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LIVESTOCK TICKS IN THE UAE: PREVALENCE, DISTRIBUTION, POPULATION DYNAMICS, AND ASSOCIATED MICROORGANISMS

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This dissertation is submitted in partial fulfilment of the requirements for the degree of Doctor of Philosophy

Under the Supervision of Dr. Mohammad Ali Al-Deeb

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Declaration of Original Work

I, Nighat Perveen, the undersigned, a graduate student at the United Arab Emirates University (UAEU), and the author of this dissertation entitled "*Livestock Ticks in the UAE: Prevalence, Distribution, Population Dynamics, and Associated Microorganisms*", hereby, solemnly declare that this dissertation is my own original research work that has been done and prepared by me under the supervision of Dr. Mohammad Ali Al-Deeb, in the College of Science at UAEU. This work has not previously formed the basis for the award of any academic degree, diploma or a similar title at this or any other university. Any materials borrowed from other sources (whether published or unpublished) and relied upon or included in my dissertation have been properly cited and acknowledged in accordance with appropriate academic conventions. I further declare that there is no potential conflict of interest with respect to the research, data collection, authorship, presentation and/or publication of this dissertation.

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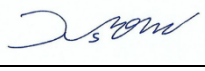
   Signature  Date 09-Dec-2021

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Abstract

Ticks are important vectors of an array of viral, bacterial, and protozoan pathogens resulting in a wide range of animal and human diseases in the Middle East and North Africa (MENA). In this study, ticks were collected from camels, cows, sheep, and goats in Abu Dhabi, Dubai, and Sharjah in the United Arab Emirates (UAE). The objectives of the study were to (1) identify tick species of livestock through taxonomic keys and using molecular markers, and determine their prevalence and distribution in the UAE, (2) assess *Hyalomma dromedarii* seasonal population fluctuation over a year under common camel breeding and management practices, (3) determine bacterial communities' composition and diversity in camel tick, *H. dromedarii* using Next-Generation Sequencing, and (4) detect tick-borne microbes and their prevalence in *Hyalomma* ticks collected from livestock. In the UAE, information on the ticks on camels and other livestock is limited, which is essential for designing and instigating effective tick control strategies in the country. In this study, four aspects of ticks and tick-borne diseases of livestock have been investigated in the UAE. First, four tick species, *H. dromedarii*, *Hyalomma anatolicum*, *Amblyomma lepidum*, and *Rhipicephalus sanguineus* were identified from livestock including camel, cow, sheep, and goat. These tick species were morphologically identified by using taxonomic keys and confirmed through molecular characterization. This study provided the first DNA molecular record of *H. anatolicum*, *A. lepidum*, and *R. sanguineus* from the UAE. Second, population fluctuation of *H. dromedarii* was evaluated over one year. Ticks were collected monthly from camels in Al-Ain, UAE, over 12 months (March 2019 to February 2020). Further, *H. dromedarii* sex ratio was calculated and parasitological indicators were measured. Results showed that the infestation prevalence was very high (94.33%) during the whole study period. The maximum infestation intensity occurred in June, while the minimum occurred in November. Overall, *H. dromedarii* ticks were found on camels during the entire year despite monthly applications of an acaricide. Third, the composition and diversity of bacterial communities associated with *H. dromedarii* collected from camels in Al-Ain, UAE, were determined. A total of 100 partially engorged female ticks were taken from tick samples and subjected to DNA extraction and Next-Generation Sequencing. The 16S rRNA gene was amplified from genomic DNA and sequenced using the Illumina

MiSeq platform to reveal the bacterial communities. Principal Coordinates Analysis (PCoA) was conducted to determine patterns of diversity in bacterial communities. Twenty-five bacterial families with high relative abundance were identified. Francisellaceae and Enterobacteriaceae coexisted in *H. dromedarii*. The dominant bacterial genus was *Francisella*. Fourth, the presence and prevalence of tick-borne *Francisella* sp., *Rickettsia* sp., and piroplasmids were determined in *Hyalomma* ticks infesting livestock. A total of 562 tick samples were collected from camels, cows, sheep, and goats from 24 locations. DNA was extracted from ticks and Polymerase Chain Reactions (PCRs) were performed. *Hyalomma dromedarii* ticks collected from camels were infected with *Francisella*-Like Endosymbionts (FLE) (5.81%) and *Candidatus* Rickettsia andeanae (1.36%). *Hyalomma anatolicum* ticks collected from cows were found positive with *Theileria annulata* (4.55%) whereas *H. anatolicum* ticks collected from goats were positive with *Theileria ovis* (10%). *Theileria ovis* was detected for the first time in the UAE. Therefore, further investigations on tick species and tick-borne microbes are required to understand ticks' biology, ecology, and microbes' interaction and their role in tick-borne diseases epidemiology in the UAE.

Keywords: Ticks, tick-borne microorganisms, population dynamics, microbial communities, UAE.

Title and Abstract (in Arabic)

قراد الحيوانات في دولة الإمارات العربية المتحدة: الانتشار، والتوزيع، وديناميكيات المجتمع، والكائنات الدقيقة المرافقة

الملخص

تعتبر القراد ناقلات مهمة لمجموعة من مسببات الأمراض الفيروسية والبكتيرية والأولية التي تؤدي إلى مجموعة واسعة من الأمراض الحيوانية والبشرية في منطقة الشرق الأوسط وشمال إفريقيا. في هذه الدراسة، جمعت القراد من الإبل والأبقار والأغنام والماعز في أبو ظبي ودبي والشارقة في الإمارات العربية المتحدة. أهداف الدراسة هي (1) تحديد أنواع القراد من الماشية من خلال مفاتيح التصنيف واستخدام الواسمات الجزيئية، وتحديد مدى انتشارها وتوزيعها في الإمارات العربية المتحدة، (2) تقييم تذبذب عشائر قراد الابل من خلال تقييم التغيرات في مرحلة الحياة ونسبة الجنس خلال 12 شهرًا، وقياس المؤشرات الطفيلية لإصابة H. dromedarii، (3) تحديد تكوين المجتمعات البكتيرية وتنوعها في قراد الإبل، H. dromedarii باستخدام تسلسل الجيل التالي، و (4) اكتشاف الميكروبات التي تنقلها القراد وانتشارها في قراد Hyalomma الذي تم جمعه من الماشية. عالمياً تم إنشاء سجل تصنيفي لـ 55 نوعًا من القراد التي تصيب الماشية تمثل الأجناس الثمانية التالية: Ornithodoros و Otobius و Amblyomma و Dermacentor و Haemaphysalis و Hyalomma و Ixodes و Rhipicephalus. في الإمارات العربية المتحدة، المعلومات عن القراد على الإبل وغيرها من المواشي محدودة، وهو أمر ضروري لتصميم وتحفيز استراتيجيات مكافحة فعالة في الدولة. حتى الآن، تم إجراء القليل من الدراسات حول القراد والكائنات الحية الدقيقة التي تنقلها القراد وانتشارها في الإمارات العربية المتحدة. في هذه الدراسة، تم التحقيق في أربعة جوانب من القراد والأمراض التي تنقلها القراد للماشية في الإمارات العربية المتحدة. أولاً، حددت أربعة أنواع من القراد، Hyalomma dromedarii و Hyalomma anatolicum و Amblyomma lepidum و Rhipicephalus sanguineus على الماشية بما في ذلك الجمال والبقر والأغنام والماعز من الإمارات العربية المتحدة. تم تحديد أنواع القراد هذه شكليًا باستخدام مفاتيح التصنيف وتم إجراء التوصيف الجزيئي لاحقًا باستخدام أدوات جزيئية مختلفة. قدمت هذه الدراسة أول سجل جزيئي DNA لـ H. anatolicum و A. lepidum و R. sanguineus من الإمارات العربية المتحدة. ثانيًا، قمت بتقييم التقلبات السكانية لـ H. dromedarii على مدى عام واحد في ظل ممارسات

تربية وإدارة الإبل الشائعة في منطقة الدراسة. علاوة على ذلك، قمت بحساب نسبة جنس H. dromedarii خلال 12 شهرًا وقمت بقياس المؤشرات الطفيلية لإصابتها. لقد أجريت عمليات تعداد بصرية شهرية في الموقع وقمت بجمعها من الإبل في مدينة العين، الإمارات العربية المتحدة، على مدار 12 شهرًا (مارس 2019 إلى فبراير 2020). أظهرت نتائجي أن انتشار الإصابة كان مرتفعاً للغاية خلال فترة الدراسة بأكملها بمتوسط 94.33%. حدثت أقصى شدة للغزو في يونيو، بينما حدث الحد الأدنى في نوفمبر. بشكل عام، تم العثور على قراد H. dromedarii على الإبل خلال العام بأكمله على الرغم من المعاملات الشهرية بمبيد القراد. ثالثًا، حددت تكوين وتنوع المجتمعات البكتيرية المرتبطة بـ H. dromedarii التي تم جمعها من الإبل في مدينة العين، الإمارات العربية المتحدة، خلال الدراسة الجينومية. تم أخذ ما مجموعه 100 أنثى قراد محتقنة جزئيًا من عينات القراد التي تم جمعها من الإبل وخضعت لاستخراج الحمض النووي وتسلسل الجيل التالي. تم تضخيم جين الرنا الريباسي S16 من الحمض النووي الجيني وتسلسله باستخدام منصة Illumina MiSeq للكشف عن المجتمعات البكتيرية. تم إجراء تحليل الإحداثيات الرئيسية (PCoA) لتحديد أنماط التنوع في المجتمعات البكتيرية. تم تحديد خمسة وعشرين عائلة بكتيرية ذات وفرة نسبية عالية. تتعايش الفرنسيسيلاسية والمعوية في بكتيريا H. dromedarii. رابعًا، حددت وجود وانتشار الفرنسيسيلا sp. و Rickettsia sp. و piroplasmids التي تنقلها القراد في القراد الذي يصيب الماشية، وقمت بتقدير معدلات الإصابة بها باستخدام مناهج مختلفة تعتمد على تفاعل البوليميراز المتسلسل. تم جمع إجمالي 562 عينة من القراد من الإبل والأبقار والأغنام والماعز في إمارات أبو ظبي ودبي والشارقة من 24 موقعًا. تم استخراج الحمض النووي من القراد وأجري PCRs. كان قراد Hyalomma dromedarii الذي تم جمعه من الإبل مصابًا بالمعايشات الداخلية الشبيهة بفرنسيسيلا (5.81%) و Candidatus Rickettsia andeanae (1.36%). تم العثور على قراد Hyalomma anatolicum الذي تم جمعه من الأبقار موجبًا مع Theileria annulata (4.55%) بينما كان H. anatolicum الذي تم جمعه من الماعز موجبًا مع Theileria ovis (10%). تم اكتشاف Theileria ovis لأول مرة من الإمارات العربية المتحدة. لذلك، يلزم إجراء مزيد من التحقيقات حول أنواع القراد والميكروبات التي تنقلها القراد لفهم بيولوجيا القراد، والبيئة ، وتفاعل الميكروبات ودورها في وبائيات الأمراض التي تنقلها القراد في الإمارات العربية المتحدة.

مفاهيم البحث الرئيسية: القراد، الكائنات الدقيقة المنقولة بالقراد، ديناميكيات المجتمع، المجتمعات البكتيرية، الإمارات العربية المتحدة.

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Dedication

To my beloved family

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List of Abbreviations

ALKV	Alkhurma Hemorrhagic Fever Virus
BIC	Bayesian Information Criterion
BLAST	Basic Local Alignment Search Tool
CDC	Centers for Disease Control and Prevention
cox1	Cytochrome c Oxidase Subunit I
CCHF	Crimean-Congo Hemorrhagic Fever
CCHFV	Crimean-Congo Hemorrhagic Fever Virus
IHR	International Health Regulations
LSM	Livestock Market
MENA	Middle East and North Africa
MSF	Mediterranean Spotted Fever
NCBI	National Center for Biotechnology Information
NGS	Next-Generation Sequencing
OTUs	Operational Taxonomic Units
PCoA	Principal Coordinates Analysis
RH	Relative Humidity
RMSF	Rocky Mountain Spotted Fever
SFG	Spotted Fever Group
TBD	Tick-Borne Disease
TBE	Tick-Borne Encephalitis
TBEV	Tick-Borne Encephalitis Virus
UAE	United Arab Emirates
VBD	Vector-Borne Diseases

Chapter 1: Introduction

1.1 Overview

Ticks are hematophagous ectoparasites of a wide range of vertebrate hosts, including livestock, wildlife, and humans, and play the main role in transmitting zoonotic diseases that are major causes of infection's related morbidity and mortality across the world (Pfäffle, Littwin, Muders, & Petney, 2013). Emerging or re-emerging infectious diseases are important global problems of great concern to domestic animals, wildlife, and human health, with many pathogens being able to infect multiple species (Cunningham, Daszak, & Wood, 2017). As antimicrobial resistance among bacterial pathogens is increasing, there has been an increase in the incidences of zoonotic diseases, occasionally causing widespread outbreaks with considerable humans and animal infections and deaths (Cunningham et al., 2017). Moreover, global warming and unstable climate are contributing factors in driving the emergence, resurgence, and redistribution of infectious diseases across the world (Epstein, 2001). Further, globalization and climatic anomalies have allowed parasites to invade into new geographical areas, and give an upsurge to epidemics and epizootics worldwide (Daszak, 2005). Therefore, infectious diseases have been recognized as a growing threat to public health and animal husbandry.

The Arabian Peninsula has significance as a source of energy, with huge oil and gas reserves that supply the energy demands globally. Consequently, this region went through massive changes and development. The side effects of this development include the expansion of farming to support the demands of the growing population particularly in the Arabian Peninsula (Faour-klingbeil & Todd, 2018). Traditional and modern farming techniques have been used for livestock farming such as the rearing

of camels, cows, buffaloes, sheep, and goats (Hasson & Al-Zubaidi, 2014). These farmed animals are a source of considerable amounts of milk, meat, wool, and hides (FAO, 2020). Ectoparasites such as ticks constitute a major threat to livestock industries in the Middle East and North Africa (MENA) region. Ticks have great importance from economic, veterinary, and human health viewpoints due to their ability to transmit a variety of tick-borne infectious diseases (Montiel-Parra, Fuentes-Moreno, & Vargas-Sandoval, 2007). Ticks and Tick-Borne Diseases (TBDs) are affecting humans and livestock with a loss of US $ 13.9 – 18.7 billion annually globally (Hussain et al., 2021). The ixodid ticks belonging to the genus *Hyalomma* may cause massive damages to the products arising from camel, cattle, sheep, and goats across the MENA region. The knowledge about the roles of diverse assemblages of ticks as reservoirs and vectors of tick-borne pathogens and their impacts on livestock and humans is crucial in Arab countries. Further, understanding the abundance of tick species, as well as their geographic distribution, is essential in evaluating the threat of TBDs outbreak and their management strategies in the MENA region. The MENA region is a hyperarid region connecting Africa to Eurasia (Abahussain, Abdu, Al-Zubari, El-Deen, & Abdul-Raheem, 2002; Khan, 2013) and with increasing levels of economic development in this region, camel farming has become enriched in recent years, particularly in the Middle East. The United Arab Emirates (UAE) in particular has enormously increased development on account of its position as a trade hub (Gardner & Howarth, 2009), however, almost 98% of the UAE (a region ranging from Oman to Saudi Arabia) is covered by desert and gives home to a large camel population (459,000 camel heads) (FCSA, 2017). In this desert ecosystem, camels are unique animals and possess distinct browsing behavior which makes this species superior to small ruminants in conserving vegetation cover (Gharbi, Moussi, Jedidi, Mhadhbi, &

Sassi, 2013). The development of camel rearing in close contact with other livestock species in the UAE may contribute to the spread of tick-borne pathogens in other livestock species and this may also be a snag in camel tick population mitigation strategies. Previously, in Mauritania, due to mixed herds of camels and cattle, the camel tick *Hyalomma dromedarii* was reported as the main vector of *Theileria annulata* in cattle (Jacquiet, Colas, Cheikh, Thiam, & Ly, 1994). Camel tick may play a significant role in the epidemiology of viruses such as Dera Ghazi Khan virus, Dhori virus, Kadam virus and Crimean-Congo hemorrhagic fever virus (Hoogstraal, 1979; Hoogstraal, Wassef, & Buttiker, 1981; Rodriguez et al., 1997; Wood, Moussa, Hoogstraal, & Biittiker, 1982). Moreover, camel tick is also the vector of bacterial pathogens including *Coxiella burnetii* that causes Q fever (Bazlikova, Kazar, & Schramek, 1984) and *Rickettsia rickettsii* that causes spotted fever rickettsia (Lange, El Dessouky, Manor, Merdan, & Azad, 1992) and protozoan pathogens, for example, *Theileria camelensis* that causes theileriosis of camels and *T. annulata* that causes theileriosis in cattle (Hoogstraal et al., 1981). Due to a multitude of factors, such as constantly changing in tick population densities, tick-borne microbes composition, tick-transmitted infections and hosts diversity, the patterns of tick-host-pathogen interrelationships are changing (Fuente et al., 2016; Hoogstraal & Valdez, 1980; Wikel, 2018). Knowledge and understanding of the facts about the epidemiology of tick-borne diseases especially on the transmission dynamics of vector ticks are critical for the devising of effective control strategies (Wikel, 2018).

