrans*, *Culicoides* and mosquitoes are more abundant to facilitate mechanical transmission (Byaruhanga et al., 2018).

2.8 Tick control measures

In Uganda, economic control of tick-borne diseases has been identified as the single most important issue as far as animal health is concerned (de la Fuente et al., 2019).

2.8.1 Chemical control methods

Currently, the most recommended control method for ticks is the use of acaricides in the appropriate concentrations by dipping or spraying at weekly intervals (Vudriko et al., 2016). However, small and medium size farms cannot afford the costs of establishing and maintaining the dip tank infrastructure (Vudriko et al., 2018). In consequence, spraying is the most commonly used method of acaricide application in Uganda (Byaruhanga, Collins, et al., 2015)

A wide range of acaricides, including arsenical, chlorinated hydrocarbons, organophosphates, carbamates and synthetic pyrethroids are being used for controlling ticks on livestock (De Meneghi et al., 2016). The performance of an acaricide in the control of ticks depends not only on the activity of a product, but on the quality and quantity of active ingredient deposited on cattle or delivered internally (A. R. Walker, 2011).

2.8.2 Biological control methods

Entomopathogens are group of organisms that attack ticks and insects. It can be macro- or microorganisms that affect arthropods (Samish & Alekseev, 2001). The biological agents, which potentially include predators like rodents, birds, ants, spiders, lizards and beetles as well as Parasitoids (destroy the host: the wasp lay the eggs in the engorged ticks and larvae eats the tick and emerges as adult to attack another tick) and parasites (Nematodes and fungus) are attack soil living stages of the ticks are effective and depending on the conditions, these predators can consume a large number of ticks. Yet, having such effective importance the development of a biological tick control methods has been neglected as compared to the control of plant pests or dipterous insects harmful to men and animals (Samish & Alekseev, 2001).

2.8.3 Anti-tick vaccine

The continuous and improper use of these chemicals (acaricides) has led to a rise in the prevalence of ticks that are resistant to acaricides (Klafke et al., 2006), and the occurrence of environmental and food contamination has prompted a growing economic and social need for the development of new methods for controlling ticks (Parizi et al., 2009). As integral components of integrated control programs, anti-tick vaccines emerge as a promising, environmentally friendly, and effective intervention for managing tick infestations and the transmission of Tick-Borne Pathogens (TBPs). These vaccines utilize immunologically active tick proteins as antigens, stimulating the production of antibodies when introduced into a vertebrate host (Nchu et al., 2020). Protection by vaccination is achieved when antigen-specific antibodies developed in immunized hosts interact and affect the function of the targeted antigen in ticks feeding on immunized hosts (Merino et al., 2011).

In Uganda, the National Agricultural Research Organisation (NARO), in partnership with IREC, Spain, has carried out on-station clinical trials for a candidate recombinant Subolesin anti-tick vaccine developed by the latter group. The trials have shown promising results (Kasaija, 2020). The two groups are also developing multi-epitope-based antigens and modifying vaccine formulations for possible enhancement of efficacy. The Molecular and Computational Biology Research group at the College of Veterinary Medicine, Animal Resources and Biosecurity (COVAB), Makerere University, is also conducting in silico protein and transcriptome/proteome studies to identify candidate anti-tick antigens for possible vaccine development (Nchu et al., 2020).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 study area

This study was conducted in Itek sub-county, Erute-south county, Lira district, Northern region, Uganda. The sub-county is situated about 17 Kilometers (Km) to the east of Lira city and it is about 389 km from Kampala city along the Lira-Kotido Road. It is bordered by Barr sub-county to the north and east, Lira city to the west, Agali subcounty to the south. The sub-county is composed of five parishes, Alebere, Onywako, Olilo, Ajja and Tetyang (Figure 15).

