



Study prevalence of hard tick infestation at South Western Kafa Zone Cheta Woreda

Dechu Jobir^{1*}, Mulatu Gure²

¹College of Agriculture and Natural Resources Department of Animal and Range Science, Dilla University, Dilla, Ethiopia. E-mail- dechujobir@gmail.com

²Hadiya Zone, Shone Town administration Livestock and fisher resource, Shone, Ethiopia.

E-mail: mulatugure@gmail.com

*Corresponding author; Dechu Jobir, College of Agriculture and Natural Resources, Department of Animal and Range Science, Dilla University, Dilla, Ethiopia, Ethiopia.

Abstract

Across sectional study was conducted in ChetaWoreda, kaffa zone from november 2019 to June 2020 G,C the estimate the prevalence of major ixodide ticks genera on cattle and identify the prevalence of tick genera level. Study animals were 'selected and randomly out of total cattle examined 291(14.7 %) were found to be infested by one or more ticks about 2024 adult ticks were collected from the animal body parts preserved with 70% alcohol and were identified to genera level by using stereomicroscope. From the total of ticks collected four genera's namely Amblyomma, Boophilus, Rhipicephalus and Hayalomma were identified and account for 34.9, 26.6 and 19.12.% and 19.21% respectively. From different variation (sex, age bread and body condition).body condition was statistical significantly with tick infestation($p<0.05$).the prevalence of tick infestation was found higher in poor body condition animal 100% medium body condition were 74% and good body condition animal 69.5% respectively, expressed as percentage. It has also exident that favorable predilection site of Amblyomma tick were external body parts and dewalp region Boophilus preferred develop under sternium ,bell, leg, head and dewalp ,Rhipicephalus had strong affinity for sternum, develop udder and scrotum ,under effective tick control program should be formulated and implemented based on the pattern of factor for tick responsible for their distribution.

Keywords: Cattle, prevalence , risk factor, tick, Cheta

1. Introduction

1.1 Background of the Study

Ethiopia is believed to have the largest livestock population in Africa. This livestock sector has been contributing considerable portion to the economy of the country, and still promising to rally round the economic development of the country (CSA, 2013). In Ethiopia, livestock production remains crucial and

represents a major asset among resource-poor small holder farmers by providing milk, meat, skin, manure and traction force (Lemma, et al., 2001).The contribution of livestock to the national economy particularly with regard to foreign currency earnings is through exploration of live animal, meat and skin and hides (MoARD , 2008).

Hides and skin account for 12-16% of the total value of exports from Ethiopia (Asfawet al., 1997). The current utilization of hides and skins in Ethiopia is estimated to be 48% for cattle, 75% for goats and 97% for sheep with exacted off take rate of 33, 35 and 7% for sheep, goats and cattle, respectively (Mahamudet al., 2000). Though hides and skin are important source of export income its contribution to the national economy may be for below the expected potential. This is due to external parasite like ticks and other (Bayou et al., 1998)

Ticks are one of the external parasites; the economic impact of tick infestation is worldwide. In 1984, FAO estimated the global cost of ixodidae tick infestation to be 7 billion us dollar annually. In Ethiopia tick and tick borne diseases cause considerable losses to the livestock economy ranking third among the prevalently parasitic disease after trypanosomes and end parasites. Ticks are directly or indirectly involved in causing substantial financial losses to livestock industry of Ethiopia accounts for 75% of the animal exports. A conservative estimate of 1 million birr loss annually was made through rejection and down grading of hides and skins in Ethiopia. Apart from a direct effect of tick infestation on animal production and productivity, ticks are inviolably efficient vectors of pathogens (protozoa, viruses, bacteria and rickettsia) to man and domestic animals (Peg rametal., 1981; Rahbariet al., 2009).

Ticks and the disease they transmit are widely distributed throughout the world, particularly in tropical and sub tropical regions. It has been estimated that 80% of the world's cattle population is exposed to ticks infestation (FAO, 1984). In different regions, ticks have developed resistance to many classes of acaricide including organophosphates, formamidines(amitraz) and other acaricide group. Target site mutations are the most common resistance mechanism observed, but there are examples of metabolic mechanism (Furlanget al., 2001)

Tick infestation has been known to cause a great deal of loss or reduction of productivity by influencing the performance and qualities of the animal yield. A lack of standardized techniques for diagnosing acaricide resistance appears to be the main difficulty and in creating and maintaining a tick resistance monitoring system. The issue of diagnosis of resistance has attracted the attention of government, international institution and academic organization (Hasenet al., 1999).

According to (walker et al., 2003) ticks that are considered to be most important to the health of domestic animals in Africa comprise about forty species. Among these the most important tick species in Ethiopia cattle are *Amblyommagemma*, *A. veriegatum*, *A. cohaerens*, *A. lepidum* (Mekonnenet al., 2001), *Boophilusdecoloratus*, (Mekonnenet al., 1988), *B. annulatus* (Haile et al., 1987), *Rhipicaphaluspulchekkus*, *R. pravus*, *R. evertsevertis*, *R. praetextatus*, *R. bergeoni*, *R.muhasmae*, *R.lumulatus*, *R.simus* (Regassaet al., 2001). *Hyalommaimpelatum*(Bekeleet al., 1996), *Haemaphysalisaciculifer* and *Haemaphysisparmata* (Decastroet al., 1994). The country environmental condition and vegetation are highly conducive for ticks and tick born disease perpetuation (Pegramet al., 1981).

Acaricides used to control ticks on livestock or in the environment should be applied in such a manner that the ticks will be killed, the treatment will not harm livestock or applicators, the tissues of treated animals will not contain illegal residues, and the environment will not be adversely affected (Shaw et al., 1966).

1.2 Statement of the Problem

In view of the world wide spread, occurrence and host related factor nature, Tick has emerged as a major global and regional concern affecting all domestic animals and secondary infection is most prevalent in regions with indigenous cattle production. From the many parasitic problems of farm domestic animals, Tick is the most important parasite, which cause for the considerable host related risk in the cattle industry, mainly through reduced production of milk, meat and causing secondary infection. Tick parasite was shown to be the most important class Archnida, order Acarina, suborder ixodida families Ixodidae and Argasidae which affect south west Ethiopia live stock with distribution over all nations of the country with out exception. Chetaworeda has the larger live stock inventories from kaffa zone, including about 98612 cattle, 80448 sheep, 36569 goats, and 2673 equins with livestock ownership currently contributing to the livelihoods of an estimated 80% of the rural population. The live stock sector in Chetaworeda has substantial contribution to the economy however, parasitic disease like Tick cause significant economic problem by decreasing the productivity of cattle in addition to direct effect of the parasite. Although a number of the studies have been undertaken with regard to prevalence and evaluation of the host related risk due

to Tick parasite in different parts of countries, but no information has been done in southern west Ethiopia especially kaffa zone Chetaworeda.