1.2 Statement of the Problem

Ticks are ectoparasites on camels and other livestock in the UAE. Ticks are the carrier of many pathogens such as bacteria, viruses, and protozoans. They may transmit zoonotic diseases to animals and humans who are working with the livestock industry, agriculture, slaughterhouses, and veterinary practice. Zoonotic diseases are the major global threats to human health and sustainable development, and the health of humans, animals, and the environment are all interconnected. In addition, warm and humid climates are most favorable for tick species in the MENA region. The UAE is known for its attachment to camels which have social and economic value in the country. *Hyalomma dromedarii* is the most abundant tick species on camels followed by *Hyalomma anatolicum* on cows and sheep in the UAE. Recently, there were reports of viral pathogens in neighboring countries, for instance, Oman and Saudi Arabia, and bacterial pathogens in the UAE from different tick species which created a huge threat to the UAE animals and people interacting with them. Several acaricides are used to control the ticks; however, many species are not going to be managed by these acaricides, and some show resistance against some of them.

1.3 Aims and Objectives

In the UAE, research studies on camel ticks and other livestock ticks are very few. Information on tick species identification, their seasonal population dynamics, the composition of microbes inside the ticks and their interaction, and molecular characterization of tick-borne microbes are obligatory for designing and initiating effective tick management strategies in the country to circumvent the future tick-borne diseases in humans and animals. The present work was aimed to generate the data on tick species through morphological and molecular characterization, seasonal

abundance, and microbes composition inside ticks by using genomic approaches. Therefore, following five hypotheses were formulated, 1) various tick species of livestock could be present in the UAE, 2) *Hyalomma dromedarii* ticks could be present all the year on camels, 3) *Hyalomma dromedarii* could harbor diverse bacterial microbes, 4) *Hyalomma dromedarii* could be infected with *Rickettsia, Francisella* and piroplasmids, 5) *Hyalomma anatolicum* could be infected with *Rickettsia, Francisella* and piroplasmids. To test all above hypotheses, the objectives of the current study were to:

1) Identify tick species of livestock through taxonomic keys and using molecular markers, and determine their prevalence and distribution in the UAE.
2) Assess *H. dromedarii* seasonal population fluctuation over a year under common camel breeding and management practices by evaluating life stage changes and sex ratio during 12 months, and measure parasitological indicators of *H. dromedarii* infestation.
3) Determine bacterial communities' composition and diversity in camel tick, *H. dromedarii* using Next-Generation Sequencing.
4) Detect tick-borne microbes and their prevalence in *Hyalomma* ticks collected from livestock.

1.4 Relevant Literature

1.4.1 Ticks as Vectors

Ticks are considered an important group of arthropod vectors due to their potential of transmitting a wide variety of human and animal pathogens (Jongejan & Uilenberg, 2004). The knowledge of tick distribution pattern and phenology is significant to identify those areas and periods involving a high risk of exposure to

specific tick-borne pathogens to promote prevention measures (Parola & Raoult, 2001). Microclimate features and relative abundance of some key hosts impact the seasonal patterns of tick activity, their distribution, and the persistence of foci of pathogens (Alonso-Carne, Garcı´a-Martın, & Estrada-Pena, 2014). Temperature and relative humidity affect tick host-seeking behavior and survival rates (Parola & Raoult, 2001; Ruiz-fons, Fernández-de-Mera, Acevedo, Gortázar, & de la Fuente, 2012) and regulate the duration of questing periods as well as the territory with suitable environmental conditions.

1.4.2 Geography of the Arab Countries

Twenty-two countries represent the Arab world in the MENA region (Tadmouri, 2004) (Figure 1). The area occupied by these countries is 14,291,469 km^2, which constitutes approximately 10.2% of the world's landmass (Khan, 2013) and is ranging from the Atlantic Ocean to the Zagros Mountains in southwest Asia (Elasha, 2010). Further, the Arab world consist of eastern and western parts and the Arab east is the Asian part of the Arab world and covers an area of about 27.55%, while the Arab west is the African part which covers the 72.45% area (Abahussain et al., 2002; Khan, 2013). The Arab region consists of mainly arid, semi-arid, and dry sub-humid areas which constitute 90% of the total area that is characterized by limited water resources, harsh environment, and arable lands. The history of this region tells us, these lands served as the major source of livestock breeding and production of animal milk and meat (Abahussain et al., 2002; Elasha, 2010). The Arab nations total population is approximately 427 million (WBG, 2020).

1.4.3 Ticks and Livestock Industry in the MENA Region

Fast development in some parts of the MENA region over the last 30 years resulted in a large urban population composed of multiple ethnicities (Gardner & Howarth, 2009) and this has been concomitant with the development of the farming industry. Certain farms in the region are located on the outskirts of cities, with a rise in camel farming across the region to support the increasing demand of camel milk and meat. Currently, there are over 1.6 million camel heads in the Arabian Peninsula (Abdallah & Faye, 2012) and over 459,000 camel heads in the UAE (FCSA., 2017). Ticks belong to genus *Hyalomma* pose major threats to camels and other livestock across Africa, Eastern Europe, the Middle East, and Western Asia (Alanazi, Abdullah, Wall, & Alharbi, 2018; Alanazi, Al-Mohammed, Alyousif, Puschendorf, & Abdel-Shafy, 2018; Wernery & Kaaden, 2002). *Hyalomma* species are medium to large-sized ticks that parasitize a wide range of animals including domestic and wild mammals, and birds (Alanazi, Al-Mohammed, et al., 2018; Jongejan & Uilenberg, 2004). *Hyalomma dromedarii* is the most common tick species in the MENA region that infests camels can transmit tick-borne diseases in camels and to some extent in humans (Al-Deeb & Muzaffar, 2020; Al-khalifa, Diab, & Khalil, 2007; Alanazi, Al-Mohammed, et al., 2018; Elghali & Hassan, 2009; Karrar, Kaiser, & Hoogstraal, 1963; Madder, Horak, & Stoltsz, 2013). However, there have been relatively few studies on the dynamics, ecology, and biology of *H. dromedarii* ticks (Alahmed & Kheir, 2003; ELGhali & Hassan, 2010). Tick infestation is the major problem in the livestock industry in the Arabian Peninsula even though a wide variety of acaricides are used to control ticks; their efficacy is not well characterized.

In the MENA region, tick populations may have a unique set of challenges due

to the natural inhospitable environment, with pockets of habitat that periodically become productive (Almazroui, Islam, Athar, Jones, & Rahman, 2012). However, the widespread livestock farming in the region provides the opportunity for ticks to sustain with high densities in artificial farming conditions (e.g. with shelter and water provided for animals). Local farms abound in the region with low to medium density of livestock, although efforts were made to modernize the livestock industry. In addition, these farms receive imported livestock including cattle, sheep, and goats more often from Australia, New Zealand, China, and Argentina (Miranda-de la Lama, Villarroel, & María, 2014). Therefore, local ticks' populations find opportunities to feed on naive imported hosts, with significant chances of boosting their populations to levels much higher than what could be normally supported in the desert ecosystem. Consequently, ticks' populations have been enhanced in the region and farms characterize the areas of high population density. The use of various acaricides is common and there is no vector (tick) control program (Meneghi, Stachurski, & Adakal, 2016). Consequently, ticks have continuous opportunity to develop resistance to acaricides (George, Pound, & Davey, 2004; Meneghi et al., 2016). In addition, the chance of importing pathogens that may remain dormant in the host also increases the due frequent influx of foreign host animals (Rottier & Ince, 2005).

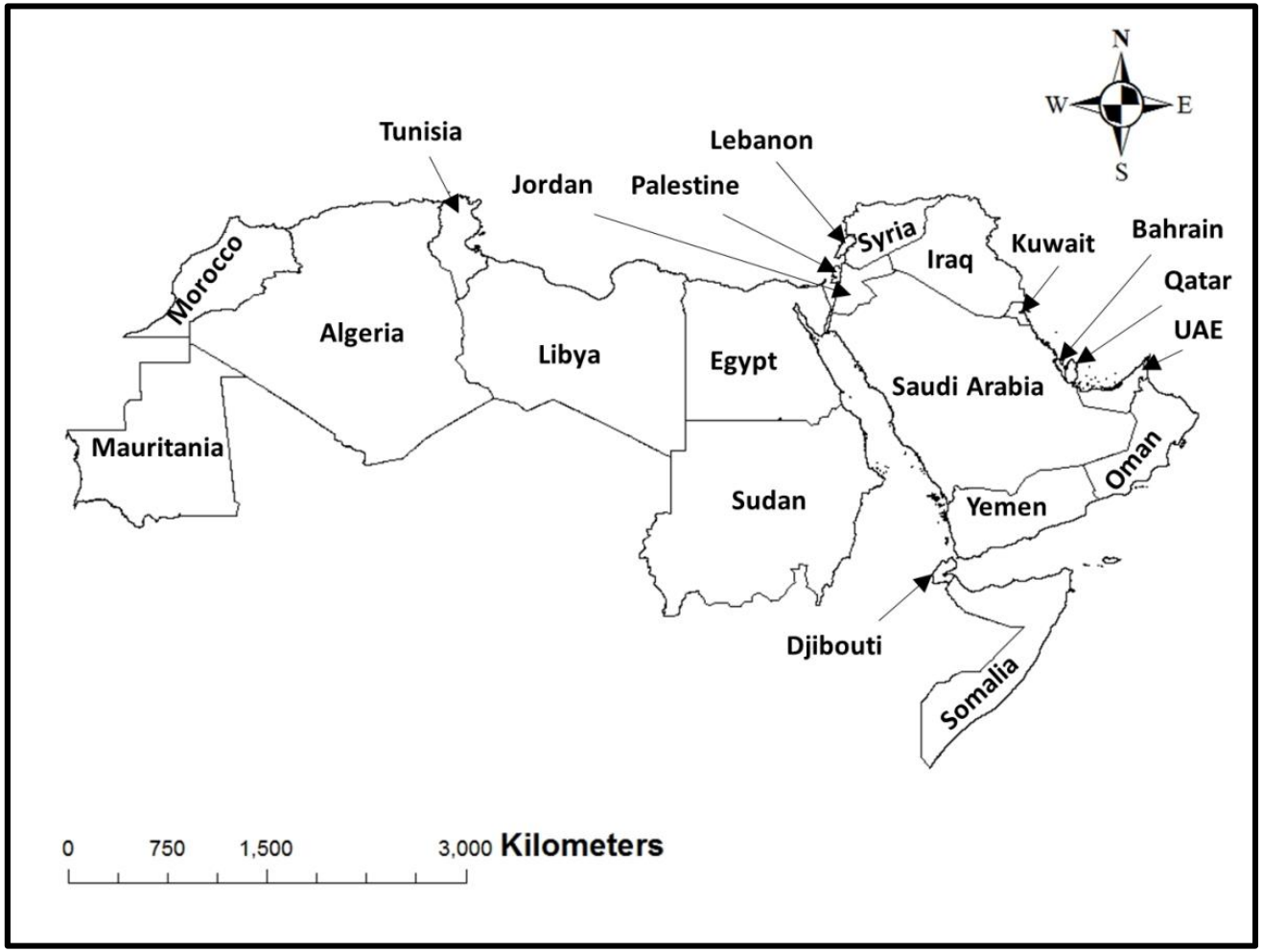

Figure 1: Regional map showing Arab countries.

1.4.4 Tick's Diversity and Distribution in the MENA Region

Ticks are economically important ectoparasites of livestock because they infest 80% of the world's cattle. Consequently, they have a negative effect on the health of their vertebrate hosts because blood-feeding causes allergic skin reactions, tick toxicosis, and severe paralysis (Champour, Chinikar, & Mohammadi, 2016; Pasalary, Arbabi, Pashei, & Abdigoudarzi, 2017). Further, ticks and tick-borne diseases among livestock in tropical and subtropical regions cause economic losses, several billion dollars yearly (Jongejan & Uilenberg, 1994, 2004). Hot dry summers and mild winters with high relative humidity are characteristic climatic conditions of the Arabian Peninsula (Almazroui et al., 2012). Hard ticks circulate in the areas where sufficient animal hosts are found. Further, ixodid ticks with two-host or three-host life cycles occur in the areas that overlap with all their hosts (assuming that different instars feed on different host species). Ticks could potentially use a combination of host species to complete their life cycles, in the case of livestock. However, sometimes, immature stages such as larvae and nymphs could feed on wild rodents or hedgehogs. In the traditional farming systems, these species are important where livestock holding ranges are accessed by small mammals (Al-Khalifa, Al-Asgah, & Diab, 1984; Diab, Hoogstraal, Wassef, Al Khalifa, & Al Asgah, 1985).

From livestock, eight tick genera including 55 species have been documented in the Arabian Peninsula (Table 1). *Ornithodoros* and *Otobius* represented the soft ticks (Argasidae). *Ornithodoros savignyi* is recorded as the most widespread soft tick species in the MENA region. Further, six genera, *Amblyomma, Dermacentor, Haemaphysalis, Hyalomma, Ixodes*, and *Rhipicephalus*, represented the hard ticks (Ixodidae). *Hyalomma impeltatum, H. dromedarii, R. annulatus*, and *R. sanguineus*

are widely distributed across the MENA region and *H. dromedarii* is reported as the most common tick species with high prevalence due to large-scale camel farming in this region. In most of the countries of the Arab world, taxonomic studies on tick species are limited. In addition, the majority of systematics studies on ticks of the Arab region were dedicated to the hard ticks. Two hard tick genera, *Hyalomma* and *Rhipicephalus* are reported as the most common genera from livestock in all Arab countries. Genus, *Hyalomma* serves as the vector and reservoir of CCHF virus in this region (Bakheit, Latif, Vatansever, Seitzer, & Ahmed, 2012). Egypt, Sudan, and Jordan, maximum numbers of tick species (N = 22) are recorded from livestock, followed by Yemen, Iraq, and Saudi Arabia (N = 19, 17, 17), respectively (Figure 2). Some tick species are common and widespread in the MENA region, in which *H. impeltatum* and *R. sanguineus* were reported in 15 countries. However, *H. dromedarii* was recorded in 13 countries (Figure 3). Knowledge of tick distribution is crucial to cope with future emerging and re-emerging tick-borne diseases and disease movement between countries that share borders with each other.