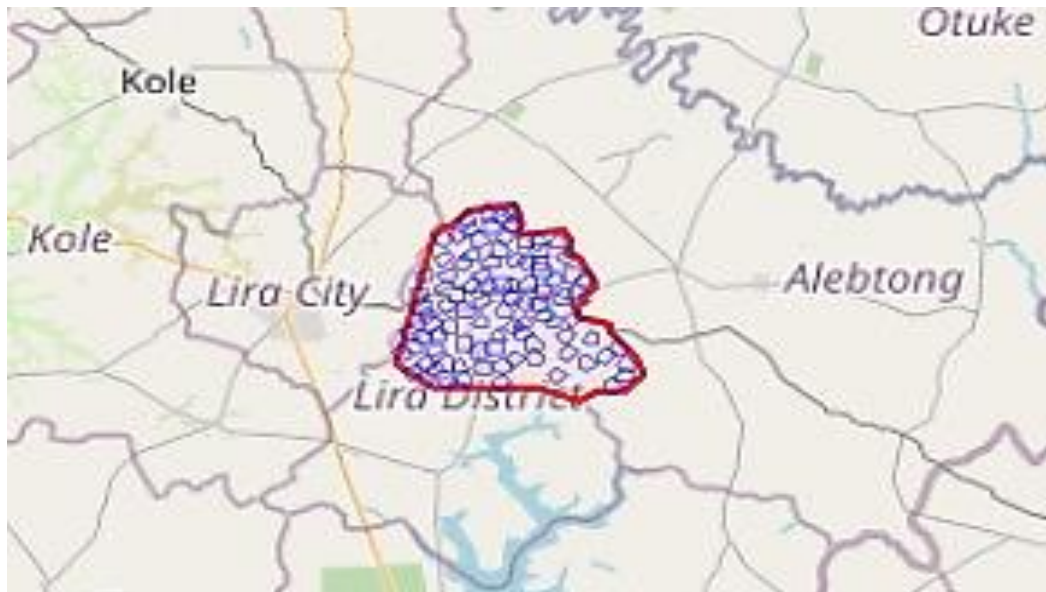


Figure 15. Map of Lira district (open street map) showing villages in Barr sub-county where Itek sub-county was carved from. (OpenStreetMap contributors, 2024)

3.2 Study design

A cross-sectional study was carried out on cattle to assess the prevalence of tick species investing cattle in Itek sub-county, Lira District from 27th March, 2024 to 2nd April, 2024.

3.3 Study population

The target population for the study were cattle kept by farmers in Itek sub-county, Lira district. Cattle owners who were not willing to offer their cattle to be used for the study were excluded from the study.

3.4 Sample size determination

The sample size was calculated using single population proportion formula with confidence level of 95% and 5% significance level. Based on the cross-sectional study design, the formulae for calculating sample size for population less than 10,000 as described by Hulley et al. (2007) was used.

$$n = \frac{2(z)^2 (p)(q)}{d^2}$$

n = the desired sample size;

z = standard normal deviation which was set at 1.96 corresponding to 95% confidence interval;

p = we assumed that there were at least 40% of cattle infested with ticks

q = 1-p, assuming that an observed difference of 10% between cattle infested with ticks and those not infested with ticks will be significant at 95% confidence interval. After substituting in the above figures, the calculated sample size is as follows;

$$n = \frac{2(1.96)^2(0.4)(0.6)}{(0.1^2)}$$

n = 184 cattle

3.5 Household and animal selection

The study households was chosen through a multistage sampling technique, involving the selection of samples from the population in progressively smaller groups at each stage. The selection of households for the study was random.

There was no inclusion or exclusion criteria applied, all animals were selected randomly, putting in mind the number of animals to be sampled per household. Different numbers of animals were sampled per parish basing on the number of animals in the parish as directed by the sub-county veterinarian.

3.6 Collection of data

3.6.1 Animal restraint

Selected animals was properly restrained in the crush in cases where the farmer had it, a figure of 8 rope on the rare legs was put and also a nose lead was applied on the animal. In absence of a

crush; animal restraining technique of casting down the animal was performed (A. I. Annatte, 2000).

3.6.2 Tick sampling

All the visible ticks (mature and immature, engorged and non-engorged) were manually removed from each animal by hand picking from whole the animals' body by using thumb forceps or by holding the tick's head firmly, turning it on its back and pulling it out sharply with attention being paid to the predilection sites (Wall & Shearer, 2001). All collected ticks from animals in each parish were preserved in collecting tubes, each filled with $\frac{3}{4}$ of 70% ethanol. The tubes were securely sealed with non-perforated lids and labeled using a permanent marker. Subsequently, they were placed in a cool box for transportation to the Parasitology Laboratory at the College of Veterinary Medicine, Animal Resources, and Biosecurity (COVAB).

3.6.3 Tick examination and identification

Ticks were examined grossly on the petri dish and then under a Stereomicroscope for microscopic morphology. The gross and microscopic characteristics of each genus and species were used to identify the ticks according to Walker et al (2003).

3.7 Statistical analysis

The gathered data was inputted into a Microsoft Excel spreadsheet. Subsequently, descriptive statistics, such as percentages and tables, was utilized to provide a concise summary of the data.