Hence, the present study was designed to generate epidemiological information and estimate economic significance of Tick parasite in Chetaworeda. Tick born disease is a serious vector of pathogens to man and domestic animals (Pegram et al., 1981; Rahbori et al., 2009).

Due to the impact of this disease many people who live at rural area. Their life depends up on the livestock production affected by the retardation of animal growth, loss of milk and meat production, general by effecting the market causes. Also economic contribution of this sector toward the country development was decreased. Therefore the effect should be considered, studies and the appropriate recommendation. So the following basic question most is raises in order to investigate problem of the disease at the selected site in and around Chetaworeda.

To what existence does the tick infestation up on the cattle?

To what percents do the economic lose occurred due to loss of production of milk and meat as well as rejects of hides?

Is there infestation of ticks up on the cattle?

1.3 Objective of the Study

1.3.1 General Objective of the Study

The overall objective of this study is to assess out the major factors for the problem of tick infestation on the cattle in the study area.

1.3.2 Specific Objectives of the Study

The specific objectives of this study area

To assess the prevalence of ticks in the study area

To assess the major factors that lead to tick infestation of the cattle.

To identify the identification of ticks and tick infestation on the cattle.in the study area

To assess the economic loss due to tick infestation.

1.4 Research Questions

The study tried to deal with answering the following research questions.

What are the major risk factors for tick infestation on the cattle?

How can we identify the tick and tick infestation ?

How we can minimize the main cause of tick infestation on the cattle?

How to reduce the economic loss in case of the tick infestation?

What are the control method of the tick and tick infestation?

1.5 The Scope of the Study

This study was conducted in the south region Kaffa Zone, Chetaworeda, KaffaZone,southern National Regional State,509 km south west of Adis Ababa and 800 km far from region town hawassa.Thisworeda has alatitude and longitude of 07°35'N 36°33'E.The 2007 national census reported atotal population for this Woreda of 43,400 of whom 21,570 were men and 21,830 were women.so the study was conducted especially in the three selected kebele within woreda; they are Duba, Kushasha, and Shakakebeles. The study is focused on the problem of tick infestation cases in reference to three selected kebele in cattle rearing farmers of 2011 E.C.

1.6. Significant of the Study

Many people of the woreda who live in a rural area, where as their life also more depends up on the livestock production and/or productivity, 7% that this time more their income lost to treat their cattle's in case of tick infestation and tick born disease, So that, the purpose of this study is focused on to creating awareness about the extent of tick infestation and tick born diseases to the community group especially farmers, society and to the concerning governmental body of other stockholder. That helps to them easily to understand and identity the problem of ticks. It is also helps to set the appropriate controlling methods and increased their annually income from cattle's sources in the study area.

1.7 Limitation of the Study

The researcher has faced so many problems when this research had studied, and the researcher set the problems, like budget and related problem such as the absence of good awareness in the community people to give clear and good information during interview, lack of sufficient and well organized data, lack of document material and documentation. Lack of constraints money, financial problem for transport and

time related problems. In other hand these parasite cause host related risk is not only headache of cattle species, it is ecto parasites of man, wild and domesticated animals, but these study will be focused only in the cattle due to the shortage of time, financial constraints and the frightens to complicity of study. however by endeavor of me I was made to fill most of limitation and generate the required information for the study.

1.8. Organization of the Paper

The researcher completed the data collection and reference materials for the study and also organized the whole content of research paper in to five chapters. These are: chapter one Introduction, statement of problem, objective of the study, research question, scope of the study, significance of the study, limitation of the study and organization of the research paper, chapter two contains about review of literature of the study, when we come to chapter three: it deals about the design and methodology of the study and chapter four gives emphasis on the analysis of data, result and discussions and finally chapter five tells about summary conclusion and recommendation and also references

2. Review of Literature

2.1 Evolution of Ticks

Studies tick evolution previously placed emphasis on the host, arguing that the main driving forces of tick's evolution are host specificity. There is, however, hypothesis suggesting that ticks evolved along their hosts and that primitive ticks had ancient hosts (Oliveret al., 1989).

However, various constituents in the ecology of tick would appear to play a significant role in their evolution: (Kim et al., 1983) and earlier researchers also hypothesized that modification in tick structure in different stages of the life cycle took place in association with the evolution and specialization of particular hosts which were parasitized by each stage. These alteration and adaptation played a major role in evolution. That was the initiation of tick classification.

2.2 Classification of Ticks

The taxonomic resemblance referred to as ticks, in a relatively small group, which are related to animals' such as insects and spiders and are all without spine,

comprising approximately 860 species (Barker et al., 2004). Within the vast phylum arthropod a, ticks and their allies can be separate from insects and other mandibulate forms (Cantipedes, Maillipedes and CrustanCeans) in to the sub phylum chelicerate'on the basis of the presence of an anterior pair of chelicerae that function as tropic appendages (Kaiser et al., 1987). Since .this group of ecto-parasites is more closely related to spider and scorpions, it has been placed in the class arachnida, order acari, which also includes alltaxas commonly referred to as mites. The sub-order of ixodida contains the hard and soft ticks of the families'ixodidae and argasidae (Beaver et al., 1984).

The super family of ixodidae includes three families of ticks, namely ixodidae, argasidae and nuttalielidae of the three families the ixodidae (hard ticks) is the largest and important groups

2.3 General Morphology of Ticks

This are classified in to two families, argasidae or "soft tick" and ixodidae or "hard tick" which differs considerably in their structule and biology (Urquhart et al., 1996).

Ixodid ticks are relatively large ranging between 2 and 20mm in length and lost all of the external signs of body segmentation. The body of the unfed tick is flattened dorsoventrally being divided in to two sections (Bow man, 1999) that is the anterior gphantostoma or capitulum, the mouth part or fusion of h~ and thorax, and posterior idiosoma which bears the legs (Wall et al., 1997).

The conscutum covers nearly the entire dorsal surface of the adult male ixodid ticks and so limits the amount of blood that can be infested. Female ticks, larvae and nymphs also have ascutum but it covers only the anterior one-third of the dorsum thus allowing the ticks to expand when engorging (Golezardyet al., 2006). Ixodidae ticks feed slowly and attach to their hosts for long periods, depending up on the stage of development (Walker et al., 2003).