Table 1: Tick species distribution in Arab countries

Families	Genera	Species	Arab Countries	Hosts	References
Argasidae	*Ornithodoros*	*O. erraticus*	Iraq, Jordan, Saudi Arabia	Animals	(FGS, 1999; Saliba, Amr, Wassef, Hoogstraal, & Main, 1990)
		O. foleyi	Oman, Libya	Farm animals	(FGS, 1999; Gabaj, Awan, & Beesley, 1992)
		O. lahorensis	Jordan		(Saliba et al., 1990)
		O. salahi	Egypt, Jordan		(Hoogstraal, 1953; Saliba et al., 1990)
		O. savignyi	Algeria, Egypt, Lebanon, Libya, Mauritania, Oman, Saudi Arabia, Yemen	Camels	(Chalon, 1923; FGS, 1999; Hoogstraal & Kaiser, 1960; Pegram, Hoogstraal, & Wassef, 1982; Sylla, Molez, Cornet, & Camicas, 2008; Wassef, Buttiker, & Gallagher, 1997)
		O. tholozani	Egypt, Libya, Jordan	Humans	(Davis & Hoogstraal, 1954; Hoogstraal & Kaiser, 1960; Saliba et al., 1990)
	Otobius	*O. megnini*	Iraq	Buffaloes	(Al-Mayah & Hatem, 2018)
Ixodidae	*Amblyomma*	*A. gemma*	Saudi Arabia, UAE, Yemen	Cows, camels	(FGS, 1999; Hoogstraal & Kaiser, 1959; Khan et al., 1997)
		A. lepidum	Egypt, Iraq, Saudi Arabia, Sudan, UAE	Livestock, camels	(FGS, 1999; El Kammah, Oyoun, El Kady, & Shafy, 2001; Khan et al., 1997; Salih, Julla, Hassan, Hussein, & Jongejan, 2008)
		A. marmoreum	Egypt	Camels	(El Kammah et al., 2001)
		A. variegatum	Egypt, Oman, Saudi Arabia, Sudan, Yemen	Camels	(El Kammah et al., 2001; FGS, 1999; Hassan, Gabr, Abdel-Shafy, Hammad, & Mokhtar, 2017a; Pegram et al., 1982)
		A. latum	Saudi Arabia, Yemen	Domestic animals	(FGS, 1999; Hoogstraal & Kaiser, 1959)
	Dermacentor	*D. marginatus*	Algeria, Lebanon	Cattle	(Dabaja et al., 2017; Mokhtaria et al., 2018)
	Haemaphysalis	*H. erinacei*	Algeria, Iraq, Jordan	Animals	(Saliba et al., 1990; Shamsuddin & Mohammad, 1988)
		H. indica	Oman		(FGS, 1999)
		H. parva	Iraq, Libya, Jordan	Domestic animals	(El-Rabie, Amr, & Hyland, 1990; Hoogstraal & Kaiser, 1960; Omer, Kadir, Seitzer, & Ahmed, 2007; Saliba et al., 1990)

Table 1: Tick species distribution in Arab countries (Continued)

Families	Genera	Species	Arab Countries	Hosts	References
		H. punctata	Algeria, Lebanon, Tunisia	Domestic animals	(Bouattour, Darghouth, & Daoud, 1999; Dabaja et al., 2017; Yousfi-Monod & Aeschlimann, 1986)
		H. sulcata	Iraq, Jordan, Saudi Arabia, Tunisia, Yemen	Sheep	(Pegram et al., 1982; Saliba et al., 1990; Shamsuddin & Mohammad, 1988)
	Hyalomma	*H. anatolicum*	Egypt, Iraq, Jordan, Kuwait, Lebanon, Oman, Saudi Arabia, Sudan, Syria, UAE, Yemen	Livestock	(Asmaa, ElBably, & Shokier, 2014; Hasson, 2012; Hoogstraal & Kaiser, 1959; Shamsuddin & Mohammad, 1988)
		H. arabica	Yemen	Goats, sheep	(Pegram et al., 1982)
		H. asiaticum	Iraq	Cattle, sheep	(Hasson, 2012)
		H. dromedarii	Algeria, Egypt, Iraq, Jordan, Kuwait, Mauritania, Oman, Qatar, Saudi Arabia, Sudan, Tunisia, UAE, Yemen	Camels	(Abdel-Shafy, Allam, Mediannikov, Parola, & Raoult, 2012; Djerbouh et al., 2012; El Kammah et al., 2001; Hagras & Khalil, 1988; Hassan et al., 2017a; Pegram et al., 1982)
		H. excavatum	Algeria, Egypt, Iraq, Jordan, Lebanon, Libya, Saudi Arabia, Sudan, Syria, Tunisia, UAE, Yemen	Cattle, camels	(Abdel-Shafy et al., 2012; El Kammah et al., 2001; Hasson, 2012; Leulmi et al., 2016; Pegram et al., 1982)
		H. franchinii	Libya, Tunisia	Camels, sheep	(Bouattour et al., 1999; Gabaj et al., 1992)
		H. hussaini	UAE	Livestock	(Khan et al., 1997)
		H. impeltatum	Algeria, Egypt, Iraq, Jordan, Kuwait, Libya, Mauritania, Oman, Qatar, Saudi Arabia, Sudan, Syria, Tunisia, UAE, Yemen	Camels, cattle, goats	(Abdel-Shafy et al., 2012; Camicast, Wilson, Cornett, & Digoutte, 1990; Djerbouh et al., 2012; El Kammah et al., 2001; Hasson, 2012; Khalil & Hagras, 1988; Khan et al., 1997)
		H. impressum	Algeria, Mauritania, Sudan	Camels	(Camicast et al., 1990; Djerbouh et al., 2012)
		H. lusitanicum	Algeria	Cattle	(Mokhtaria et al., 2018; Yousfi-Monod & Aeschlimann, 1986)

Table 1: Tick species distribution in Arab countries (Continued)

Families	Genera	Species	Arab Countries	Hosts	References
		H. marginatum	Algeria, Egypt, Iraq, Jordan, Kuwait, Libya, Syria, Tunisia, UAE, Yemen	Camels, cattle, sheep	(Abdel-Shafy et al., 2012; Bitam et al., 2006; Hasson, 2012; Leulmi et al., 2016; Omer et al., 2007)
		H. nitidum	Mauritania		(Sylla et al., 2008)
		H. rufipes	Algeria, Egypt, Libya, Mauritania, Oman, Saudi Arabia, Sudan , Yemen	Camels, cows	(Camicast et al., 1990; Djerbouh et al., 2012; El Kammah et al., 2001)
		H. scupense	Algeria, Iraq, Jordan, Sudan, Tunisia	Cattle, sheep	(Bitam et al., 2006; Hasson, 2012; Leulmi et al., 2016; Saliba et al., 1990; Shamsuddin & Mohammad, 1988)
		H. schulzei	Jordan, Kuwait, Lebanon, Saudi Arabia	Domestic animals	(Alanazi, Al-Mohammed, et al., 2018; El-Rabie et al., 1990; Saliba et al., 1990)
		H. truncatum	Egypt, Mauritania, Sudan, UAE	Livestock, camels	(Camicast et al., 1990; El Kammah et al., 2001; Khan et al., 1997)
		H. turanicum	Egypt, Iraq, Jordan, Libya, Saudi Arabia	Sheep, goats	(El-Rabie et al., 1990; Hasson, 2012; Shamsuddin & Mohammad, 1988)
	Ixodes	*I. hoogstraali*	Oman		(FGS, 1999)
		I. ricinus	Algeria, Tunisia	Cattle, sheep	(Bouattour et al., 1999; Dib, Bitam, Bensouilah, Parola, & Raoult, 2009; Leulmi et al., 2016)
		Ixodes sp.	Jordan	Animals	(Saliba et al., 1990)
	Rhipicephalus	*R. annulatus*	Algeria, Egypt, Iraq, Jordan, Kuwait, Lebanon, Libya, Oman, Sudan, Syria, Tunisia, Yemen	Cattle	(Abdel-Shafy, 2000; Abdel-Shafy et al., 2012; Asmaa et al., 2014; El Kammah et al., 2001; Hassan, Gabr, Abdel-shafy, Hammad, & Mokhtar, 2017b; Pegram et al., 1982)
		R. appendiculatus	Sudan, UAE	Livestock, camels	(Khan et al., 1997)
		R. bursa	Algeria, Iraq, Jordan, Lebanon, Libya, Tunisia	Sheep, goats	(Bitam et al., 2006; Omer et al., 2007; Shamsuddin & Mohammad, 1988)

Table 1: Tick species distribution in Arab countries (Continued)

Families	Genera	Species	Arab Countries	Hosts	References
		R. camicasi	Egypt, Jordan, Saudi Arabia, Sudan, Yemen	Sheep	(El Kammah et al., 2001; Saliba et al., 1990)
		R. decoloratus	Libya, Sudan, Yemen	Cattle	(Abaker et al., 2017; Camicast et al., 1990; El-Tigani & Mohammed, 2010; Gabaj et al., 1992)
		R. evertsi	Libya, Mauritania, Sudan, UAE, Yemen	Livestock, camels	(Gabaj et al., 1992; Khan et al., 1997; Nabeth et al., 2004)
		R. geigyi	Mauritania	Livestock	(Camicast et al., 1990)
		R. guilhoni	Egypt, Mauritania, Sudan	Sheep	(Camicast et al., 1990; El Kammah et al., 2001)
		R. kohlsi	Jordan, Iraq, Jordan, Saudi Arabia, Yemen	Goats, sheep	(Pegram et al., 1982; Shamsuddin & Mohammad, 1988)
		R. microplus	Libya	Goats	(Gabaj et al., 1992)
		R. muhsamae	Sudan	Cattle	(Ali, 2007)
		R. praetextatus	Saudi Arabia, Sudan	Livestock	(Diab, Al-Khalifa, Al-Asgah, Hussein, & Khalil, 2006)
		R. pulchellus	Egypt, UAE	Camels, livestock	(Hassan et al., 2017a; Khan et al., 1997)
		R. sanguineus	Algeria, Egypt, Iraq, Jordan, Kuwait, Lebanon, Libya, Mauritania, Oman, Saudi Arabia, Sudan, Syria, Tunisia, UAE, Yemen	Livestock	(Bitam et al., 2006; Nabeth et al., 2004; Pegram et al., 1982; Pegram, Zivkovic, & Jongejan, 1989)
		R. simus	Sudan, Yemen	Sheep, cattle, camels	(Ahmed, El Hussein, & El Khider, 2005; Pegram et al., 1982, 1989)
		R. sulcatus	UAE	Livestock, camels	(Khan et al., 1997)
		R. turanicus	Algeria, Egypt, Iraq, Jordan, Lebanon, Oman, Saudi Arabia, Sudan, Tunisia, Yemen	Cattle, sheep, goats	(Asmaa et al., 2014; Bitam et al., 2006; El Kammah et al., 2001; Omer et al., 2007; Shamsuddin & Mohammad, 1988)

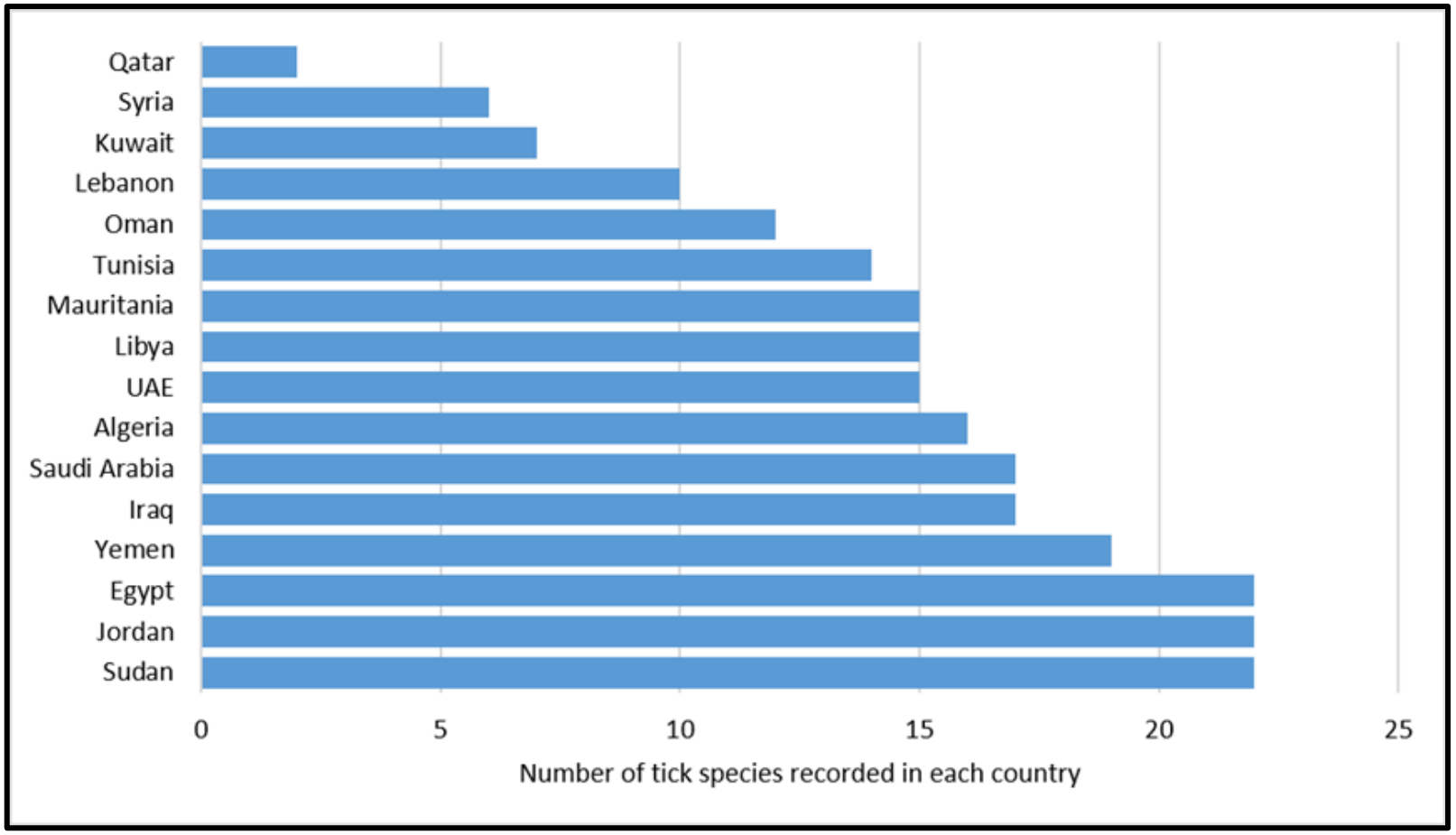

Figure 2: Arab countries with number of tick species reported.

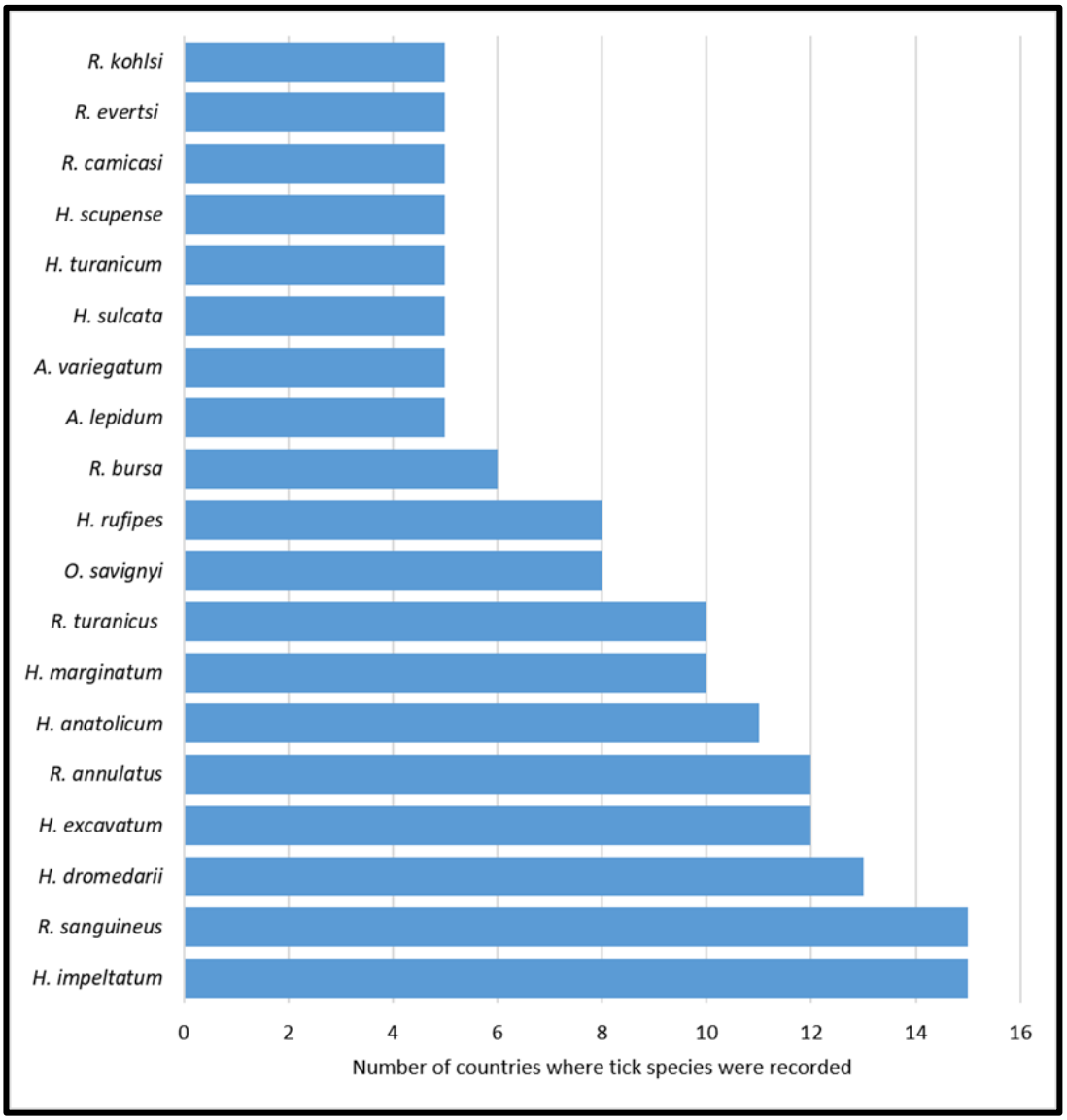

Figure 3: Tick species reported in more than four countries in the MENA region.