3.8 Ethical considerations

Approval was requested from dean of School of Veterinary Medicine and Animal Resources (SVAR). Additionally, permission was sought from the District Veterinary Officer. Prior to conducting the study, farmers received a comprehensive explanation of the study's purpose, potential risks and benefits, and their verbal consent was obtained. To ensure the well-being of the animals, restraint methods that minimize stress, discomfort and harm, using the least force necessary was employed.

CHAPTER FOUR

4.0 RESULTS

4.1 Morphological identification of ticks

Of the 210 cattle examined, 201 (95.7%) were found to be infested with one or more ticks. A total of 702 ticks were found attached to cattle at the time of sampling from the five parishes in Itek sub-county. The adults were 627 (188 females, 439 males), 68 nymphs and 7 larvae. Additionally, 46 (7.34%) were engorged, 32 (5.1%) fully engorged, 14 (2.23%) partially engorged and 535 (85.33%) were not engorged of the adult ticks collected.

On analysis, four species of ticks in the two genera, *Rhipicephalus* and *Amblyomma* were identified. The majority of them 52% (365) were *Amblyomma* species and the rest 48% (337) were *Rhipicephalus* species.

In *Rhipicephalus* genera; *Rhipicephalus appendiculatus* was the most prevalent 33.97% (213), followed by *Rhipicephalus decoloratus* 11.16% (70) and the least being *Rhipicephalus evertsi* 0.16% (1/627) and all ticks in *Amblyomma* genera were *Amblyomma variegatum* at a prevalence of 54.71% (343) of the total adult ticks examined.

Table 1. Proportion of tick species identified from cattle in Itek sub-county

Tick species	Number of ticks collected (percentage)
Rhipicephalus species	
<i>Rhipicephalus appendiculatus</i>	213 (33.97)
<i>Rhipicephalus decoloratus</i>	70 (11.16)
<i>Rhipicephalus evertsi</i>	1 (0.16)
Amblyomma species	
<i>Amblyomma variegatum</i>	343 (54.71)
TOTAL	627 (100)

4.2 Tick species distribution in Itek Sub-county

Table 2 illustrates the distribution of tick species across various parishes in Itek sub-county. The table highlights the number of *Rhipicephalus appendiculatus*, *Rhipicephalus decoloratus*, *Rhipicephalus evertsi*, and *Amblyomma variegatum* among sampled cattle. Notably, *Amblyomma variegatum* is most abundant in Olilo parish, while *Rhipicephalus appendiculatus* shows significant presence in Alebere and *Rhipicephalus evertsi* was only found in Olilo parish.

Table 2. Number of tick species per parish in the Itek sub-county

Tick species	Number of tick species per parish (Number of cattle sampled)				
	Olilo (60)	Ajia (40)	Tetyang (40)	Alebere (30)	Onywako (40)
<i>Rhipicephalus appendiculatus</i>	47	33	15	79	39
<i>Rhipicephalus decoloratus</i>	14	6	32	7	11
<i>Rhipicephalus evertsi</i>	1	0	0	0	0
<i>Amblyomma variegatum</i>	121	55	45	70	52
TOTAL	183	94	92	156	102

4.3 Prevalence of tick infestation per parish in Itek sub-county

Out of 5 parishes in Itek sub-county, Ajia had the highest prevalence of 100% and the least being Tetyang with 92.5%.

Table 3. Prevalence of tick infestation per parish in Itek sub-county

Parish	Total number of animals inspected	Tick-infested Animals	Prevalence (%)
Olilo	60	57	95
Ajia	40	40	100
Onywako	40	38	95
Alebere	30	29	96.7
Tetyang	40	37	92.5

4.4 Sex distribution of tick species in Itek sub-county

Figure 16 presents the sex distribution of various tick species collected from cattle. The data highlights a significant predominance of male ticks across species, particularly in *Amblyomma variegatum*, which shows the highest male count. *Rhipicephalus appendiculatus* also demonstrates a considerable male presence, while *Rhipicephalus evertsi* and *Rhipicephalus (Boophilus) decoloratus* are largely composed of female ticks.