In addition to that the ixodid ticks have long mouth parts (Amblyomma and Hyalomma) and short mouth parts (Boophilus, Rhipicephalus and Haemaphisalis) which projecting towards the of development (Walker et al., 2003). Anterior end of tick. All ixodid ticks except Haemaphisalis and ixodes have eyes which located dorsally. The anal grooves of ixodid ticks pass

to the posterior end of anus except for the ixodes which passes anteriorly like that of soft ticks. There are some sorts of coloration on Amblyomma, Hyalomma and in few Rhipicephalus general (Uraguahartet al., 1996).

Male ixodid ticks are smaller than females. Males imbibe relatively little blood when they feed and show little increase in size. Ixodid ticks posse's asclerotized dorsal shield or plate on the idiosoma known as scutum. In males, the scutum covers the entire dorsal surface, where as in females scutum is relatively small to facilitate the size increase which occurs during feeding. The scutum is difficult to see a fully engorged female (Wall et al., 1997)

In some genera (Derma center and Amblyomma) the dorsal shield scutum is ornated silvery marking and is then said to be "ornate". In several genera, the dorsal shield of male is marked with "festoons" on the posterior border (Reed et al., 1961).

2.4 Life Cycle (Biology) of the Ixodid Ticks

2.4.1Biology of Ticks

Ticks are among the most significant blood-sucking arthropods and distributed worldwide. They transmit various pathogens that can cause disease and death in cattle. Ticks have several morphologic features and physiologic mechanisms that facilitate host selection, ingestion of vertebrate blood, mating, survival and reproduction (Berihun et al., 2016). Although the

natural history of ticks varies considerably among species, these arthropods are well-adapted to survive in tropical, temperate, and even subarctic habitats. Most ticks require three different hosts to complete one full cycle. These three-host ticks detach on completion of feeding, drop from the host, molt and wait for another host. The life cycle of tick involves according to feeding habitat and characteristic number of host individuals (Walker et al., 2003). In the hard ticks mating takes place on the host, except with Ixodes where it may also occur when the ticks are still on the vegetation. Male ticks remain on the host and will attempt to mate with many females where as they are feeding. The lifecycle of ticks (both Ixodids and Argasids) undergo four stages in their development (Figure 1); eggs, 6-legged larva, 8-legged nymph and adult (Walker et al., 2003). According to the numbers of hosts, Ixodids ticks are classified as onehost ticks, two-host ticks, three-host ticks and Argasids classified as multi-host ticks. In one-host ticks, all the parasitic stages (larva, nymph and adult) are on the same hosts; in two- host ticks, larva attach to one host, feed and molt to nymph stage and engorged, after which they detach and molt on the ground to adult; and in three-host ticks, the larva, nymph and adult attach to different hosts and all detach from the host after engorging, and molt on the ground. In multi-host ticks (Argasids), a large number of hosts are involved and it is common to have five molts, each completed after engorging and detaching from the hosts (Be Evans et al., 2000)



Figure-1: Life cycle of ticks (Walker et al., 2003)

2.5 Host Finding

All ticks spend most of their life cycle away from their hosts (without host), hiding either in soil and vegetation or in the nests of their hosts. So they need to be able to find hosts on which to feed. Ticks do this in several ways. Many ticks have the eggs and molting stages in soil or vegetation in the environment in which their hosts graze or hunt. The survival of a population of ticks depends on the presence of hosts suitable for reproduction by the adults. These hosts are known as maintenance hosts. These hosts are more limited in variety than the hosts on which larvae and nymphs of three-host ticks can survive. They are also more limited than those on which adults may attempt to feed but not necessarily survive. (Walker et al., 2003).

2.5.1. One-host tick

Cattle tick (*Boophilus*) is a single-host species. The parasitic stage of the tick life cycle (the stage spent on an animal) is spent entirely on a single host. The parasitic stage of the life cycle involves 3 phases; larvae, nymph and adult. Cattle ticks undergo a moult on the host between the larval, nymph and adult phases. During the moult, the tick sheds the previous skin or shell, in order to emerge into the next life stage (i.e. larvae moult into nymphs, and nymphs moult into adults). The parasitic stage begins when a larval tick climbs from the pasture onto an animal. The parasitic stage ends when a fully engorged female detaches from the animal and falls to the pasture (Jelalu et al., 2016).

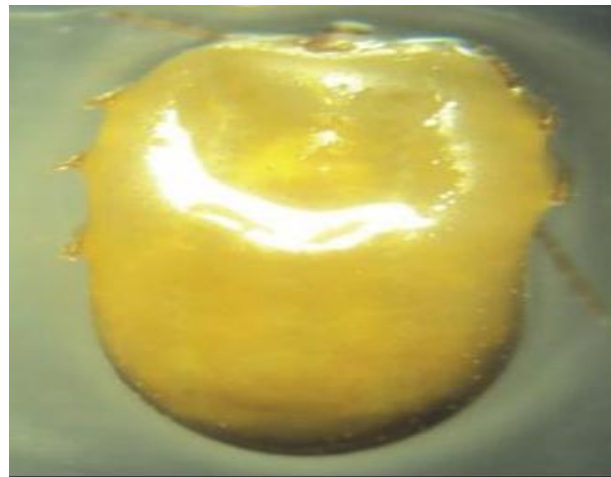


Figure 2: *Boophilus decoloratus* (Jelalu.,et al.,2016)

2.5.2. Two-host tick

In these species, the larval and nymph stages are spent on the same animal, but the nymph drops off to molt to the adult stage, which then seeks a final host. A few species in the genera *Hyalomma* and *Rhipicephalus* are experienced two-host life cycle (Nateneal et al., 2015).

2.5.3. Three-host tick

Three-host tick life-cycle showing relative sizes of the instars and unfed and engorged ticks, approximately 4 times life size (*Rhipicephalus appendiculatus*). The fed male has not expanded but shows a caudal appendage.

Hypostome and palps, ventral view showing palps spread out to enable only the hypostome and chelicerae to penetrate the host. Engorged nymph of *Amblyomma* compared with engorged female of *Boophilus* sub-genus, ventral view. The two species shown may occur at the same time on the same cattle. They can be distinguished by the shape of their mouthparts (Walker et al., 2003).