1.4.5 Tick Microbiota and Next-Generation Sequencing (NGS) Technology

Vector-borne pathogens and the diseases they cause are of high significance to human and animal health (Cunningham et al., 2017; Daszak, Cunningham, & Hyatt, 2000). Infections such as parasitic and microbial are common in the populations of animals and humans, and pathogenic organisms are often found abundant in healthy ecosystems (Cable et al., 2017; Cunningham et al., 2017). In recent years, due to

extensive anthropogenic changes in the environment, long-term host-pathogen associations disrupted primarily and resulting in emerging and re-emerging infectious diseases (Cunningham et al., 2017; Daszak et al., 2000). In any geographic area, the density of hosts, vectors, and pathogens are determinants of disease transmission (Martinez & Merino, 2011). Globally, ticks act as carriers and vectors of many human and animal pathogens including viruses, bacteria, and protozoa, often mediate the transfer of diseases from one host species to another (Drew & Samuel, 1987; Estrada-Pena, Aviles, & Munoz, 2011; Jongejan & Uilenberg, 2004; Magnarelli, 2009; Pfäffle et al., 2013). To meet the demands of growing populations, farming of animals across the world resulted in increased domestic animal populations artificially. Consequently, tick abundance and distribution have been increased, mainly in peri-urban livestock industries. Tick acquire various endosymbionts and pathogenic microbes by feeding exclusively on the blood of their vertebrate hosts (Sonenshine & Roe, 2014; Piesman & Eisen, 2008; Scoles, 2004). The collection of bacteria retained and transmitted by ticks represents a wide range of genera such as *Anaplasma, Borrelia*, *Coxiella, Cowdria, Ehrlichia, Francisella,* and *Rickettsia* and these bacteria are adapted to undergo development in the tick vector for at least a portion of their lifecycle (Noda, Munderloh, & Kurtti, 1997; Scoles, 2004). The bacterial diseases such as rickettsial infections transmitted by arthropod vectors affect about one billion people globally, though humans are considered as accidental hosts (Parola, Paddock, & Raoult, 2005; Walker & Ismail, 2008).

Next-Generation Sequencing (NGS) technology has revolutionized genomic research by providing opportunities to analyze substantial amounts of genetic data at a reduced cost (Kchouk, Gibrat, & Elloumi, 2017; Kulski, 2016; Loman et al., 2013; Mardis, 2011). Increasingly, microbiome studies are utilizing newer NGS platforms,

such as the Illumina MiSeq, which have been reported to be more cost-effective and accurate (Loman et al., 2013; Quail et al., 2012). The V4 hypervariable region provides adequate data for taxonomic profiling of microbial communities and has revealed a lower error rate on the Illumina platform, therefore, it is usually selected for work on the MiSeq (Kozich, Westcott, Baxter, Highlander, & Schloss, 2013). Metagenomic approaches allow the investigation of entire bacterial microbiota associated with their vectors allowing better assessment of the diversity of circulating microbes and the reservoir potential of vectors (Boissiere et al., 2012; Carpi et al., 2011; Walter, Carpi, Evans, & Caccone, 2016). For example, selected genera of bacterial communities inhabiting the salivary glands of *Anopheles* mosquitoes have been linked with enhanced transmission of *Plasmodium* (Boissiere et al., 2012). Knowledge about the bacterial community structure associated with ticks is very limited. There is an increasing number of studies that have utilized metagenomic analysis of viral and microbial communities associated with ticks (Carpi et al., 2011; Karim et al., 2017; Khoo et al., 2016; Thapa, Zhang, & Allen, 2019; Walter et al., 2016; Xu et al., 2011; Zhuang et al., 2018). The formation of the microbial communities may be determined by the host specificity of the microbes, perhaps certain bacterial genera dominate in certain tick host species (Qiu, Nakao, Ohnuma, Kawamori, & Sugimoto, 2014). The majority of the microbes appear to be endosymbionts and in their tick hosts, some of these endosymbionts often form complex interactions with pathogenic microbes (Gall et al., 2016; Khoo et al., 2016). The diversity of microbial communities changes due to environmental conditions including temperature (Thapa et al., 2019) has been reported previously which suggests that seasonality of the microbiota, could be linked with the seasonality of pathogen transmission. Previously, the presence of some endosymbionts, such as *Rickettsia bellii* in *Dermacentor andersoni* ticks, is often

associated with lowered infection rates of pathogenic species such as *Anaplasma marginale*, proposing that endosymbionts may play an essential role in suppressing pathogen transmission (Gall et al., 2016). Consequently, knowledge about the classification of microbial communities inside ticks is of great significance to understandng the transmission of pathogens to animals and humans (Gall et al., 2016; Thapa et al., 2019). In the MENA region, so far there is a limited record of the composition of microbial communities in ticks (Alreshidi, 2020; Elbir, Almathen, & Alhumam, 2019; Ravi et al., 2019).

1.4.6 Tick-Borne Diseases in the MENA Region

Vector-borne zoonotic diseases emerge or re-emerge in many parts of the world and causing global health issues that involve humans, pathogens, vectors and wildlife (Collinge & Ray, 2006; Harrus & Baneth, 2005). Various tick-borne diseases such as viral, bacterial, and protozoan have been reported in the Arabian Peninsula (Table 2). Ticks can be infected with bacteria, viruses or protozoa (Bowman & Nuttall, 2008; Bratton & Corey, 2005; de la Fuente, Estrada-Pena, Venzal, Kocan, & Sonenshine, 2008; Gayle & Ringdahl, 2001; Jongejan & Uilenberg, 2004; Nicholson, Sonenshine, Lane, & Uilenberg, 2009). Co-infection of multiple tick-borne pathogens is usually found in dogs (Kordick et al., 1999; Suksawat, Pitulle et al., 2001; Suksawat, Xuejie et al., 2001). The persistence and transmission of vector-borne diseases depend on the overlapping distributions of hosts and vectors along with the favorable environmental conditions for a given pathogen (Cunningham et al., 2017). Tick-borne diseases such as babesiosis, theileriosis, anaplasmosis, and cowdriosis constitute one of the main difficulties for the development of the livestock breeding industry and cause serious economic losses (Iqbal, Song-hua, Wan-jun, G, & Chen-wen, 2006;

Jongejan & Uilenberg, 2004). Most of the arthropod-borne infections may be associated with 116 tick species (32 argasid species and 84 ixodids), from the medical and veterinarian viewpoint. Moreover, tick-borne diseases are common in the medical and veterinary clinical settings and good surveillance is crucial for the management of such zoonotic diseases (Dantas-Torres, Chomel, & Otranto, 2012).

In the MENA region, the ticks that carry pathogens and transmit diseases belong to both the soft-tick family, Argasidae including genera, *Argas*, *Ornithodoros*, and *Otobius*), as well as the hard-tick family, Ixodidae including genera, *Dermacentor, Hyalomma, Haemaphysalis, Ixodes, Amblyomma* and *Rhipicephalus* (A'aiz & Dhaim, 2014; Abaker et al., 2017; Alkishe, Peterson, & Samy, 2017; Babudieri, 1957; El Kammah et al., 2001; Hassan et al., 2017a). The major tick-borne diseases in the tropical region consist of theileriosis, babesiosis, cowdriosis, and anaplasmosis (Ali, 2007). However, following main tick-borne diseases of human and animal importance in the MENA region have been reported: protozoan diseases e.g. babesiosis and theileriosis; bacterial diseases e.g. anaplasmosis, ehrlichiosis, Lyme borreliosis, Mediterranean Spotted Fever, Spotted Fever Rickettsioses, Tick-borne Relapsing Fever, and Tularemia; and viral diseases e.g. Alkhurma Hemorrhagic Fever, Crimean Congo Hemorrhagic Fever, and Tick-borne encephalitis (Table 2). The distributions of widely occurring four tick-borne diseases, CCHF, anaplasmosis, babesiosis and theileriosis in the Arab countries are shown in Figure 4.

Table 2: Tick-borne diseases, pathogens, vectors and hosts in Arab countries.

Pathogens	Host species	Tick vectors	Locality	References
CCHFV	Camels, sheep	*H. anatolicum, H. marginatum H. rufipes, H. impeltatum, R. sanguineus, R. turanicus, R. annulatus*	Egypt	(Darwish, Imam, Omar, & Hoogstraal, 1978)
CCHFV	Human, livestock		Egypt	(Hoogstraal, 1979)
CCHFV	Patients		United Arab Emirates	(Suleiman et al., 1980)
CCHFV	Sheep, cattle	*H. marginatum*	Iraq	(Al-Tikriti et al., 1981)
CCHFV	Died patient		United Arab Emirates	(Baskerville, Satti, Murphy, & Simpson, 1981)
CCHFV	Patients		Kuwait	(Al-Nakib et al., 1984)
CCHFV	Patients		Mauritania	(Saluzzo, Aubry, & Digoutte, 1985)
CCHFV	Patients		Mauritania	(Gonzalez, LeGuenno, Guillaud, & Wilson, 1990)
CCHFV	Camel		Egypt	(Morrill et al., 1990)
CCHFV	Livestock	*Hyalomma* sp.	Oman	(Scrimgeourl et al., 1996)
CCHFV	Patients		United Arab Emirates	(Schwarz, Nsanze, & Ameen, 1997)
CCHFV	Camels, cattle, goat	*H. impeltatum H. excavatum H. anatolicum*	United Arab Emirates	(Khan et al., 1997)
CCHFV	Livestock	*Hyalomma* sp.	United Arab Emirates	(Rodriguez et al., 1997)
CCHFV			Saudi Arabia	(Hassanein, El-Azazy, & Yousef, 1997)
CCHFV	Domestic livestock	*H. anatolicum, R. evertsi*	Oman	(Williams et al., 2000)
CCHFV	Livestock	*A. variegatum, R. decoloratus, R. geigyi, H. impeltatum, H. impressum, H. rufipes, H. truncatum, R. guilhoni*	Mauritania	(Nabeth et al., 2004)
CCHFV	Patients		Sudan	(Elata et al., 2011)
CCHFV	Patients		Sudan	(Aradaib et al., 2011)
CCHFV	Livestock	*Hyalomma* sp.	Iraq	(Abul-Eis, 2012)

Table 2: Tick-borne diseases, pathogens, vectors and hosts in Arab countries (Continued).

Pathogens	Pathogens	Pathogens	Pathogens	Pathogens
CCHFV	Patients		Oman	(Al-Zadjali, Al-hashim, Al-gh, & Balkha, 2013)
CCHFV	Patients		Sudan	(Osman et al., 2013)
CCHFV		*H. marginatum*	Iraq, UAE, Oman, Yemen, Saudi Arabia, Mauritania	(Cecaro, Isolani, & Cuteri, 2013)
CCHFV	Cow		Egypt	(Horton et al., 2014)
CCHFV	Cattle		Sudan	(Ibrahim, Adam, Osman, & Aradaib, 2015)
CCHFV	Cattle, camel, sheep, goat	*Hyalomma* sp.	Oman	(Body et al., 2016)
CCHFV	Patients		United Arab Emirates	(Al-Dabal et al., 2016)
CCHFV	Cattle, camel, sheep		Oman	(Body et al., 2016)
CCHFV	Patients		Mauritania	(Kleib et al., 2016)
CCHFV	Patients		Tunisia	(Wasfi et al., 2016)
CCHFV			Algeria	(Kautman, Tiar, Papa, & Široký, 2016)
CCHFV	Cattle		Mauritania	(Sas et al., 2017)
CCHFV	Camels		Sudan	(Suliman et al., 2017)
CCHFV			Egypt	(Helmy, El-Adawy, & Abdelwhab, 2017)
CCHFV	Humans		Iraq, Kuwait, Oman, Saudi Arabia, Sudan, UAE	(Al-Abri et al., 2017)
CCHFV	Livestock		Egypt, Somalia, Tunisia	(Al-Abri et al., 2017)
CCHFV	Patient		Oman	(Al-Abri et al., 2019)
CCHFV	Cattle, camel	*H. rufipes, H. dromedarii, H. impeltatum*	Mauritania	(Schulz et al., 2021)
CCHFV	Camel	*H. dromedarii*	United Arab Emirates	(Camp et al., 2020)
ALKV	Patients		Saudi Arabia	(Charrel et al., 2005)

Table 2: Tick-borne diseases, pathogens, vectors and hosts in Arab countries (Continued).

Pathogens	Pathogens	Pathogens	Pathogens	Pathogens
ALKV	Patients			(Madani, 2005)
ALKV	Patient, dead camel		Saudi Arabia	(Charrel, Zaki, Fagbo, & de Lamballerie, 2006; Charrel et al., 2005)
ALKV	Camels	*O. savignyi*	Saudi Arabia	(Charrel, Fagbo, & Moureau, 2007)
ALKV	Camel, sheep	*H. dromedarii*	Arabian Peninsula	(Memish, Charrel, Zaki, & Fagbo, 2010)
ALKV	Human		Egypt	(Carletti et al., 2010)
ALKV	Human		Najran, Saudi Arabia	(Alzahrani et al., 2010)
ALKV	Soldier		Jazan, Saudi Arabia	(Memish et al., 2011)
TBEV	Human, livestock	*I. ricinus*	Europe, North Africa, Middle East	(Alkishe et al., 2017)
Rickettsia africae	Humans	*O. foleyi*	Libya	(Franchini & Taddia, 1930)
R. africae	Humans		Mauritania	(Niang et al., 1998)
R. africae	Cattle	*A. variegatum, R. appendiculatus, R. microplus*	Comoros	(Yssouf et al., 2014)
Anaplasma. marginale	Cattle		Libya	(El-Maghrbi, El-Sayed, Hassaneen, & Ezeldin, 2008)
A. marginale	Cattle	*R. annulatus*	Sudan	(Awad et al., 2011)
A. marginale	Cattle, sheep, goat	*R. sanguineus, R. turanicus, H. excavatum, H. anatolicum, H. marginatum, H. turanicum, H. scupense, R. annulatus*	Iraq	(Ameen, Abdullah, & Abdul-Razaq, 2012)
A. ovis	Sheep	*Rhipicephalus* sp.	Iraq	(Renneker et al., 2013)
A. marginale, A. central A. bovis	Cattle		Tunisia	(Belkahia et al., 2015)
A. marginale	Cattle		Libya	(Al-Bassam, Al-Garib, El-Attar, Abdunaser, & Abdouslam, 2016)
A. marginale	Buffalo		Egypt	(Elhariri, Elhelw, Hamza, & Soliman, 2017)

Table 2: Tick-borne diseases, pathogens, vectors and hosts in Arab countries (Continued).

Pathogens	**Pathogens**	**Pathogens**	**Pathogens**	**Pathogens**
Anaplasma sp., *Anaplasma platys*	Livestock	*Rhipicephalus* sp., *R. sanguineus*	Palestine	(Zaid, Ereqat, Nasereddin, & Al-jawabreh, 2019)
Rickettsia conorii		*R. sanguineus*	Libya	(Giordano & Nastasi, 1935)
Ehrlichia ruminatium	Livestock	*A. variegatum, A. lepidum*	Sudan	(Muramatsu et al., 2005)
Ehrlichia sp.	Livestock		Egypt	(Loftis et al., 2006)
Ehrlichia sp.	Livestock	*R. sanguineus*	Palestine	(Zaid et al., 2019)
Rickettsia sp.	Humans	*R. sanguineus*	Algeria, Morocco, Sudan	(Hoogstraal, 1967)
R. conorii	Humans		Mauritania	(Niang et al., 1998)
R. conorii	Camels	*H. dromedarii*	Tunisia	(Demoncheaux et al., 2012)
R. conorii	Humans	*R. sanguineus*	South Jordan	(Nafi, Tarawnah, & Tarawnah, 2017)
R. aeschlimannii	Livestock	*Hyalomma* sp.	Egypt	(Loftis et al., 2006)
R. aeschlimannii	Cattle, sheep	*H. marginatum, H. scupense*	Algeria	(Bitam et al., 2006)
R. conorii, R. aeschlimannii	Camels	*H. dromedarii*	Tunisia	(Demoncheaux et al., 2012)
Rickettsia sp.	Camels	*H. dromedarii*	UAE	(Al-Deeb, Muzaffar, Abu-Zeid, Enan, & Karim, 2015)
Coxiella burnetii	Humans		Libya	(Fellers, 1952)
C. burnetii	Humans		Mauritania	(Niang et al., 1998)
Borrelia burgdorferi, B. lusitaniae, B. garinii		*I. ricinus*	Tunisia	(Younsi, Postic, Baranton, & Bouattour, 2001)
B. burgdorferi	Humans	*I. scapularis*	Egypt	(Adham, Abdel-samie, Gabre, & Hala, 2010)
B. burgdorferi	Humans		Egypt	(Elhelw, El-enbaawy, & Samir, 2014)
B. burgdorferi	Human, livestock	*I. ricinus*	Europe, North Africa, Middle East	(Alkishe et al., 2017)
Borrelia sp.	Humans	*Ornithodoros* sp.	Sudan	(Kirk, 1939)
	Humans	*O. tholozani*	Libya	(Coghill, Lawrence, & Ballantine, 1947)
Borrelia sp.	Humans	*Ornithodoros* sp.	Egypt	(Davis & Hoogstraal, 1954)

Table 2: Tick-borne diseases, pathogens, vectors and hosts in Arab countries (Continued).