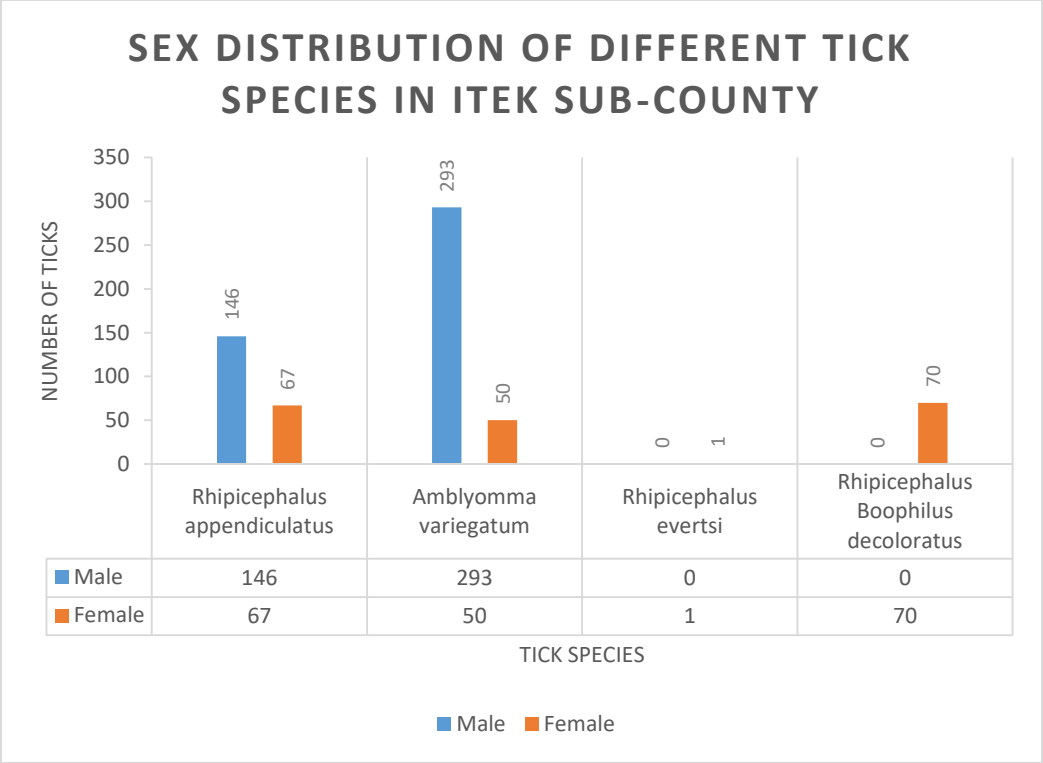


Figure 16. Sex distribution of different species of ticks infesting cattle in Itek sub-county

4.5 Level of engorgement of the tick species

Figure 17 below provides a breakdown of engorgement status for each tick species identified on cattle. The data reveals a notable prevalence of non-engorged ticks among *Amblyomma variegatum*, while *Rhipicephalus decoloratus* displays a balanced mix of engorgement levels, with a considerable number fully engorged. *Rhipicephalus appendiculatus* mainly consists of not-engorged ticks, and *Rhipicephalus evertsi* shows negligible feeding activity