Figure 3: *Amblyomma variegatum* (Jelalu., et al., 2016)

2.6 Epidemiology of Ticks

2.6.1 Host relationship

Some ticks live in open environments and crawl onto vegetation to wait for their hosts to pass. This is a type of ambush and the behavior of waiting on vegetation is called questing. Thus in general such as *Rhipicephalus*, *Haemaphysalis* and *Ixodes* the larvae, nymphs and adults will quest on vegetation. The tick grabs onto the host using their front legs and crawl over the skin to find a suitable place to attach and feed. Adult tick of genera *Amblyomma* and *Hyalomma* are active hunters, they run across the ground after nearby hosts (Abdela et al., 2016).

2.7. Pathogenic Effects of Ticks and Tick-Borne Diseases

Infested cattle lose condition due to 'tick worry' and loss of blood. Heavy infestations can kill calves and even adult cattle. Animals in poor condition are especially vulnerable (susceptible). Previously unexposed cattle become heavily infested until they build up a degree of resistance. *Bos Indicus* (tropical breeds of cattle) and their crosses, develop a greater degree of resistance than *Bos Taurus* (British and European breeds of cattle). Ticks transmit the organisms that cause tick fever, which is a serious blood parasite disease of cattle. It also causes lower reproductive & productive efficiency of their hosts. Economic loss through morbidity & mortality of animal hosts and control cost. Hides of infested cattle are damaged by tick bites, reducing their value. In severe cases the hides may be unsalable (Walker 2004). Especially ticks with long mouthparts cause considerable direct

damage to hides of cattle. Secondary infections can cause septic wounds or abscesses, and lesions on the teats of cows may affect milk production. Ticks also have adverse effect on livestock in several ways and parasitize a wide range of vertebrate hosts and transmit a wide variety of pathogenic agents than any other group of arthropod (Walker et al., 2003).

2.8 Consequences of Infestation

Ticks present three main dangers to their hosts: the physical damage from the bite itself, other systemic effects of the tick's saliva and transmission of infectious diseases. When ticks attach themselves to the host in the first stage of feeding, they cut the skin with their mouthparts and cause damage to tissues and capillaries. Host reactions, such as mast cell degranulation leading to histamine release and inflammatory cell infiltration, further contribute to tissue damage. This tissue damage tends to be quite painful and may result in secondary bacterial infections. (Shearer, 2010).

2.8.1 Direct effect of ticks

Ticks are responsible for direct damage to livestock through their feeding habits. The damage is manifested as hide damage, damage to udders, teats and scrotum, myiasis due to infestation of damaged sites by maggots and secondary microbial infection. Ticks bite can directly debilitate to domestic animals, causing mechanical damage, irritation, inflammation and hypersensitivity (Abdela et al., 2016).

Feeding by large numbers of ticks causes reduction in live weight and anemia among domestic animals, while tick bites also reduce the quality of hides. Apart from irritation or anemia in case of heavy infestations, tick can cause severe dermatitis. These parasites generate direct effects in cattle in terms of milk production and reduce weight gain (Tiki and Addis, 2011).

2.8.2 Physical effect of tick

Ticks are attached to the body for a blood meal and may cause irritation and serious physical damages to livestock. Included tick worry, irritation, unrest and weight loss due to massive infestation of ticks. The direct injury to hides due to tick bites, loss of blood due to the feeding of ticks. When cattle are heavily infested, ticks can be found anywhere on the body of animals (Walker, 2004).

2.8.3 Vector of pathogens.

The main diseases transmitted by ticks to livestock are anaplasmosis (ruminants), babesiosis (ruminants, horses, and dogs), theileriosis (ruminants, horses) and cowdriosis (ruminants). Ehrlichiosis in ruminants and dogs is also important in certain tropical and subtropical regions. Endemic stability can often be achieved, especially in indigenous livestock and even East Coast fever may cause no more than slightly increased calf mortality in local zebu in fully endemic areas. *A. variegatum* (vector of *Cowdriaruminantium* and *Theileriamutans*) and *Boophilus* species (vector of *Anaplasma marginale* and *Babesia bigemina*) are the most widespread ticks in Ethiopia. *R. evertsievertsi* is also known to occur across different ecological zones of the country serving as a vector for *Babesia bigemina* in cattle, (Ferede, et al., 2010). Estimates of economic losses due to ticks and tick-borne diseases are often little more than educated guesses. Any form of control in local resistant livestock is not always cost-effective, whereas intensive and expensive control measures are often required for valuable exotic breeds (Latif and Walker, 2004).

Babesiosis

Babesiosis is an emerging, tick-transmitted, zoonotic disease caused by hematotropic parasites of the genus *Babesia*. *Babesia* parasites (and those of the closely related genus *Theileria*) are some of the most ubiquitous and widespread blood parasites in the world, second only to the trypanosomes, and

consequently have considerable worldwide economic, medical, and veterinary impact. The parasites are intraerythrocytic and are commonly called piroplasms due to the pear-shaped forms found within infected red blood cells. The major species of *Babesia* which causes bovine babesiosis are *B. bovis*, *B. bigemina*, *B. divergens* and *B. major*. *Babesia* parasites can be transmitted transovarially between tick generations in the case of *Ixodes*, surviving up to 4 years without a vertebrate host. *Babesia* may also be transmitted by fomites and mechanical vectors contaminated by infected Mary et al., (1996)

Anaplasmosis

Anaplasmosis, formerly known as gall sickness, traditionally refers to a disease of ruminants caused by intraerythrocytic organisms of the order of genus *Anaplasma*. Anaplasmosis is a vector borne infectious blood disease in cattle caused by the rickettsial parasites. It is not contagious, but numerous species of tick vectors (*Boophilus*, *Dermacentor*, *Rhipicephalus*, *Ixodes* and *Hyalomma*) can transmit *Anaplasma* species. It causes outbreaks in a herd, which can lead to the death of adult cattle. Other economic losses include abortions, decreased weight gain, bull infertility and treatment costs. Although many outbreaks of anaplasmosis occur in the spring and summer, they can occur at any time of the year (Latif and Walker, 2004).

Theileriosis

Theileriosis results from infection with protozoa in the genus *Theileria* of the suborder Piroplasmorina. *Theileria* species are obligate intracellular parasites. *Theileria* is considered to be spread by bush ticks and/or introduction of infected animals from an endemic area. It may also be spread via standard husbandry practices that include blood transfer (such as using needles on multiple animals) and across the placenta. Bush ticks only transfer *Theileria* to cattle, not other species (Vivian, 2010).