Pathogens	**Pathogens**	**Pathogens**	**Pathogens**	**Pathogens**
Borrelia sp.	Humans	*O. tholozani*	Jordan	(Babudieri, 1957)
Borrelia persica	Livestock	*O. tholozani*	Syria, Egypt	(Rebaudet & Parola, 2006)
Borrelia sp.		*Ornithodoros* sp.	Iraq, Syria, Jordan, Egypt	(Assous & Wilamowski, 2009)
Francisella tularensis	Human		Egypt	(Trevisanato, 2004)
F. tularensis	Human		Iraq, Syria	(Trevisanato, 2007)
F. tularensis	Human		Syria, Egypt, Lebanon	(Gürcan, 2014)
Francisella spp.	Camel	*H. dromedarii*	Egypt	(Ghoneim, Abdel-Moein, & Zaher, 2017)
Babesia bigemmina, B. ovis	Camel, cows, sheep	*Hyalomma* sp., *Boophilus* sp., *Rhipicephalus* sp., *Amblyomma* sp., *Argas* sp.	Egypt	(El Kammah et al., 2001)
B.bigemina	Cattle		Libya	(El-Maghrbi et al., 2008)
B. bovis, B. bigemina	Cattle	*R. annulatus, I. ricinus, H. punctata, H. sulcata, H.excavatum, H. scupense, H. marginatum*	Tunisia	(M'ghirbi et al., 2008)
Babesia spp.	Livestock	*H. scupense, R. bursa*	Tunisia	(M'ghirbi, Hurtado, & Bouattour, 2010)
B. bigemina, B. bovis	Cattle	*R. annulatus*	Sudan	(Awad et al., 2011)
B. occultans		*H. marginatum*	Tunisia	(Ros-García, M'ghirbi, Bouattour, & Hurtado, 2011)
Babesia sp.	Cattle	*H. anatolicum*	Iraq	(Hadi & Al-Amery, 2012)
B. bigemina	Cattle, sheep, goat	*R. sanguineus, R. turanicus, H. excavatum, H. anatolicum, H. marginatum, H. turanicum, H. scupense, R. annulatus*	Iraq	(Ameen et al., 2012)
B. ovis	Sheep	*Rhipicephalus* sp.	Iraq	(Renneker et al., 2013)
Babesia sp.	Camel		Saudi Arabia	(Swelum, Ismael, Khalaf, & Abouheif, 2014)
B. ovis, B. motasi, B. foliate B.taylori	Sheep, goat	*H. anatolicum*	Iraq	(Abdullah, 2014)

Table 2: Tick-borne diseases, pathogens, vectors and hosts in Arab countries (Continued).

Pathogens	**Pathogens**	**Pathogens**	**Pathogens**	**Pathogens**
Babesia sp.	Camel, cow	*O. savignyi, R. annulatus*	Egypt	(Hassan et al., 2017b)
Theileria annulata	Calf	*H. anatolicum*	Bahrain	(Uilenberg, Franssen, & Perié, 1986)
T. annulata	Cow	*H. dromedarii*	Mauritania	(Jacquiet et al., 1990)
T. annulata, T. ovis, T. hirci	Camel, Sheep, goat	*Ixodid* sp.	Saudi Arabia	(Hussein, Al-Asgah, Al-Khalifa, & Diab, 1991)
T. annulata		*H. dromedarii*	Mauritania	(Jacquiet et al., 1994)
T. annulata	Camels, cows, sheep	*Hyalomma* sp., *Rhipicephalus* sp., *Amblyomma* sp., *Argas* sp.	Egypt	(El Kammah et al., 2001)
T. lestoquardi	Sheep	*H. anatolicum*	Oman	(Tageldin, Fadiya, Sabra, & Ismaily, 2005)
T. mutans	Cattle		Libya	(El-Maghrbi et al., 2008)
T. annulata, T. buffeli	Cattle	*R. annulatus, I. ricinus, H. scupense, H. sulcata, H. punctata, H.excavatum, H. marginatum*	Tunisia	(M'ghirbi et al., 2008)
T. annulata	Cattle	*Hyalomma* sp.	Iraq	(Al-Saeed et al., 2010)
Theileria spp.	Livestock	*H. scupense, R. bursa*	Tunisia	(M'ghirbi et al., 2010)
T. lestoquardi	Sheep		Oman	(Shayan, Ebrahimzadeh, Tageldin, Amininia, & Eckert, 2011)
Theileria sp.	Cattle	*H. anatolicum*	Iraq	(Hadi & Al-Amery, 2012)
T. ovis, T. lestoquardi, T. uilenbergi	Sheep	*Rhipicephalus* sp.	Iraq	(Renneker et al., 2013)
T. ovis, T. annulata, T. lestoquardi	Sheep	*Ixodes* sp.	Iraq	(A'aiz & Dhaim, 2014)
T. annulata	Camel	*H. dromedarii*	Egypt	(Hassan et al., 2017a)
T. annulata	Camel	*H. dromedarii*	United Arab Emirates	(Al-Deeb et al., 2015)
T. annulata	Cattle		Oman	(Al-Hamidhi et al., 2015)

Table 2: Tick-borne diseases, pathogens, vectors and hosts in Arab countries (Continued).

Pathogens	Pathogens	Pathogens	Pathogens	Pathogens
T. annulata, T. ovis,T. lestoquardi	Cattle, sheep, goat	*H. anatolicum*	Oman	(Al-Fahdi et al., 2017)
T. annulata	Camel	*H. dromedarii*	Egypt	(Hassan et al., 2017a)

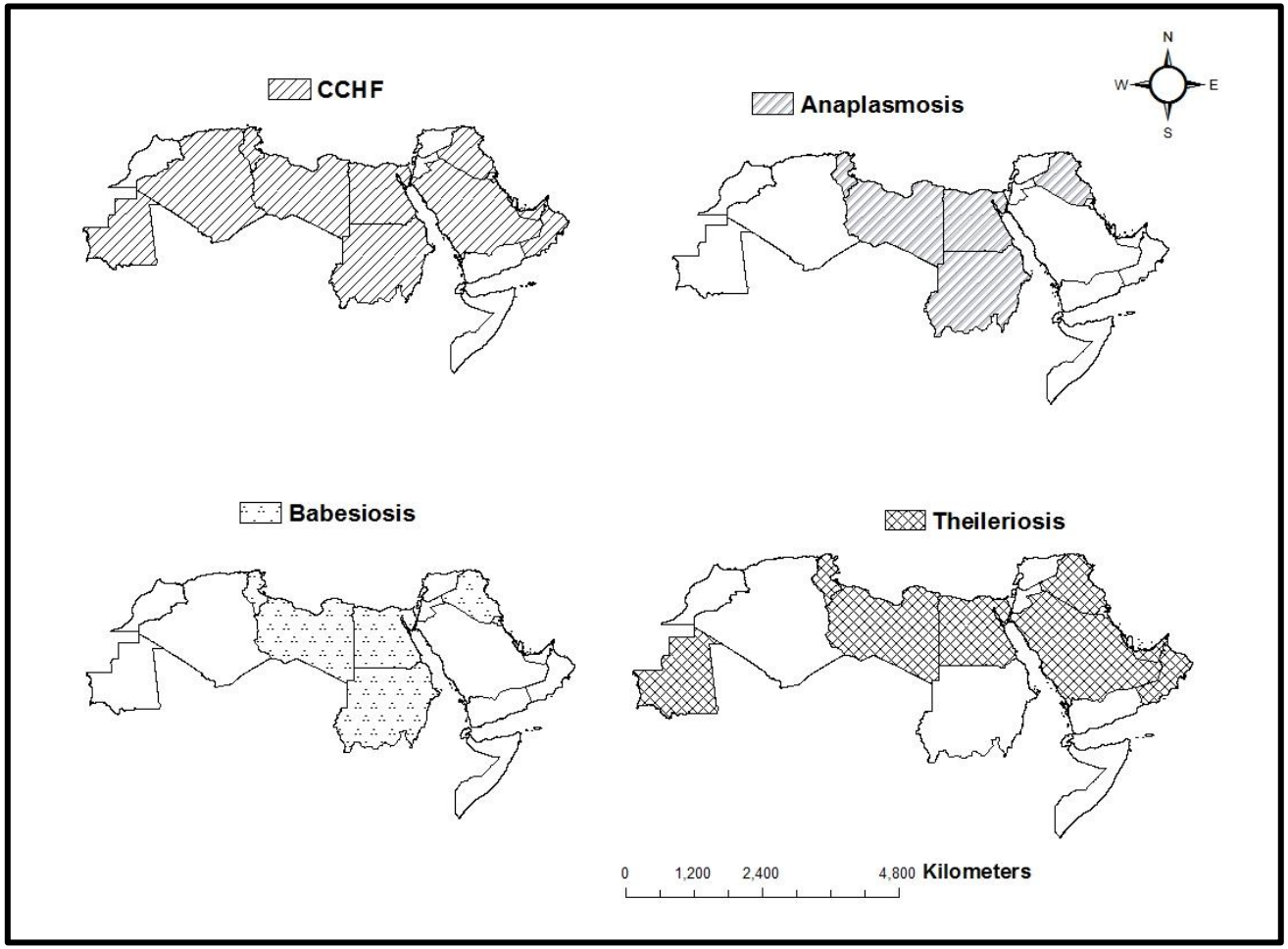

Figure 4: Maps show the distribution of tick-borne diseases. CCHF, Anaplasmosis, Babesiosis and Theileriosis in the Arab countries.

1.4.6.1 Tick-Borne Viral Diseases in the MENA Region

1.4.6.1.1 Alkhurma Hemorrhagic Fever Virus (ALKV)

Alkhurma Hemorrhagic Fever Virus (ALKV) virus is a member of the tick-borne hemorrhagic fever group of the genus *Flavivirus*. Livestock including camels and sheep are considered the hosts of ALKV (Carletti et al., 2010). It was detected in an *O. savignyi* collected in Jeddah, Saudi Arabia (Charrel et al., 2007). In the MENA region, these ticks have been associated with camels and their resting places, and also found from the same places where the cases of ALKV infections in humans occurred. In the Arabian Peninsula, the Alkhurma virus has been reported from Egypt (Carletti et al., 2010) and Saudi Arabia (Alzahrani et al., 2010; Charrel et al., 2001, 2005; Madani, 2005; Memish et al., 2011) (Table 2).

1.4.6.1.2 Crimean-Congo Hemorrhagic Fever (CCHF)

Crimean-Congo Hemorrhagic Fever (CCHF) has been reported worldwide from more than 30 countries in Africa, Asia, South-East Europe, and the Middle East and the majority of CCHF infections have been reported in workers associated with the livestock industry, slaughterhouses, and veterinary practice (Ergönül, 2006; Whitehouse, 2004). The infection of CCHF was reported asymptomatic in animal species including cattle, sheep, goats, and camels (Schwarz et al., 1997; Wernery, 2014). Thirty species of ticks, mainly the genus *Hyalomma*, act as vectors and reservoirs for the Crimean-Congo Hemorrhagic Fever virus (CCHFV). CCHF can be transmitted to humans by tick bites or through close contact with infected animals and humans (Ergönül, 2006). Ticks belong to the genus, *Hyalomma* were probably responsible for outbreaks of CCHF in humans with a high fatality rate in the UAE,

Oman, and Saudi Arabia (Scrimgeourl et al., 1996; Suleiman et al., 1980). CCHFV in ticks can be transmitted transstadially (from larva to nymph to adult) and transovarially in various tick species including *H. marginatum*, *H. rufipes*, *D. marginatus* and *Rhipicephalus rossicus* (Hoogstraal, 1979). CCHFV has been reported from almost all countries of the Arabian Peninsula (Table 2) (Al-Abri et al., 2017; Al-Tikriti et al., 1981; Al-Zadjali et al., 2013; Cecaro et al., 2013; Darwish et al., 1978; Horton et al., 2014; Ibrahim et al., 2015; Rodriguez et al., 1997; Scrimgeourl et al., 1996; Wasfi et al., 2016; Williams et al., 2000).

1.4.6.1.3 Tick-Borne Encephalitis (TBE)

Tick-Brne Encephalitis (TBE) is caused by the tick-borne encephalitis virus (TBEV) which belongs to the genus *Flavivirus*, has been a rising public health concern in Europe and other parts of the world for decades (Kunze, 2016), and ticks are considered both reservoirs and vectors TBEV (Katargina et al., 2013). Humans may get TBE disease by consuming unpasteurized milk and cheese (Mansfield et al., 2009) and via the bite of an infected tick (*I. ricinus* or *I. persulcatus*). *Ixodes ricinus* and *I. persulcatus* can acquire from their vertebrate hosts, wild and domestic animals and the virus occurrence in ticks can vary significantly within and among areas of risk (Kunze, 2016). Mammals and migratory birds species may also play a significant role in the transmission and distribution of TBEV (Soleng et al., 2018). TBE cases have been reported in Saudi Arabia in 1995 (Zaki, 1997). No causal treatment is known yet, infections can be prevented by avoiding bites of ticks and vaccination (Kunze, 2016). Therefore, this could be an emerging disease in the MENA region.

1.4.6.2 Tick-Borne Bacterial Diseases in the MENA Region

1.4.6.2.1 Anaplasmosis

Anaplasmosis is reported in tropical and subtropical areas across the world and in many countries, and this disease is a major constraint to cattle production (Kocan, Fuente, Blouin, Coetzee, & Ewing, 2010). Anaplasmosis in humans and animals, caused by *Anaplasma* species including *A. marginale, A. centrale, A. bovis,* and *A. ovis* for ruminants, and *A. phagocytophilum* for human and domestic animals (Inci, Yildirim, Duzlu, Doganay, & Aksoy, 2016). The infection in humans is called 'Human Granulocytic Anaplasmosis' (Dumler et al., 2005), and in cattle is called bovine anaplasmosis (Kocan et al., 2010). Belkahia et al. (2015) reported three *Anaplasma* species including *A. marginale, A. centrale*, and *A. bovis*, from Tunisian cattle. Whereas, Awad et al. (2011) reported *A. marginale* in cattle from Sudan, and *R. annulatus* was the vector. Anaplasmosis has also been reported from other countries including Libya, Egypt, Iraq, and Palestine (Table 2) (Elhariri et al., 2017; Renneker et al., 2013; Zaid et al., 2019).

1.4.6.2.2 Ehrlichiosis

Ehrlichiosis has been detected in both humans and animals. In humans, it is called 'Human Monocytotropic Ehrlichiosis' and is caused by *A. phagocytophilum, Ehrlichia chaffeesis,* and *Ehrlichia ewingii*. Ixodid ticks transmit this disease (Inci et al., 2016). In the MENA region, ehrlichiosis is reported from Egypt, Palestine, and Sudan (Table 2) (Loftis et al., 2006; Muramatsu et al., 2005; Zaid et al., 2019).