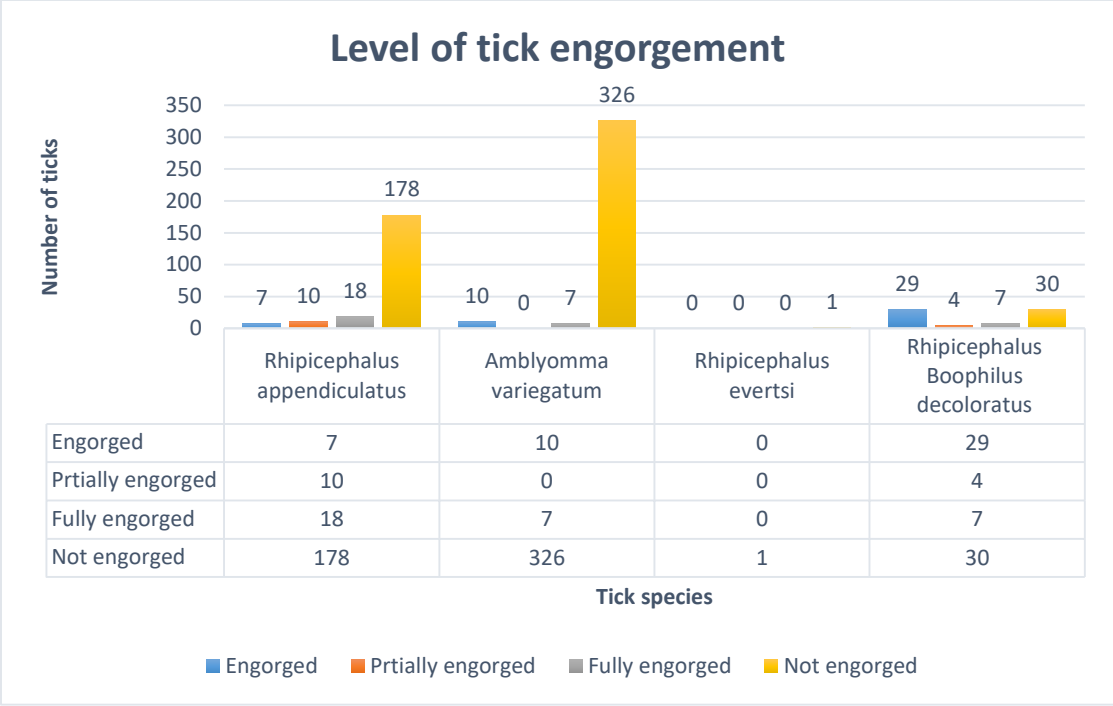


Figure 17. Level of engorgement of different species of ticks collected from Itek sub-county

CHAPTER FIVE

5.0 DISCUSSION

The main purpose of this study was to document the diversity of species of ticks infesting cattle in Itek sub-county, Lira District, Northern Uganda. In total, four (4) tick species were identified. *Amblyomma* species was the most abundant, *Amblyomma variegatum* was the commonest tick species, followed by *Rhipicephalus* species, of which *Rhipicephalus appendiculatus* was the second abundant tick species in the sub-county. This is in disagreement with what Muhanguzi et al., (2020) and Etiang et al., (2024) found out. They reported that *Rhipicephalus appendiculatus* was the most abundant species. The difference in these results is probably because of the different seasons and places of tick collection. However the study was in consistence with (Corrigan et al., 2023) which reported that *Amblyomma variegatum* was the most prevalent in Katakwi district during dry season.

Solomon et al. (2001) stated that *Amblyomma variegatum* causes the greatest damage to hides and skins because of its long mouth part, which renders the commodity valueless on world market if the infestation was high. The abundance of *Amblyomma variegatum* in study area may be associated with massive damage to hides and skin. Furthermore, ulcers caused by this tick species can be favorable sites for secondary bacterial infection like *Dermatophilus congolensis* and other pyogenic organism.

These findings show that tick infestation on cattle is mainly dominated by the species of these two genera, and amongst these species, are vectors of major diseases of economic importance in the region. This explains the high morbidity and mortality rates reportedly caused by mainly ECF and anaplasmosis in the sub-county as it was also described in study Byaruhanga, Oosthuizen, et al, (2015) in North Eastern uganda.

Being a three- host tick and a short lifecycle, enables *Rhipicephalus appendiculatus* to avoid desiccation (Troughton & Levin, 2007). According to (A. R. Walker et al., 2014) , *Rhipicephalus appendiculatus* covers a more eastern and central African distribution, ranging from South Sudan to the northern parts of South Africa, while *Rhipicephalus evertsi* is more widespread including parts of western Africa.

Rhipicephalus decoloratus is widely distributed in most areas south of the Sahara, typically within grasslands and wooded areas used as pasture for cattle (Walker et al., 2014). *Rhipicephalus decoloratus* was the second most abundant of *Rhipicephalus* species. It being a one-host tick (A. R. Walker et al., 2014), *Rhipicephalus decoloratus* completes all its growth stages on a single host animal. The availability of these host animals promotes the tick's multiplication and rapid development. This species can survive across various elevations and thrive in both wet and dry conditions year-round (Makwarela et al., 2023). However, unlike in the recent findings of Muhanguzi et al., (2020) who concluded that *Rhipicephalus decoloratus* is being displaced by *Rhipicephalus microplus* in Serere district, this was not found in the current study in Itek sub-county, Lira district.

Amblyomma variegatum, the African bont tick was found in all parishes in the sub county where ticks were collected. This finding is consistent with previous studies that reported the presence of *Amblyomma variegatum*, throughout the year, in the entire country spanning different vegetation and microclimates (Matthysee and Colbo, 1987; Balinandi et al., 2020). The presence of many animals in the area facilitated its multiplication since it is a three-host tick (Anderson, 2002). According to Walker et al. (2014), *Amblyomma variegatum* in most subtropical and tropical Africa has a northern borderline that stretch from Senegal to Ethiopia and a southern borderline that covers parts of Zambia, Namibia, Mozambique and Zambia.