2.8.4 Tick bite paralysis

It is characterized by an acute ascending flaccid motor paralysis caused by the injection of a toxin by certain ticks while feeding. Examples are paralysis caused by the feeding of *Dermacentor andersoni*, sweating sickness caused by *Hyalomma truncatum*, tick toxicosis caused by *Rhipicephalus* species and other tick paralysis caused by *Ixodes holocyclus*. Effective

diagnosis of tick borne hemoparasitic diseases of ruminants is helpful to implement appropriate prevention and control strategies. Tick control, chemoprophylaxis and immuno-prophylaxis are the basic methods to control tick borne hemoparasitic diseases of ruminants. (Latif and Walker, 2004). Housing in tick proof buildings, Separate housing of cattle from others, Quarantine, Pasture spelling and rotational grazing, Manual removal of ticks, Clearance of vegetation, Use of acaricides, Use of biological control methods(e.g. birds, rodents, shrews, ants and spiders play some role in tick control measures), Breeding cattle for tick resistance, Ethno veterinary practices against ticks (e.g. Several plants and herbs have been shown to possess anti-tick insecticidal, growth inhibiting, anti- molting and repellent activity and Tick vaccine (Vivian, 2010).

2.9 Control Methods

2.9.1 Biological control methods of ticks

Ticks have numerous natural enemies, but only a few species have been evaluated as tick biocontrol agents. Some laboratory results suggest that several bacteria are pathogenic to ticks, but their mode of action and their potential value as bio-control agents remain to be determined (Jongejan, 1994). Natural enemies of ticks include insectivorous birds, parasitoid wasps, nematodes, *Bacillus thuringiensis* bacteria, and deuteromycetefungi (largely *Metarhizium isopliae* and *Beauveria bassiana*). The potential of each of these taxa as bio-control agents will be discussed in turn. Mammals and birds typically consume ticks during self-grooming. (Jongejan and Uilenberg, 1994)

2.9.2 Stock breeding and pasture management.

Cattle breeds indigenous to Africa, typically *Bos Indicus* or zebu have a good heritable ability to acquire natural resistance to the feeding of ticks. This characteristic can be used in breeding programmed to produce crosses with more productive exotic cattle of the *Bos Taurus* type which will give good resistance to ticks and good production (Latif and Walker, 2003).

2.9.3 Application of chemicals methods

The use of acaricides in the control of ticks has improved the viability of cattle farming in the tick infested areas. Ticks can be killed by dipping or spraying cattle with an appropriate chemical (acaricides). Ticks can develop resistance to acaricides (Blackwell science, 2001).

Dipping

In this method, animals are immersed in a dipping tub containing solution of chemicals. Infested cattle should be dipped in the organophosphate acaricide coumaphos (0.3% active ingredient). In general dipping vats provide a highly effective method of treating animals with acaricides for tick control (Latif and Walker, 2003).

Spray

The application of fluid acaricides to an animal by means of a spray has many advantages and has been successfully practiced for controlling ticks on most of the animals (Barnett, 1961). Spraying equipment is highly portable, and only small amounts of acaricides need to be mixed for a single application. However, spraying is generally less efficient in controlling ticks than immersion in a dipping vat because of problems associated with applying the acaricides thoroughly on all parts of the animal body (Walker et al., 2003)

Spot treatment or hand dressing

There are predilection sites for certain tick species on part of the body which are not effectively treated by spray or dips. The inner parts of the ear, under part of the tail, the tail brush and the areas between the teats and the legs in cattle with large udder are especially liable to escape treatment. The application of insecticides with aerosols and in oils, smears, and dusts by hand to limited body areas is time-consuming and laborious, but in certain instances it may be more effective and economical (in terms of cost) of acaricides than treating the entire animal (Blackwell science, 2001). The future of tick control in the communal areas. Indigenous Sanga and Zebu cattle which are predominantly reared by communal farmers have a high degree of tick and tick-borne disease resistance and require minimal tick control methods. This tick control method is suitable and cost effective (minimize) for usages, even farmers Kaur et al., (2015) Some other control methods and applications. Ticks are commonly controlled by using conventional synthetic acaricides, however it has certain drawbacks like high cost, non biodegradable, toxic to environment, left residuals in animal body and above all development of resistance in ticks. Therefore, the search for herbal alternatives is ongoing process and various researchers are exploring different genera of plants to find extracts with acaricidal properties that

can be used in association with or even as an alternative to synthetic.

Ticks are among the most significant blood-sucking arthropods and distributed worldwide. They transmit various pathogens that can cause disease and death in cattle. Ticks have several morphologic features and physiologic mechanisms that facilitate host selection, ingestion of vertebrate blood, mating, survival and reproduction (Berihun et al., 2016). Although the natural history of ticks varies considerably among species, these arthropods are well-adapted to survive in tropical, temperate, and even subarctic habitats. Most ticks require three different hosts to complete one full cycle. These three-host ticks detach on completion of feeding, drop from the host, molt and wait for another host. The life cycle of tick involves according to feeding habitat and characteristic number of host individuals (Walker et al., 2003). In the hard ticks mating takes place on the host, except with Ixodes where it may also occur when the ticks are still on the vegetation. Male ticks remain on the host and will attempt to mate with many females where as they are feeding. The lifecycle of ticks (both Ixodids and Argasids) undergo four stages in their development (Figure 1); eggs, 6-legged larva, 8-legged nymph and adult (Walker et al., 2003). According to the numbers of hosts, Ixodids ticks are classified as one-host ticks, two-host ticks, three-host ticks and Argasids classified as multi-host ticks. In one-host ticks, all the parasitic stages (larva, nymph and adult) are on the same hosts; in two-host ticks, larva attach to one host, feed and molt to nymph stage and engorged, after which they detach and molt on the ground to adult; and in three-host ticks, the larva, nymph and adult attach to different hosts and all detach from the host after engorging, and molt on the ground. In multi-host ticks (Argasids), a large number of hosts are involved and it is common to have five molts, each completed after engorging and detaching from the hosts (Be Evans et al., 2000).

3. Materials and Methods

3.1 Description of the Study Area

The study was conducted in chetaworeda and the surrounding rural kebel and clusters, kaffazone, Southern Nations Nationalities and Peoples Region. The town is found in 509 Km away from Adis Ababa, The capital city of Ethiopia. The total area of the woreda is 77,774 hr wide it is found in 5% kola, 80% woinadega and 15% dega ecological zone with an

altitude of 500-2500 Km above sea level and annual temperature ranging from 21 -25 annual rain fall between 1200-1600mm, the rain fall patterns are characterized as low and erratic. Chetaworeda is surrounded by 16 rural kebel, which is divided into five clusters including more than three kebel each, i.e. Adisalem, Diya, Kolla, Booba and Kora. It has the total population of 43,400 from these 6455 households, 36,945 the household members. The study was employed from the beginning of February to the end of April, 2019, three months period.