1.4.6.2.3 Spotted Fever Rickettsioses (SFR)

Spotted Fever Rickettsioses (SFR) is an important tick-borne disease group and comprises Pacific Coast Tick Fever, Rocky Mountain Spotted Fever (RMSF), and Rickettsialpox. Rickettsial pathogens are categorized into the order Rickettsiales, which includes the family Rickettsiaceae and Anaplasmataceae (Dumler et al., 2001; Raoult & Roux, 1997). Most of the rickettsial diseases are caused by infection with obligate intracellular Gram-negative bacteria transmitted by arthropod vectors (Parola et al., 2005; Walker & Ismail, 2008). In the MENA region, spotted fever group *Rickettsia* sp. was reported in *H. dromedarii* in the UAE (Al-Deeb et al., 2015), *Rickettsia aeschlimannii* was detected in *H. dromedarii* from Tunisia (Demoncheaux et al., 2012) and *R. aeschlimannii* was detected in *Hyalomma* sp. from Egypt (Loftis et al., 2006).

1.4.6.2.4 Mediterranean Spotted Fever (MSF)

Mediterranean Spotted Fever (MSF) was first described as an infection associated with high fever and spots, a century ago. The disease was 1st reported in Tunisia in 1910, and after that, it was reported in other regions around the Mediterranean basin (Parola et al., 2005). This is caused by *Rickettsia conorii* which is a gram-negative, obligate intracellular bacterium that is extremely fastidious (Parola et al., 2005). *Rhipicephalus sanguineus* is the main vector of this disease. Humans have no role in maintaining this bacterium in nature and are an accidental host of *Rickettsia* (Nafi et al., 2017). People of all ages can be affected by this bacterium. This disease has been reported from the MENA region including Algeria, Morocco, Sudan, Jordan, Mauritania, and Tunisia (Table 2) (Demoncheaux et al., 2012; Hoogstraal,

1967; Nafi et al., 2017; Niang et al., 1998).

1.4.6.2.5 Lyme Borreliosis

Lyme Borreliosis is transmitted by hard ticks of the genus *Ixodes* (Arco, Dattwyler, & Arnaboldi, 2017; Margos et al., 2009) and is most common in temperate forested regions of North America, Europe, and Asia (Kilpatrick et al., 2018). This disease is caused by members of the *B. burgdorferi* sensu lato (s.l.) species-complex including *B. burgdorferi* sensu stricto in North America (Pritt et al., 2016) and five species in Europe, *Borrelia afzelii, B. garinii, B. burgdorferi, Borrelia spielmanii* and *Borrelia bavariensis* (Stanek, Wormser, Gray, & Strle, 2012). *Borrelia burgdorferi* is transmitted by nymphs of *I. scapularis* to humans more frequently than adults. Efficient transmission of this spirochete requires a minimum of 24 to 48 hours of tick attachment, at which time the nymph obviously is engorged with blood (Re III, Occi, & Macgregor, 2004). Prediction of disease risk for better target control interventions requires an understanding of the pathogen and disease dynamics (Kilpatrick et al., 2018). Various pathogens such as *B. burgdorferi, B. lusitaniae, B. garinii* were reported in *I. ricinus* from Tunisia (Younsi et al., 2001). However, *B. burgdorferi* was detected in ixodid ticks (*R. annulatus, H. dromedarii, H. excavatum* and *R. sanguineus*) and soft ticks (*O. savignyi*) from Egypt in 2010 and 2014 (Adham et al., 2010; Elhelw et al., 2014) and in *I. ricinus* from other Middle East and North African countries (Alkishe et al., 2017) (Table 2).

1.4.6.2.6 Tick-Borne Relapsing Fever (TBRF)

Relapsing fever cases were reported from Sudan in 1939 (Kirk, 1939), from Egypt in 1954 (Davis & Hoogstraal, 1954), and from Jordan in 1957 in humans

(Babudieri, 1957). TBRF is caused by spirochete *Borrelia* mainly. Further, this disease is transmitted by *Ornithodoros* which is the most important tick vector, found in Iraq, Syria, Jordan, and Egypt (Assous & Wilamowski, 2009). Previously, a louse-borne relapsing fever outbreak in Sudan had probably affected 20,000 members of the Dinka tribe in 1998 and 1999; the death rate was 10%–14% (Cutler, 2006). Further, tick-borne relapsing fever was reported as in clusters of infection and was associated with exposure of susceptible human hosts to tick vectors and domestic animals. In the Middle East, *B. persica* infections were transmitted by *O. tholozani* vector, in 2005 (Cutler, 2006).

1.4.6.2.7 Tularemia

Tularemia is a zoonotic disease that is caused by the gram-negative bacterium *F. tularensis* (Eisen, 2007; Jellison, 1974). Ticks serve as a reservoir and vector and play important role in the epidemiology of tularemia. In addition, ticks can carry the bacterium by both transovarial and transstadial transmission (Gürcan, 2014). The main vectors responsible for this disease spread include *Amblyoma, Dermacentor*, and *Ixodes* species (Gürcan, 2014). In the MENA region, the genus *Francisella* is reported in *H. dromedarii* from Egypt (Ghoneim et al., 2017). Moreover, the presence of *F. tularensis* (IgG antibodies) in patients in Egypt (Ghoneim et al., 2017) poses a serious threat to the emergence and re-emergence of tularemia due to the high prevalence of tick species in this region.

1.4.6.3 Tick-Borne Protozoan Diseases in the MENA Region

1.4.6.3.1 Babesiosis

Babesiosis is caused by *Babesia* spp. to humans and animals, and can be transmitted by ixodid ticks transovarially and transstadially (Uilenberg, 2006). The disease is characterized by high temperature, anemia, restlessness, anorexia, and death. Babesiosis has been reported in domestic animals, including sheep and goats. The major economic impact of this infection is on the livestock industry worldwide due to high losses (Abdullah, 2014; Inci et al., 2016; Ranjbar-Bahadori, Eckert, Omidian, Shirazi, & Shayan, 2012). This infection was investigated in blood samples of sheep and goats in Kurdistan, Iraq from June to September 2012, and four *Babesia* species were detected: *B. ovis, B. motasi, B. foliata,* and *B. taylori*. One species of tick vector *H. anatolicum* was found on these animals (Abdullah, 2014). In the MENA region, *Babesia* spp. have been recorded from Egypt, Libya, Tunisia, Iraq, Sudan, Saudi Arabia (Table 2) (Awad et al., 2011; El-Maghrbi et al., 2008; Hadi & Al-Amery, 2012; Hassan et al., 2017b; Renneker et al., 2013; Swelum et al., 2014).

1.4.6.3.2 Theileriosis

Theileriosis has been studied in a wide range of livestock including cattle, sheep, and goats, and is one of the most common tick-borne diseases. Few people studied theileriosis-infected camels (El Kammah et al., 2001; Hassan et al., 2017a; Youssef & Yasien, 2015). This disease is caused by *Theileria*, obligate intracellular protozoan parasites, and transmitted by *Hyalomma* spp. which cause severe and mild infections in their vertebrate hosts (Hassan et al., 2017a). This pathogen (*Theileria* spp.) has been detected in all livestock species and can cause significant economic

losses to the farming industry (Florin-christensen & Schnittger, 2009). In the MENA region, *Theileria* sp. has been reported from Egypt, Sudan, UAE, Mauritania, Saudi Arabia, Oman, Libya, Tunisia, and Iraq (Table 2) (Al-Deeb et al., 2015; Al-Hamidhi et al., 2015; El-Maghrbi et al., 2008; Hadi & Al-Amery, 2012; Hassan et al., 2017a; Hussein et al., 1991; M'ghirbi et al., 2008; Renneker et al., 2013).

1.4.7 Global Travel and Trade and Infectious Diseases

With the emergence of a series of new diseases and the spread of HIV/AIDS across the world, infectious diseases have become an increasing priority in health policy and political agendas (Lederberg, Shope, & Oaks, 1992), however, due development of effective vaccines, antibiotics, and improved sanitation, infectious diseases were significantly reduced in the developed world in the 1970s. More than 60% of human infectious diseases that emerged between 1940 and 2004 were zoonotic, resulting in significant global morbidity, mortality, and economic costs (Jones et al., 2008). In the emerging zoonoses, 22.8% are arthropod vector-borne infections and 71.8% are from wildlife (Jones et al., 2008). The frequency of emerging vector-borne zoonoses has been increased during the last ten years significantly (Jones et al., 2008; Wikel, 2018). Because of their rapid spread and high case fatality rates, public concern about emerging infectious diseases was increased (Krause, 1994; Morse, 1995). Infectious diseases' emergence was concomitant with human behavioral changes, their interaction with wildlife, and environmental changes (Weiss & McMichael, 2004). Emergence was found to be intensified by increasing human travel and globalized trade (Morse, 1996). Novel infectious diseases continued to emerge via new pathways often from unexpected reservoirs (Cunningham et al., 2017). Travel and trade, and an altered attitude of humans towards domestic animals, wildlife, and nature affect

vectors and pathogens distribution worldwide. In the MENA region, livestock import is from as far as Australia, China, and Argentina, mixed transportation systems are often used (Miranda-de la Lama et al., 2014) which cause often considerable morbidity and mortality of livestock. The surviving imported livestock could carry non-native pathogens. Furthermore, increased tick densities in the region could permit non-native pathogens to maintain their populations and circulate within farming systems. The trigger of the problem seems to be associated with the increased human population size and farming activities (Heyman et al., 2010). Multiple causes of novel disease emergence have been documented. However, the human-mediated transport of vectors or pathogens across geographical regions (pathogen pollution) has been recognized as a major cause of diseases emergence in animals (Cunningham et al., 2017). Therefore, tick-borne diseases may have more chances to remain in livestock farming systems and will become an increasing problem (Heyman et al., 2010). Some scattered reports were found in the literature of *Hyalomma* species being imported into the USA, most commonly on animals and animal products. Five tick species of genus *Hyalomma* reported on ostriches that were imported from Africa and Europe (Mertins & Schlater, 1991). In addition, Keirans and Durden (Keirans, 2001) reported one case of *H. marginatum* found on a human with a travel history to Greece. Consequently, a detailed travel history may be essential for the identification of ticks and assessment of the risk of vector-borne diseases. *Hyalomma* is considered a medically-important tick genus because of transmiting a variety of pathogens including viral, bacterial, and parasitic (Bakheit et al., 2012). For example, previously *H. marginatum* transmitted the CCHF in Crimean Peninsulas, Russia and Turkey, *H. anatolicum* in Pakistan, Iran, Tajikistan and Turkmenistan, *H. rufipes* in Africa, and *H. asiaticum* from Central Asia to China (Bakheit et al., 2012; Goddard, 2012; Hoogstraal, Oliver, & Guirgis, 1970).

Further, *Hyalomma* species have also been implicated in tick paralysis in humans (Doğan, Devge, Pata, & Sönmezoğlu, 2012). In the MENA region, thousands of livestock are imported each year from different countries including India, Pakistan, Sudan, Somalia, Turkey, Argentina, Australia, Iran, and Uruguay in UAE and Saudi Arabia. Livestock farming and production contribute significantly to the food resources of the countries (Alanazi, Al-Mohammed, et al., 2018). Tick record from the UAE was limited before 1995 and only the ixodid ticks, *H. anatolicum, H. impeltatum, H. dromedarii, R. sanguineus,* and *R. turanicus,* were on the file at the U.S. National Tick Museum (Khan et al., 1997). During the CCHF outbreak in 1994-1995, ticks were collected and investigated. And data indicate that several competent vectors of the CCHF virus were found on imported animals (Khan et al., 1997). Further, it was found that CCHF virus-infected animals were imported mostly from Somalia and with fewer numbers arrived from Iran (Khan et al., 1997) . Preventing the importation of infected hosts may stop the import of many diseases of economic or public health significance. Presently, countries endorse this principle for the movement of people, by surveillance of infected persons arriving at their international borders, particularly during pandemics and the World Health Organization (WHO) provides guidance and training through its International Health Regulations (IHR). Further, rules and regulations for international trade including animals and their products are created and enforced by the WHO, for smooth flow of trade (Cunningham et al., 2017).

1.4.8 Climate Change and Ticks and Tick-Borne Diseases

The MENA region has been affected by climate change like the rest of the world. Climate change is considered a contributing factor in ticks and tick-borne pathogens' distribution and impact (positive or negative) on the biology of ticks (e.g.,

high mean temperatures and high humidity facilitate tick survival in certain areas, however in others, the opposite effect occurs). Along with climate change, other factors, for example, habitat fragmentation, demographic modification, and other environmental changes may complicate the processes through facilitating the survival and establishment of tick colonies in the regions where these tick species were not prevalent before (Estrada-Pena, 2009). Understanding about disease-causing pathogens and diseases epidemiology has been improved significantly during the past decade. Further research is required to explain the lifecycle of these pathogens' antigenic differences and human immune response for effective vaccine development and treatments (Heyman et al., 2010). For the development of novel approaches for better target control interventions, the prediction of the disease risk requires an understanding of both pathogen and disease dynamics (Kilpatrick et al., 2018). In the meantime, for prevention of the disease, the best way is to educate the risk groups (especially farmworkers and laborers at slaughterhouses/abattoirs) and awareness in healthcare personnel. The distribution and abundance of tick populations also depend on the interaction of large-scale climate influences, microclimates, habitat characteristics, and host densities (Kilpatrick et al., 2018). Microclimate conditions may impact tick survival directly by increasing tick mortality rate, and indirectly by influencing the activity (Dobson, Finnie, & Randolph, 2011; Ogden, Bigras-poulin, Callaghan, Barker, & Lindsay, 2005). Temperature, both cold and hot, significantly reduces the host-seeking activity and tick survival. Further, low humidity reduces the tick activity and probably kills ticks through desiccation; high levels of rainfall also inhibit activity (Eisen, Eisen, & Ogden, 2017). In adverse climate conditions, ticks often seek microclimate refuges in leaf litter or debris to reduce the impacts of extreme temperature and humidity on survival (Lindsay et al., 1999). Therefore, the risk of

human infection is thought to be increased with the density of questing infected nymphs in the environment and varies at a local scale (Kilpatrick et al., 2018).

1.5 Potential Contribution of the Study

Ticks are important arthropod vectors and serve as reservoirs, and harbor a wide variety of pathogens. Tick identification is a very important step in any successful tick-borne disease control program. The data on tick morphological and molecular identification in the UAE is scarce. This study on ticks' species provided the first morphological and molecular record of three tick species from livestock in the UAE. Further, this research provided the first one-year study on camel tick, *H. dromedarii* to better understand the pattern of tick population seasonal dynamics. Tick population size was determined and was linked the climatic factors. By combining some of these elements of ecology, the pattern of population fluctuation over a year was evaluated with its link to a variety of factors including host-parasite interaction, microclimatic conditions, and host range. This study has shown how camel tick managed to survive in the UAE under extreme conditions. Moreover, strategic tick control measures could be implemented from March to June, which is the time of the tick population peak. Tick microbiota plays a significant role in vector competence, pathogen transmission, and tick reproductive success. Tick-borne pathogens can significantly decrease the production of milk, meat, and hide of livestock. In addition, this work represents the first study on tick microbes in the UAE. A diversity of microbes in *H. dromedarii* ticks was assessed through Next-Generation Sequencing, which highlights the reservoir potential of this tick species for significant pathogens and understand the patterns of tick-borne bacteria that circulate in camels. Ticks play a significant role in the transmission cycles of various zoonotic diseases caused by viruses, bacteria, and

protozoans. Mixed tick-borne pathogen infections are common in nature. Epidemiological investigations suggest that infections caused by mixed tick-borne microbiota can modulate their pathogenicity and disease burden in various livestock hosts. Early detection of pathogens is crucial in curtailing their spread and subsequently in reducing the risk of exposure and possible outbreaks. Four microorganisms in *Hyalomma* ticks including endosymbionts and pathogens were detected. *Theileria ovis* is reported for the first time in the UAE which shows that continuous surveillance of pathogens is crucial at the domestic animal-human interface to maintain good health of livestock, workers and for the early detection of disease catastrophes in ruminants. In addition, published systematic review on ticks and tick-borne diseases in the MENA region, which is part of this study, provides new insight into the tick world in the desert ecosystem and provide management strategies to deal with this serious threat to humans and animal health. Mitigation strategies are well established in the UAE; however, continuous tick prevalence is a conundrum that needs to be solved and this research outputs can contribute a lot to tick management strategies. Therefore, this research results are a good combination of ecology and molecular biology to tackle the research questions in the field of ticks and tick-borne diseases.

1.6 Limitations of the Study

The limitations of my study are the following:

1) With the availability of funds and labor, more sampling would reveal more details about tick populations.

2) The population dynamic study was conducted for one year and at only one location. More years and more sites would reveal more data about tick prevalence and their distribution.
3) Though 16S rRNA gene-based analysis is good to provide bacterial composition and diversity, it does not provide identification of bacteria at the species level. Future research should use a technique that identifies bacteria at the species level.