Majority of ticks across the species are not engorged, with *Rhipicephalus appendiculatus* and *Amblyomma variegatum* having the highest numbers of non-engorged ticks. Fully engorged ticks were relatively few across all species, with *Rhipicephalus decoloratus* having a slightly higher count of fully engorged ticks compared to the other species. This pattern is consistent with observations in tick ecology, where non-engorged ticks are more likely to be found on the host because engorged females detach after feeding to lay eggs in the environment (Walker et al., 2003). Additionally, the presence of partially and fully engorged ticks suggests active feeding and the potential for disease transmission during the study period.

The male to female sex ratio of all tick species except *Rhipicephalus decoloratus* and *Rhipicephalus evertsi* is greater than suggesting that the male outnumbered the female ticks. Huruma et al. (2015) also reported a higher male to female sex ratio of tick species in and around Sebeta town, Ethiopia. The male to female ratios of *Rhipicephalus decoloratus*, *Rhipicephalus*

appendiculatus and *Amblyomma variegatum* in the present study is similar to previous report by (Gedilu et al., 2014). The more plausible explanation why male ticks outnumbered the females could be due to the fact that fully engorged female tick drops off to the ground to lay eggs while male tend to remain on the host up to several months to continue feeding and mating with other females on the host before dropping off (Solomon et al., 2001). The females of *Rhipicephalus decoloratus* were the only one found with no male in this study probably due to small size of males, which may not be seen during collection (Tessema & Gashaw, 2010).

In this study, the lower infestation rates of immature stages of ticks on cattle compared to adult stages was registered. This is because the pre-adult stages typically exhibit low host specificity (Walker et al., 2003). Furthermore, the method used to collect ticks was not the most efficient for immature stages. More larvae and nymphs would probably have been collected had the cattle hair been combed. In previous study, a poor correlation between larval and adult results has also been observed and was due to the differential efficiency in sampling method (Randolph & Rogers, 1997).

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

There is a high diversity and abundance of the tick species (4) infesting cattle in Itek sub-county in Lira district. The proportion of tick-infested cattle in Itek sub-county is 95.7%. *Amblyomma variegatum* was the most abundant species, followed by *Rhipicephalus appendiculatus*. There was no invasive species of ticks identified in the current study. This study demonstrates high tick infestation rates in cattle by different tick species with potential to transmit several tick-borne diseases including zoonotic pathogens in Itek sub-county, Lira district.

6.2 Recommendations

Tick occurrence and prevalence data are useful in the design of targeted tick control strategies which are affordable and environmentally friendly. And also since the present study only investigated tick species infesting cattle, the general composition of tick species being hosted by other domestic and wild animals in Uganda is poorly described, therefore extensive studies have to be done to capture the definite ecological ranges of tick species around the region and Uganda at large.

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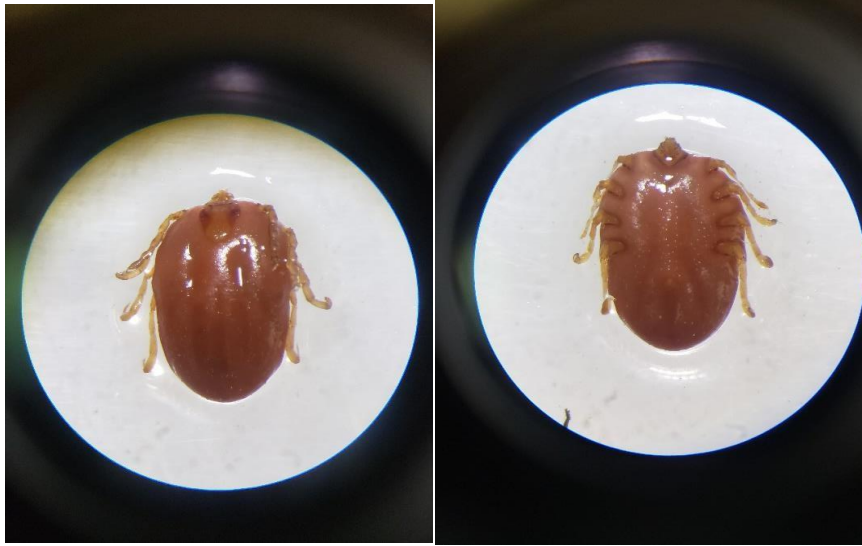
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APPENDICES



Rhipicephalus decoloratus



Amblyomma variegatum



Rhipicephalus appendiculatus



Rhipicephalus evertsi