3.2 The Study population

The study population consisted of cattle which were brought from different cattle were categorized under different groups different age young, adult and old and based on management practice categorized grazing or extensive and intensive were cattle of any age, sex, and body condition scores found in the area. Kushasha, Shaka and Duuba randomly selected kebeles of Chetaworeda. The study populations were constituted local and cross breeds that were available in the study area. The management type of animals was also considered.

3.3 Study Design

A cross sectional study was conducted February to the end of April, 2019, three months period to estimate the prevalence of ticks, identification the major ticks, their predilection sites of tick, and tick burden in different age groups, breeds body conditions scores, sex of animal and different areas in the woreda. All the animal selected as sampling unit were checked for any tick infestation based up on the numbers of ticks found on the animal and the study record period. Ticks were collected from ears, dewlaps, belly/ flank, udder/ scrotum, fore/ hind legs, perineum and tails in the separated sample bottles with 70% ethyl alcohol (ethanol).

3.3.1 Questionary survey

The data for this study were collected from primary and secondary data sources. The primary sources of data include questionnaires, interview and observation as well as focus group discussion. All questionnaires are arranged in sequential manner that helps for discussion and analysis as well as respondents. The researcher prepared questions to 40 model farmers selected purposively for total number of 50 and the

total number of 3 selected by using random sampling techniques. Among those 12 are females. The secondary data were collected by referring the woreda recorded document such as unpublished and published research resource and check list from recorded in woreda agricultural office like case books concerning the general case on the tick infestation problem on the cattle.

3.4 Sample Size

Determination and Sampling Method

The sampling method employed to select the study animal was simple random sampling method and multi stage sampling strategy was used to determine the three study kebeles from the whole 3 kebeles in the woreda the total 384 cattle were selected simple random by and the sample size was determined by using the formula given in (Thrusfield et al., 1995). The expected prevalence of ixodidae ticks of cattle in Chetaworeda was assumed as 50% since there were no known researches conducted in the study area. The parameters used were 95% confidence interval and 5% desired level of precision. By substituting these values in the formula, the sample size taken was $n=384$

$$N = \frac{1.96^2 p \exp(1 - P_{\exp})}{D^2}$$

Where:

(N = Required Sample

P_{\exp} = Expected Prevalence

D = Desired Absolute Precision)

3.5 Tick Collection, Identification and Count

The ticks sample was collected from different randomly selected cattle. The entire body surface of the animal was examined thoroughly and all visible adult ticks were collected from half-body on alternative sides. Ticks were removed carefully and gently in a horizontal pull to the body surface. The collected ticks were preserved in universal bottles

(Petridish) containing 70% ethyl alcohol and labeled with the animal identification and predication site, age, sex, and data of collection. The specimens were transported to the parasitological laboratory of the school of veterinary medicine of Hawassa University for counting and identification. The ticks were counted and subsequently identified to genus and species level by using stereomicroscope, according to standard identification keys given by (Walker et al., 2003). The half-body tick counts of cattle were doubled to obtain the whole body tick burdens. During examination of the selected animals for tick infestation, the age, sex, body condition score, breed and kebele of the sampled animals were recorded on a special format designed for this purpose. During the study, distribution of ticks and total count of each tick genera were done.

3.6 Data Analysis

The data were entered and managed in Microsoft-excel. The overall prevalence of tick was determined by dividing the number of positive animals by total sample size, and was expressed as percentage. If there was a statistically significant association in tick infestation between ages, sex, kebeles, and season and body conditions of the animals. There was no significant difference between occurrences of tick infestation in study kebeles.

4. Results

4.1 Result

4.1.1 Overall Prevalence of Tick Infestation

In the current study out of the total of 384 cattle examined for the presence of ticks, 291 (75.5%) were found to be infested with varying number of tick genera. Slightly higher prevalence was recorded in Kushasha (85%) and lower prevalence was recorded in Duubakebele (68.7%).

Table 1 Prevalence of Tick Infestation in Cattle by Kebele

Study area	No of animals Examined	No of animals Infested	Prevalence %
Kushasha	128	94	73.4%
ShakA	128	88	68.7%
Duuba	128	109	85%
Total	384	291	75.7%

4.1.2 Prevalence of Tick Infestation in Cattle by Kebele

The prevalence of tick infestation in the present revealed that the occurrence of tick infestation both animals were not significantly different (table 2).

Table 2: Prevalence of tick infestation in cattle by Sex

Sex value	No of animals Examined	No of animals infested	Prevalence%
Male	135	101	74.8%
Female	249	190	76.3%
Total	384	291	75.8%

4.1.3 Prevalence of Tick Infestation in Cattle by Body Condition

Statistical analysis was performed to determine the relationship between the development animals with

different body conditions and it shows that there was statistically significant variation between body conditions of animals. In the current study, higher prevalence was in very fat animal (obesity) than animal (medium baly) animas table 3

Table 3: Prevalence of tick infestation in cattle by body condition

Body condition	No of animals Examined	No of positive animals	Prevalence %
Good	46	32	69.5%
Medium	306	227	74%
Poor	32	32	100%
Total	384	291	75.7%

4.1.4 Prevalence of Tick Infestation in Cattle by Age

The association between tick infestation and age of animals were assessed statistically. According to the present study findings, there was statistically

significant association between age of the animal and level of tick infestation.

Table 4: Prevalence of tick infestation in cattle by age

Age	of animals examined	of positive animals	prevalence%
Young	127	76	59.8%
Adult	194	153	78.8%
Old	63	62	98.4%
Total	384	291	75.7%

4.1.5 Prevalence of Tick Infestation in Cattle by Season

During this study samples were collected both in dry and wet season of the year and there was higher

prevalence of tick infestation in wet season (77.6%) than dry season of the year (72.7%), but there was no significant statistical variation between tick infestation and season of the year.

Table 5: Prevalence of tick infestation in cattle by season

Season	of animals Examined	of positive Animals	Prevalence %
Dry	147	107	72.7%
Wet	137	184	77.6%
Total	384	291	75.7%

4.1.6 Tick Burden and Species Identification in ChetaWoreda

During the study period, total 2024 adult ticks were collected from 291 cattle in three study areas (kebeles)

the mean tick burden of a single animal was 6.9 and the counts ranged from 2 to 18 for four different genera. The tick genera encountered take a count amblyomma (34.9%) Boophilus (26.6%), Hyalomma (19.2%) and Rhipicephalus (19%) in a diminishing hierarchy of overall abundance .