Chapter 2: Methods

2.1 Research Design

This research was designed to answer questions regarding the biology and ecology of ticks, their abundance, population fluctuation pattern, the composition of microbes to understand tick-borne diseases epidemiology.

2.2 Ethical Approval

This study was carried out in accordance with the recommendations of the Animal Research Ethics Committee (A-REC) of the UAE University (ethical approval# ERA_2019_5953). In addition, the experimental protocol was approved by the UAE University Research Office.

2.3 Prevalence, Distribution, and Molecular Record of Hard Ticks from Livestock

2.3.1 Study Area for Tick Collection

This study was conducted mainly in two types of areas. The first type included open farms in the desert ecosystem of the UAE (Figure 5), where livestock holding areas are accessed by large and small mammals, reptiles, and birds, and most of these areas also lie at the border of Oman, and livestock markets (Figure 6). In addition, sampling was done at areas in which animals were reared on farms and housed in a homestead, locally called Izba. The open areas have a typical desert ecosystem climate, which is characterized by high amplitudes of seasonal temperatures. The area is the home to a camel farming community for whom keeping livestock is their cultural tradition and way of life. The second type included livestock markets in Dubai and Al Ain. (Figure 6). The details about the animal hostssampled, the distribution of samples

among the sites, tick species recorded at each site, and the years when each site was sampled are given in Appendix 1.

Figure 5: Tick collection site (open farm in UAE desert ecosystem).

Figure 6: Tick collection site (livestock market).

2.3.2 Tick Sampling from Livestock

The current work is a cross-sectional study. Animals were sampled from three emirates in the UAE (Abu Dhabi, Dubai, and Sharjah) (Figure 7). At desert farms, tick collection was undertaken early mornings and in the evenings from animals. A total of 587 domestic animals were examined, including 300 camels, 119 cows, 97 sheep, and 71 goats. Ticks were collected from the whole body of animals and a total of 5950 ticks were collected from camels (4803 ticks), cows (651 ticks), goats (219 ticks), and sheep (277 ticks) (Figure 8 and 9). All adult ticks from the animals were removed manually using forceps and placed in 50 mL plastic vials. The vials containing ticks were retained inside an icebox and were taken to the Animal Ecology and Entomology Laboratory at the UAE University, where they were frozen at -80°C until further processing. All vials were labeled and ticks were counted.

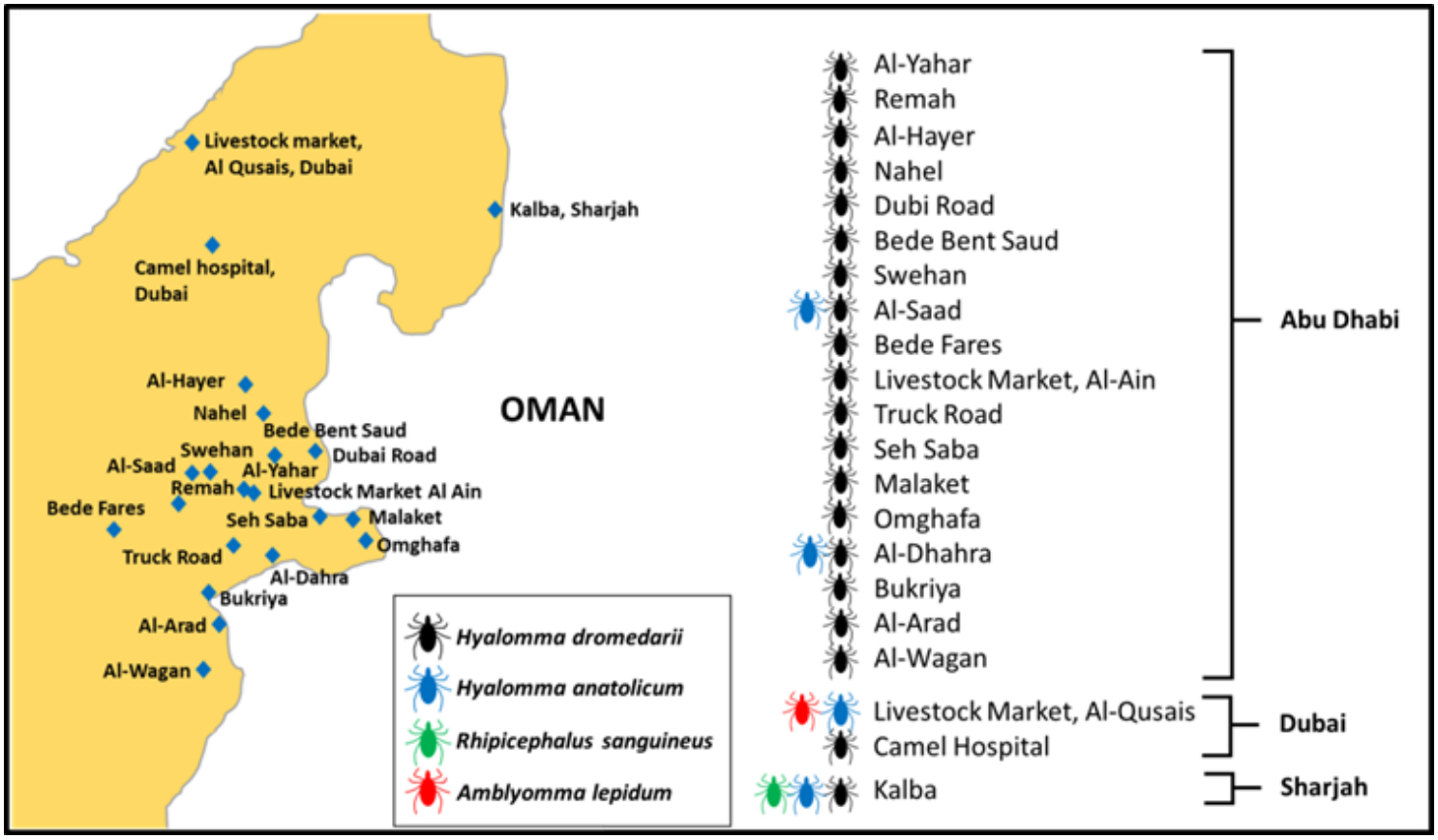

Figure 7: Tick collection sites and distribution of tick species on livestock in study area, in 2019-2021 in UAE.

Figure 8: Tick sampling from livestock.

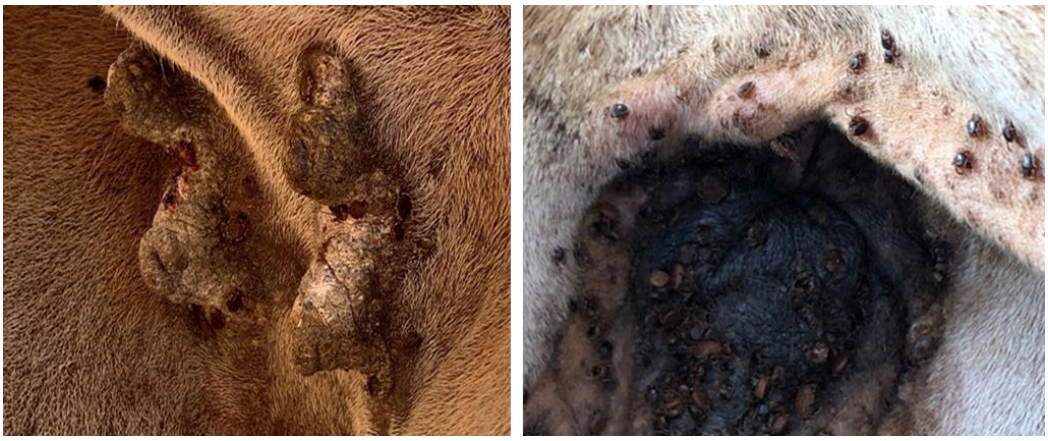

Figure 9: Tick infestation on camel, and cow. camel (left side), and cow (right side)

Labeling for all samples included location, host, date of sampling, number of ticks, and the gender of the host (male or female).

2.3.3 Morphological Identification of Ticks

In the laboratory, the sampled ticks were counted, rinsed with 70% ethanol, and then with deionized water for five minutes to remove environmental particulate contamination (Carpi et al., 2011), and then air-dried (Figure 10). Ticks were examined using a dissecting Nikon SMZ1500 Stereoscopic zoom microscope (Nikon, Tokyo, Japan). Further, ticks were morphologically identified to the species level using taxonomic keys (Apanaskevich, 2003; Apanaskevich, Schuster, & Horak, 2008; McCarthy, 1967; Robinson, 1962; Walker et al., 2003) based on morphological characteristics and sorted according to sex and stage of development.

Figure 10: Ticks counting, washing and morphological identification.

2.3.4 Molecular Characterization of Ticks

2.3.4.1 DNA Extraction

After washing the ticks and finishing the morphological identification, DNA was extracted from 44 individual ticks to confirm their identification and provide a molecular record in the GenBank. Briefly, legs were removed from ticks with a sterile

scalpel blade and homogenized in a 1.5 mL tube by using liquid nitrogen. DNA extraction was done using a DNA Mini Kit (Qiagen, Hilden, Germany) following the manufacturer's protocol. The DNA concentration of each sample was estimated using a NanoDrop 2000 UV spectrophotometer (Thermo Scientific, Waltham, USA) (Figure 11). Extracted DNA samples were stored in a freezer at -20°C.

Figure 11: DNA extraction and quality check.

2.3.4.2 Polymerase Chain Reaction Amplification

Extracted tick DNAs were subjected to a polymerase chain reaction, which amplified regions of the *cox1* and the *16S* rRNA genes. Gene fragments were separately amplified from each individual DNA sample representing each tick specimen using specific oligonucleotide primer pairs (Table 3) to amplify 710 bp of the *cox1* gene, and 460 bp of the *16S* rRNA gene-based on published protocols (Black & Piesman, 1994; Folmer, Black, Hoeh, Lutz, & Vrijenhoek, 1994). The PCR amplifications were performed in a Swift MaxPro thermo-cycler (ESCO, Singapore, Singapore) and each PCR was done in 25 µL reaction volume containing 12.5 µL Taq PCR master mix (Qiagen, Hilden, Germany), 1.0 µL (10 pM) of each primer, 3.0 µL

of tick genomic DNA, and 7.5 µL nuclease-free water. Thermo-cycle conditions are given in Table 3 and in each PCR negative control (no template DNA) was used to detect any contamination. In addition, positive control was used to indicate that the primers were properly annealing to the target region on the template DNA.

2.3.4.3 Agarose Gel Analysis, Amplicon Purification and Sequencing

Aliquots (6 µL) of individual amplicons were visualized in 1.5% agarose gel (Promega, Madison, USA) stained with ethidium bromide (Promega, Madison, USA). A100-bp DNA ladder (Promega, Madison, USA) was used as a standard marker. Gel photographs were taken using a gel documenting system (Major Science, Taipei, Taiwan). Further, amplicons were purified using QIAquick PCR Purification Kit (Qiagen, Hilden, Germany) according to the manufacturer's protocol and DNA concentration was measured using a spectrophotometer, NanoDrop 2000 UV spectrophotometer (Thermo Scientific, Waltham, USA). DNA fragments were sequenced by Sanger Sequencing at the Sequencing Unit, Biology Department, UAE University.

Table 3: Primers and cycle conditions used to amplify gene fragments for molecular identification of ticks.

Target gene	**Primer**	**Sequence (5'–3')**	**Cycle conditions**	**Amplicon size (bp)**	**Reference**
16S rRNA	16S+1 16S−1	CTGCTCAATGATTTTTTAAATTGCTGTGG CCGGTCTGAACTCAGATCAAGT	94°C 5 min 32 cycles: 94°C 1 min 52.9°C 1 min 72°C 1 min 72°C 15 min	460	(Black & Piesman, 1994)
cox1	LCO1490 HCO2198	GGTCAACAAATCATAAAGATATTGG TAAACTTCAGGGTGACCAAAAAATCA	95°C 5 min 30 cycles: 94°C 1 min 54°C 1 min 72°C 1 min 30 S 72°C 10 min	710	(Folmer et al., 1994)

2.3.4.4 DNA Sequence Analysis

Sequences of the *cox1* and *16S* rRNA genes from the present study were compared with the available data in the GenBank using the Basic Local Alignment Search Tool (BLAST), the National Center for Biotechnology Information (NCBI). Based on DNA similarity with sequences of the current study using BLAST, representative *16S* rRNA gene and *cox1* gene sequences of *H. dromedarii*, *H. anatolicum*, *R. sanguineus*, and *A. lepidum*, were downloaded from the GenBank. The sequences acquired in this study were deposited in the GenBank database to get accession numbers.

2.3.4.5 Statistical Analysis

The number of ticks was recorded on camels, cows, sheep, and goats. The prevalence (proportion of hosts infested with ticks), mean intensity (number of ticks per infested host), and mean abundance (number of ticks per host) were calculated for all hosts (Rózsa, Reiczigel, & Majoros, 2000). Mean intensities and mean abundance values were compared between hosts using bootstrap *t*-tests, and the *p*-values were generated using 2000 replications. The prevalence of ticks was compared between hosts, and within the same host on the basis of sex using Fisher's exact test and 95% confidence levels using the Clopper–Pearson method. All comparisons were made using the Quantitative Parasitology Software Version 3.0 (Rózsa et al., 2000).

2.4 Seasonal Population Dynamics of *Hyalomma dromedarii* on Camels

2.4.1 Study Site

The study was carried out on a private farm near Umm Al Zammol Road, Al Dhaharah, Al Ain (Figure 12), located about 120 km south-southeast of Abu Dhabi (N 24° 11' E 55° 45'). Al Ain covers an area of approximately 13,100 km^2. The inland location of Al Ain makes its environment warmer and drier compared to Abu Dhabi and Dubai. The vegetation cover comprises sparse halophytes. This region is characterized by high amplitudes of seasonal temperatures with mean monthly temperature varies in the study area between 17.1°C in winter and 38.1°C in summer. Annual rainfall averages 91.1–201.0 mm (Suleiman, 2007). The owner of the farm treated the camels with the acaricide Phoxim (Ectofox®50 EC; Dammam, Saudi Arabia) monthly (during the whole year), which is a common practice among all camel breeders in Al-Ain region. The acaricide was sprayed on the animals at a known concentration (2 mL/1L of water) and this may influence the actual population dynamics of the ticks. However, one of the goals of this study is to assess *H. dromedarii* population fluctuation over time under common camel breeding and management practices in the study area. Therefore, the population dynamics was monitored despite of acaricide use.

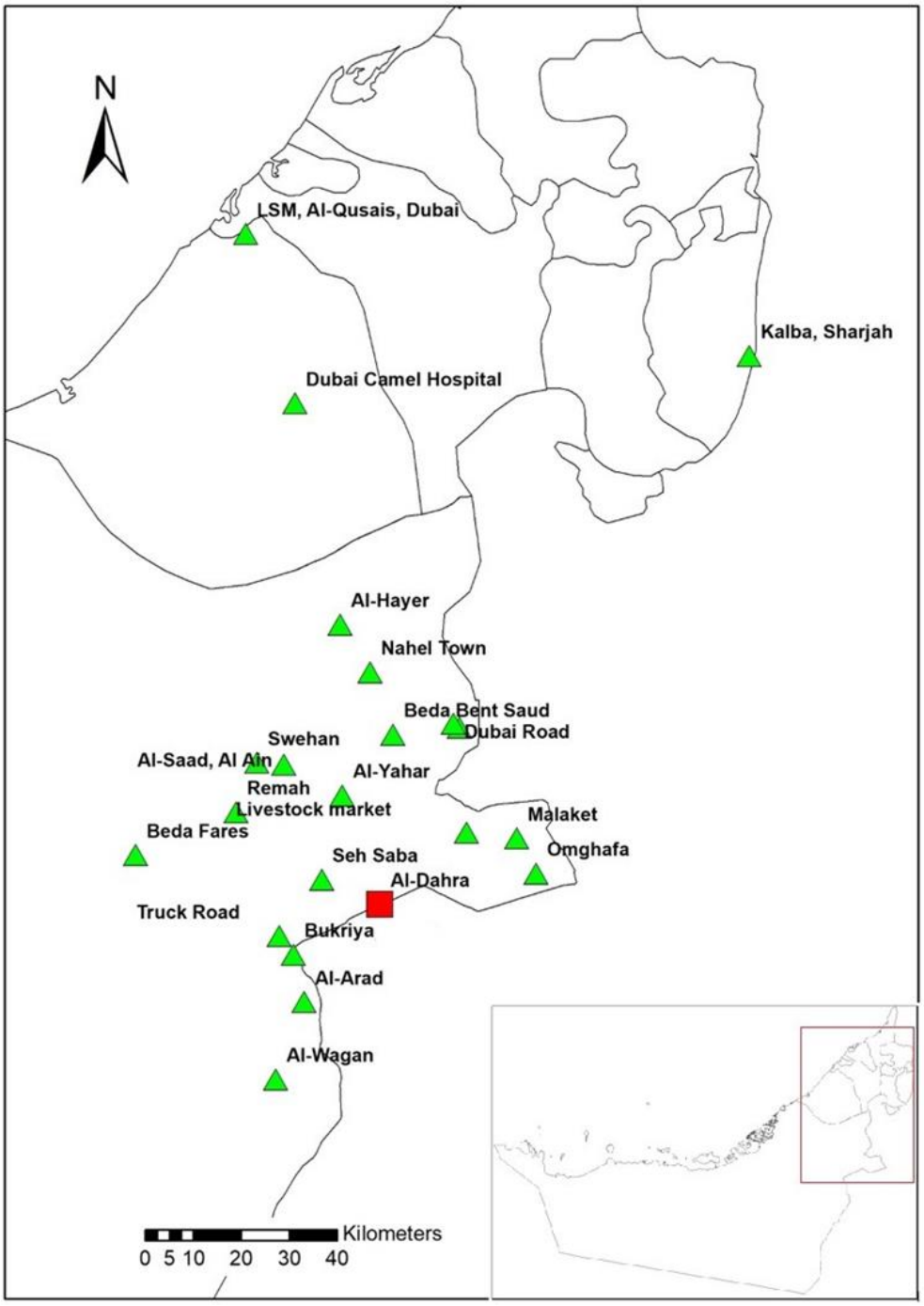

Figure 12: Study area for population dynamics. Red square shows the farm location.