Table 6: Prevalence of Tick Species Identified in ChetaWoreda

Tick genera	Proportion
Amblyomma	708 (34.9%)
Boophilus	540 (26.6%)
Hyalomma	389 (19.21%)
Rhipicephalus	387 (19.12%)
Total	2024 (100%)

A total of 2024 tick samples belonging to four genera were identified in the study area. Amblyomma was the most abundant tick in the study area followed by

Boophilus, Hyalomma and Rhipicephalus in decreasing order (table 6)

Table 7: General of ticks and their distribution on body regions of cattle in Cheta woreda

Ticks general and No of infected animals (No of ticks per sampled body part)

Body region	Amblyomma	Boophilus	Hyalomma	Rhipicephalus	Total
Dewlap	31(234)	26(174)	19(163)	2(7)	78(578)
Udder	28(226)	17(114)	9(35)	3(9)	57(384)
Scrotum	22(157)	-	6(31)	2(8)	30(196)
Anal region and under tail	-	20(131)	7(14)	25(194)	52(339)
Sternum	18(91)	19(121)	15(146)	3(11)	55(369)
Ear	-	-	-	19(158)	19(158)
Total	110(10)	33(225)	64(363)	45(356)	291(2024)

With respect to risk factors influencing the prevalence of ticks only age and body condition were found to be significantly affecting between groups.

Table 8: Factors affecting tick infestation status of animals,

Risk factors	No of animal examined	No of positive animals
Keble		
Kushasha	128	94(73.4%)
Shaka	128	88(68.7%)
Duuba	128	109(85%)
Sex		
Male	135	101(74.8%)
Female	249	290(76.3%)
Age		
Young	127	76(59.8%)
Adult	194	153(78.8%)
Old	63	62(98.4%)
Body condition		
Good	46	32(69.5%)
Medium	306	227(74%)
Poor	32	32(100%)
Season		
Dry	147	107(72.7%)
Wet	237	184(77.6%)

Questionnaire was prepared to collect data from respondents concerning to tick infestation problem on the cattle's from their local area, 40 model farmers

were selected randomly in both sex to give their responses on major actors that lead cattle to tick infestation problem (table 9)

Table 9: Questionnaire data reorienting the question items and respondents response

No	Question contents	Alternative	Response			
			M	F	T. No	Total %
1	Have you known the ticks?	ayes	28	12	40	100
		b. no	-	-	-	0
2	Is there tick problem in our Keble?	a. yes	25	10	35	87.5
		b. no	3	2	5	12.5
3	Based on question No 2 if your answer is yes, what type of ticks you know in your locality?	a. hard tick	16	8	24	60
		b. soft tick	4	2	6	15
		c. both	8	2	10	25
4	On the above question No 2 which ticks are seriously damage (affects) cattle your local area?	a. hard tick	18	6	24	60
		b. soft tick	8	2	10	25
		c. both	4	2	6	15
5	Do you know the season of tick infection out break?	a. yes	28	12	40	100
		b. no	-	-	-	0
		a. at the end of rain season	2	1	3	7.5
		b. at the beginning or rain season	16	6	22	55
		c. at the mid of rainy season	4	3	7	17.5
6	The above question number 5 your answer is yes, in which season?	d. at the dry season	6	2	8	20
7	Do you know the major tick born disease?	a. yes	10	4	14	35
		b. no	18	8	26	65
8	Which species of livestock costly infected by ticks?	a. bovine	28	12	40	100
		b. ovine	-	-	-	0
		c. caprice	-	-	-	0
		b. exotic breed	19	4	23	57.5
		a. no	10	4	14	35

Discussion

In the present study, detailed investigations were carried out to identify and determine the type of genera and predication site of ticks infesting cattle in Chetaworeda of Kaffa zone. The prevalence and distribution of the most common tick genera infesting cattle is different from one area to another. Although there are different genera of ticks, only four genera of ticks, Amblyomma, Boophilus, Hyalomma and Rhipicephalus were identified.

In this study, Amblyomma were found to be the most abundant tick genera in Chetaworedadistinct accounting for 34.9%. The result of the study is in compatible with tick survey conducted in Western ShoaBako distinct by Husen (2009) that indicated the distribution of this tick genus as the first most

abundant genera in that area with prevalence of 34.9%. Amblyomma is a potential vector of disease caused by Cowdriarumintium (Sileshiet al., 2007).

Boophilus was the second most abundant tick genera (26.6%) in this study. This is in agreement with (Sileshiet al., 2007) who described that Boophilus is the commonest and most wide spread tick in Ethiopia, collected in all administrative regions except in Afar region. This is also in line with (Tamruet al., 2008) who reported the highest prevalence of Boophilus (80%). According to (Shiferawet al., 2005).Boophilus had highest frequency in the observed area during dry seasons(January, February and early March) in Kafa Zone. This result disagreed with the findings of (Alekawet al., 1998) at Metekel Ranch, Ethiopia showing prevalence of 5.7%.

This may be due to the geographical location and altitude factors which is 1500 to 1600mm above sea level of Metekelranch. The females were abundant from September to April and transmitted Babesiabigemina to cattle and sever infestation can lead to tick worry, anorexia and anemia (Seyoumet al., 2005).

Hyalomma is the third most abundant tick genera (19.2%) in this study. This tick can transmit a toxin causing sweating sickness in cattle and also damage the skins and hides (Mahmud et al., 2000). Rhipicephalus was the fourth and least abundant ticks genera (19%) in this study. This finding was lower than the works conducted by Seleshi and Solomon et al. (2007) at GhibeTullary in central Ethiopia who reported 21.2 of prevalence. This tick shows no apparent preference for any particular altitude, rainfall or season (Pegramet al., 1981). It is a possible vector of Babesia, Ricketssia and Theileria (Kettle et al., 1995).

With regard to distribution pattern of ticks, Amblyomma (Boophilus) was observed from all the body region of animals, even though the frequency of occurrence is more in the areas extending from Dewlap, Udder, Scrotum, and Anal and under tail and Sternum regions. Rhipicephalus were the most restricted tick genera identified. They were restricted to the Anal region and under tail and Ear areas, with very few of them observed on the scrotal, udder, Dewlap and Sternum area. Amblyomma were more fairly distributed than their Rhipicephalus counter parts, with the exceptions of their absence from the ear and anal region. Amblyomma were more concentrated in the areas of scrotum and udder. The dewlap, udder and scrotum were affected with the highest number of ticks followed by sternum. Scrotum and udder area was the most commonly affected body region in the current study of the total 291 animals.

The mean burden was significantly associated with age of animal in that older animal had significantly higher tick load than adult animals. This is probably associated with decreases in immunity as the animals get older.

In this study the prevalence of tick infestation between different body condition score groups was 100%, 74% and 69.5% in poor, medium and good animals respectively and the difference was significant. Poor body condition animal is most likely due to season of the years and statistical sample size. The higher prevalence in the emaciated animals is most likely due to low immunity associated with inadequate nutrition. In this study the prevalence of tick infestation between different kebeles was 85%, 73.4% and 68.7% in Kushasha, Shaka and Duuba respectively with significant difference this is due to the different Agro ecological conditions.

5. Conclusion and Recommendations

5.1 Conclusion

Ticks are obligate blood feeding ectoparasites of vertebrates and induce huge production loss in livestock industry. The main tick genera found in Ethiopia are Amblyomma, Boophilus, Haemaphysalis, Hyalomma and Rhipicephalus. Tick-borne diseases of cattle such as anaplasmosis, babesiosis, cowdriosis and theileriosis (T. mutans) are present in Ethiopia. Heavy infestations by different tick species suppress the immunity of cattle and also damage teats and reduce productivity of animals and there are direct effects associated with tick infestation that leads to tick worry, anorexia and anemia. These all are the impacts of tick infestation. The conventional method of controlling tick infestations in Ethiopia is application of acaricide, either by hand spraying, by hand dressing. The availability of vaccine is very small. Problems of acaricide resistance, chemical residues in food and the environment and the unsuitability of tick resistant cattle for all production systems make the current situation unsatisfactory and require the development of absolute control through effective vaccine.

5.2 Recommendations

In light of the above conclusion the following recommendations are forwarded:

More attention should be given to the selection of resistant cattle breeds and types, and good performance with regards to production of local breeds.

Tick control program (application of acaricides) should be continued with an increasing frequency of application in wet months.

Awareness should be created to the community to control the tick and tick infestation problems.

Appropriate pasture management in communal grazing area is important.

Detection of acaricide resistance tick species which are economically important since limited types of acaricides were used in the area.

Governmental and non-governmental institution should have pay attention to control and prevent the prevalence of tick infestation.

6. References

- Alekaw, S. 1998. Distribution of ticks and tick-borne diseases at Metekel Ranch. *Eth. Vet.* 1.,4(1): 30.
- Asfaw, W. 1997. Country report: Ethiopia in: proceedings of seminar on livestock Development policies in Eastern and South Africa, Mbabany, organization by CTA, OAU, the ministry of agriculture, cooperative, Swaziland.
- FAO. 1984. Ticks and TBDs control. A practical field manual volume m 1 tick control. FAO, Rome Pro 1-299.
- Hourrigan, JL. (1979) Spread and detection of Psoroptic scabies of cattle in the United States, *Journal of American Veterinary Association.* 175:1278-1280.
- Husen, U. 2009. Survey of cattle tick species and tick burden in and around Bako town DVM thesis, College of Agriculture and Veterinary Medicine, Jimma University, Jimma, Ethiopia.
- Kettle, D.S. 1995. Medical and Veterinary Entomology. 2nd Ed. UK: CAB International Pp. 420- 460.
- Mahmud. M. 2000. Raw hides and skin improvement In Ethiopia status and challenges in Preceding the opportunities and challenges of goat production in East Africa, a Conference held 10_12 ovember. 2000. Dehub University, Hawassa, Ethiopia.
- Martins and FurJang, 2001. A vermetin resistance of the cattle tick *Boophilus microplus* In Brazil. *The Veterinary Record* (2001),149(2), p.64
- Okello-Onen, J., S.Hassan and S. Essuman. 1999. Taxonomy of African ticks, an identification Manual.
- Pegram, R.G, Hoogstraal, Wassef, H.Y. 1981. Ticks (Acari: Ixodidae) of Ethiopia. Distribution, Ecology and relation or species infesting livestock. *Bull. Entomol. Res.* 71 :335-359.
- R.G, Preton, P.M. 2003. Ticks of domestic animals in Africa: A guide to identification
- Rahbori, S; Shvan, P., Iladdodzadeh, R. (2009); Domestic ruminants in Iran, Korean, *Journal of Parasitology.* www.ncbi.nlm.nih.gov/pmc/articles/pmc2526304.
- Regassa, a. 2001. The use of herbal preparations for tick control in western Ethiopia. *J S. Afr. Vet. Assoc.* 71,240-243.
- Seyoum, Z. 2005. Distribution and host parasite relationship of Ixodids ticks in Eastern Amhara, Kombolcha regional veterinary Laboratory, kombolcha, Ethiopia Pp. 34-47. Shearer
- Shiferaw. 2005. Cattle tick dynamics in different agro-ecological zones of Wolaita, Southern Ethiopia. Master degree thesis. Faculty of veterinary Medicine, Addis Ababa University, DebreZeit, Ethiopia Pp. 1-137.
- Sileshi, M., Pegram, R.G. Solomon, G., Abebe, M., Vilma, J., Sileshi, Z. 2007. A synthesis of review of Ixodids (Acari: Ixodidae) and Argas (Acari: Argasidae) ticks in Ethiopia and their possible Role in diseases transmission. *Ethiopia Vet.* 1. 2: 1-22. species. The University of Edinburgh, UK Pp. 67-80.
- Tamru, T. 2008. Survey of Bovine tick species in and around Asela Town, DVM Thesis, School of Veterinary Medicine, Jimma University. Jimma, Ethiopia.
- Thrustfield. M. 1995. *Veterinary Epidemiology.* 2nd Ed. UK Black well science Ltd; Pp. 182-198.

Walker, A.A, Bouatour, A, Camicas, J.L, Estadapena, A.A, Harok, I.G, Hatif, A.A, Pegram, Yacob, H.T., Atakly, H., Kumsa, B. 2008. Major ectoparasites of cattle in and around Mekelle, northern Ethiopia, Entomological Research, 38, 126-130

Access this Article in Online	
	Website: www.ijarbs.com
	Subject: Entomology
Quick Response Code	
DOI: 10.22192/ijarbs.2021.08.06.024	

How to cite this article:

Dechu Jobir, Mulatu Gure. (2021). Study prevalence of hard tick infestation at South Western Kafa Zone Cheta Woreda. Int. J. Adv. Res. Biol. Sci. 8(6): 206-223.

DOI: <http://dx.doi.org/10.22192/ijarbs.2021.08.06.024>