2.4.2 Selection of Animals for Survey

The current study was conducted on a farm, which had 30 local breed camels (25 females, 1 male and 4 calves). Camels were separated from sheep and goats on the

farm by a fence. A total of 25 adult camels (24 females, and 1 male) aged 3–14 years were chosen and included in the survey. They were examined monthly from March 2019 to February 2020. The origin, breed, age, sex, and reproduction status of animals were recorded at the beginning of the study (Anullo, Alemu, & Ayele, 2018). Most individuals were reproductively active, with the exception of four calves.

2.4.3 Tick Sampling, Counting, Labeling and Identification

On each monthly tick collection, all visible ticks were removed manually using forceps from the entire right side of the body of each animal. The right side was arbitrarily selected for this purpose. The number of ticks collected was doubled to determine the approximate tick load per camel (Zeleke & Bekele, 2004). Ticks were retained in 50 mL plastic vials. The vials containing ticks were placed on ice inside a cool box and were taken to the Entomology Laboratory at UAE University, where they were frozen at –80°C until further processing. All ticks were counted. Labeling for all specimens included location, host, and date of sampling. Ticks were morphologically identified under a stereomicroscope using available taxonomic keys (Walker et al., 2003) and classified according to species, sex, and stage of engorgement.

2.4.4 Parasitological Indicators

The following parasitological indicators (Gharbi, Moussi, et al., 2013) were determined:

Infestation prevalence (%) = 100 × number of infested animals/total number of animals

Infestation intensity = number of ticks/number of infested animals

Tick load = number of ticks/total number of animals

2.4.5 Meteorological Data Collection

The effects of temperature and humidity were mainly studied because this study was conducted in a desert ecosystem wherein these are the major factors affecting every living thing. Meteorological data was used from the nearest meteorological station. The mean monthly relative humidity (RH) was in percent and the mean monthly temperature was in degrees Celsius (°C).

2.4.6 Statistical Analysis

The relationship between the monthly tick average burdens and the monthly average temperature was assessed through the Pearson correlation test using GraphPad Prism 8.3.1 for Windows (San Diego, CA, USA, www.graphpad.com). Tick loads (number of ticks per host) were compared between different months by One-way ANOVA. In addition, descriptive statistics of tick counts and percentages for male and female ticks and life stages were calculated.

2.5 Bacterial Communities Composition and Diversity in Camel Tick, *Hyalomma dromedarii* using Next-Generation Sequencing

2.5.1 Tick Collection

In a cross-sectional study, ticks were collected manually from camels in 2010 and 2019. In 2010, a project was completed (in Animal Ecology and Entomology Laboratory) in which a large number of *H. dromedarii* ticks were collected and stored in − 80°C. In 2019, a new project was started on this tick species and it was interesting to collect ticks from the same locations sampled in 2010 so that a comparison of microbial communities could be made between the samples collected in both projects and detect changes over time. Farms and camels were selected randomly. In 2010,

ticks were collected from 10 locations (Al-Wagan, Al-Yahar, Bede' Fares, Bede'Bent Suod, Dubai Road, Dwar Al-Shahenat, Malaket, Omghafa, Remah, and Swehan) in Al-Ain area at the eastern part of the UAE. In each location, five camels were selected, and from each camel, 10 ticks were collected. In the laboratory, one partially engorged female tick was picked out of the 10 ticks collected per animal to be subjected to DNA extraction and sequencing. The same strategy of tick sampling was followed in 2019. As a result, 1000 ticks were gathered in total in 2010 and 2019 and from them 100 partially engorged female ticks were used at the rate of 50 ticks each year. Ticks were kept in plastic vials (50 ml) in − 80°C freezer until DNA extraction. The experiment protocol is shown in (Figure 13).

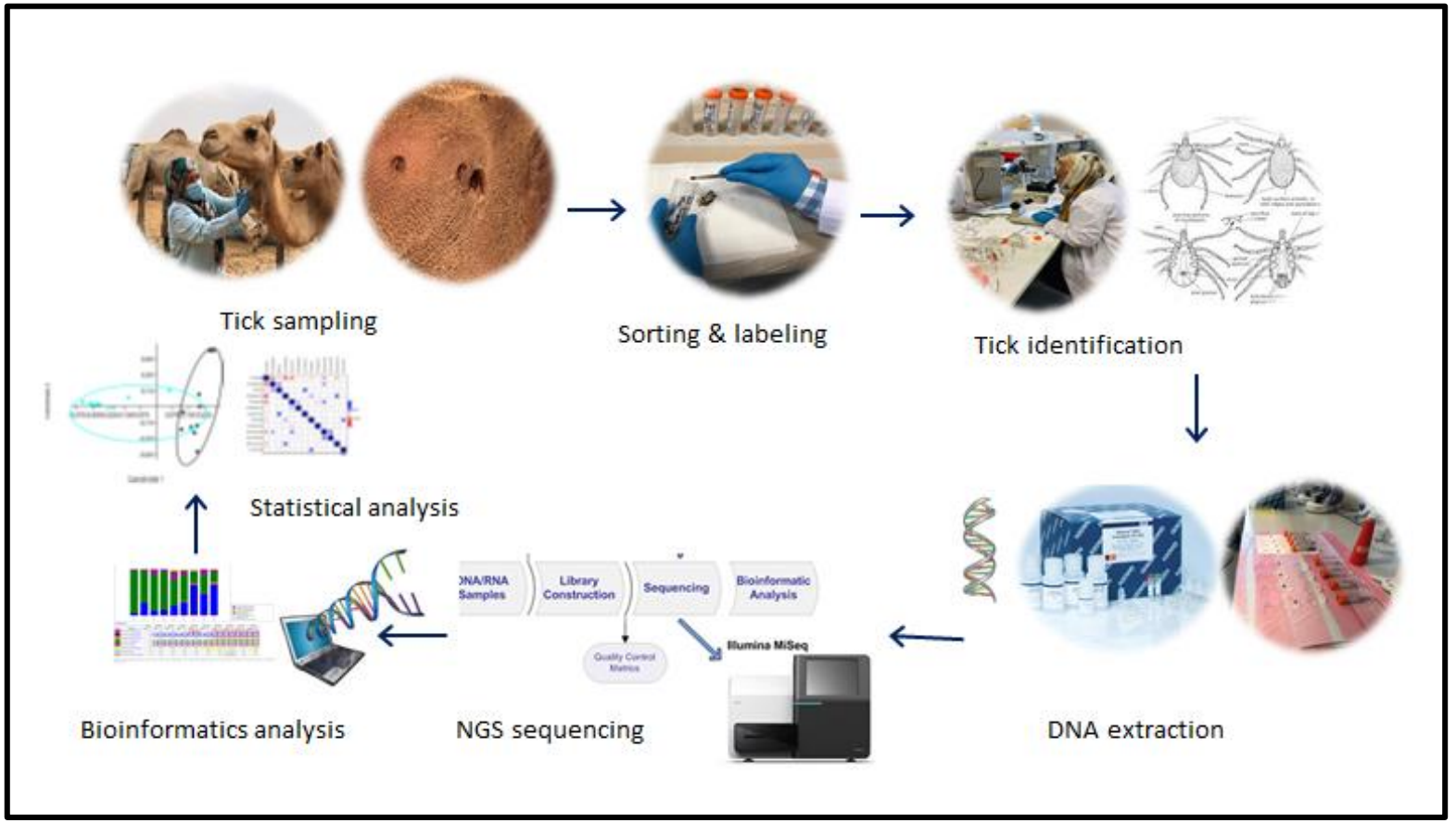

Figure 13: Experimental protocol for assessment of bacterial communities'composition of camel tick.

2.5.2 Tick Identification, Genomic DNA Extraction and Pooling

The identification of ticks as *H. dromedarii* was done morphologically using the keys of Apanaskevich et al. (2008) and Walker et al. (2003) and based on DNA sequencing using Cytochrome Oxidase subunit I (COI) gene (Al-Deeb et al., 2015). Briefly, in the males the sub-anal plates are aligned outside the adanal plates. In addition, the adanal plates have a characteristic shape with both long margins strongly curved in parallel. In the females, the genital aperture has posterior lips with a narrow V, which is also found in *Hyalomma impeltatum*, but the posterior margin of their scutum is distinctly sinuous compared to a slightly sinuous margin in *H. dromedarii*. With molecular identification, a segment of the COI gene was amplified in polymerase chain reaction using a primer pair Fish1F: 5′ -TCA ACC AAC CAC AAA GAC ATT GGC AC-3′ and Fish1R: 5′ -TAG ACT TCT GGG TGG CCA AAG AAT CA-3′ 69 under the following thermocycling conditions: 2 min at 95°C followed by 30 cycles of 1 min at 94°C, 1 min at 54°C, and extension for 90 s at 72°C. It is already mentioned that in each sampling year, 50 partially engorged female ticks were collected from which DNA was extracted individually. Before DNA extraction, each tick was thoroughly washed with distilled water. Each whole tick was crushed manually using a sterile Kimble Kontes pellet pestle (Thermo Fisher, Waltham, MA) inside a sterile 1.5 ml microcentrifuge tube. Genomic DNA was extracted from each individual tick using QIAamp Tissue Kit (Qiagen, Hilden, Germany) following the manufacturer's protocol. Quality and concentration of the extracted DNA was determined with a spectrophotometer (Nano Drop ND-1000, Erlangen, Germany). In addition, DNA quality was assessed on a 1% agarose gel stained with ethidium bromide and visualized under UV light. DNA was stored in a − 20°C freezer until used. Prior to sequencing,

extracted DNA samples from individual ticks were pooled according to collection location. This resulted in having 10 DNA pools for each sampling year.

2.5.3 16S rRNA Gene Amplicon Sequencing and Bioinformatics Analysis

To evaluate the microbial communities in camel ticks, a 16S rRNA gene-based analysis was conducted. A total of 20 DNA pooled samples were shipped to Macrogen Inc (Seoul, South Korea). The following primers were used for amplifying the V3 V4 region: Bakt_341F: CCT ACG GGNGGC WGC AG Bakt_805R: GAC TAC HVGGG TAT CTA ATC C70 using the Herculase II Fusion DNA polymerase Nextera XT Index Kit V2. Sequencing was performed on a Illumina MiSeq platform with a read length of 301 bp. Demultiplexed paired-end sequence reads in FASTQ format for each sample were merged using fast length adjustment of short reads (FLASH) version 1.2.1171. Next, CD-HIT-OTU72 was used to cluster the reads from 2010 and 2019 into OTUs using default options. CD-HIT-OTU filters out low-quality reads, trims extra-long tails, identifies chimeric reads and clusters reads into OTUs with a cutoff of 97% identity. Finally, taxonomic assignment of OTUs was performed using the *assign_taxony.py* script from QIIME 1.9.173 by performing a Basic Local Alignment Search Tool (BLAST)74 search against the National Center for Biotechnology Information (NCBI) 16S microbial database. Taxonomic levels of bacteria from phylum to genus were profiled in samples across all locations. Taxonomic abundance ratios were calculated from taxonomic abundance count to summarize and interpret the results at phylum, class, family and genus levels. Sequences were deposited in NCBI Sequence Read Archive under the BioProject ID PRJNA639925.

2.5.4 Quantification and Statistical Analyses

Principal Coordinates Analysis (PCoA) was conducted to determine patterns of diversity in bacterial communities. The PCoA was conducted and visualized using the software PAST 5.27 Paleontological statistics software package (Hammer, Harper, & Ryan, 2001) (Øyvind Hammer, Natural History Museum, University of Oslo, Norway, ohammer@nhm.uio.no). OTU count of each genus was entered and the samples were categorized by year (2010 and 2019). Eigenvalues were examined to determine the extent of variation explained by the first three principal coordinates (Coordinates 1–3) (Paliy & Shankar, 2017). Different indices of diversity were calculated, since a single index often does not reflect the true nature of diversity and a combination provides an approximation of diversity. Richness (total number of genera, based on OTUs obtained for each genus); Shannon Wiener Index; and the Index of Dominance were estimated. The Shannon Wiener Index of diversity was calculated using the following formula:

$$\text{Shannon-Wiener Index } H = -\sum_{i}^{S} p_i \log p_i$$

where S—the total number of genera, *i*—the number of OTUs for genus i; and *pi*—relative proportion of genus *i*. Index of Evenness (relative abundance of each genus, based on OTUs) was calculated as follows:

$$\text{Index of Evenness, } E = e^{H}/S$$

where *H*—Shannon–Weiner's Index and S is the total number genera.

The Index of Dominance (D) was calculated using the following formula:

D = number of OTUs for the dominant genera/the total number of OTUs.

All these indices were compared between years using paired two sample *t*-test using PAST (Hammer et al., 2001). Pearson's Correlation Coefficient (*r*) was calculated to determine associations between different genera that occurred in 2010 and 2019 (Sokal & Rohlf, 2012). Genera with significant correlations were subjected to stepwise regression analysis, with backward selection (Sokal & Rohlf, 2012). One genus was used as the response variable and all other genera that had significant correlations with the response variable were used as explanatory variables. Genera were removed individually based on significance to see the effect on the overall model. Only those genera that improved the overall model were retained, while genera that did not affect the model were removed. The process was repeated with each genus that had a significant correlation with other genera. For all tests, the value of α was set at 0.05.

2.6 Tick-Borne Microorganisms and their Prevalence in *Hyalomma* Ticks Collected from Livestock

2.6.1 Study Area, Tick Collection and Identification

This is a cross-sectional study in which tick collection was done from January 2019 to February 2020. A total 562 tick samples were collected from camels, cows, sheep and goats in the Emirates of Abu Dhabi, Dubai, and Sharjah from 24 locations (Appendix 2; Figure 14). The largest number of ticks was collected from camels (516 samples), which represented the main animal in this study, whereas fewer samples were collected from sheep, goats, and cows (46 samples). Animals were selected randomly, and from each host 10 ticks were removed manually using a pair of forceps. Ticks were collected in 50 mL plastic tubes (Sterilin, UK). All tick samples were placed in an icebox and transported to the Entomology Laboratory at the UAE

University. Ticks were frozen at –20°C until further processing. In Sharjah, a pool of ticks was created for each host (camels (3), sheep (25), goats (25), and cows (6)) (Appendix 2). Similarly, a pool of ticks was created from the sheep (36) samples in Abu Dhabi. Further, a total of 15 cows (Australian origin) and 30 goats (Pakistani and Indian origin) were sampled in Abu Dhabi for tick collection; however, these animals were not infested with ticks (Appendix 2). All ticks were identified at the species level on the basis of their morphology by using taxonomic keys (Apanaskevich et al., 2008; Walker et al., 2003). The experiment protocol is shown in Figure 15. The figure was created with BioRender.

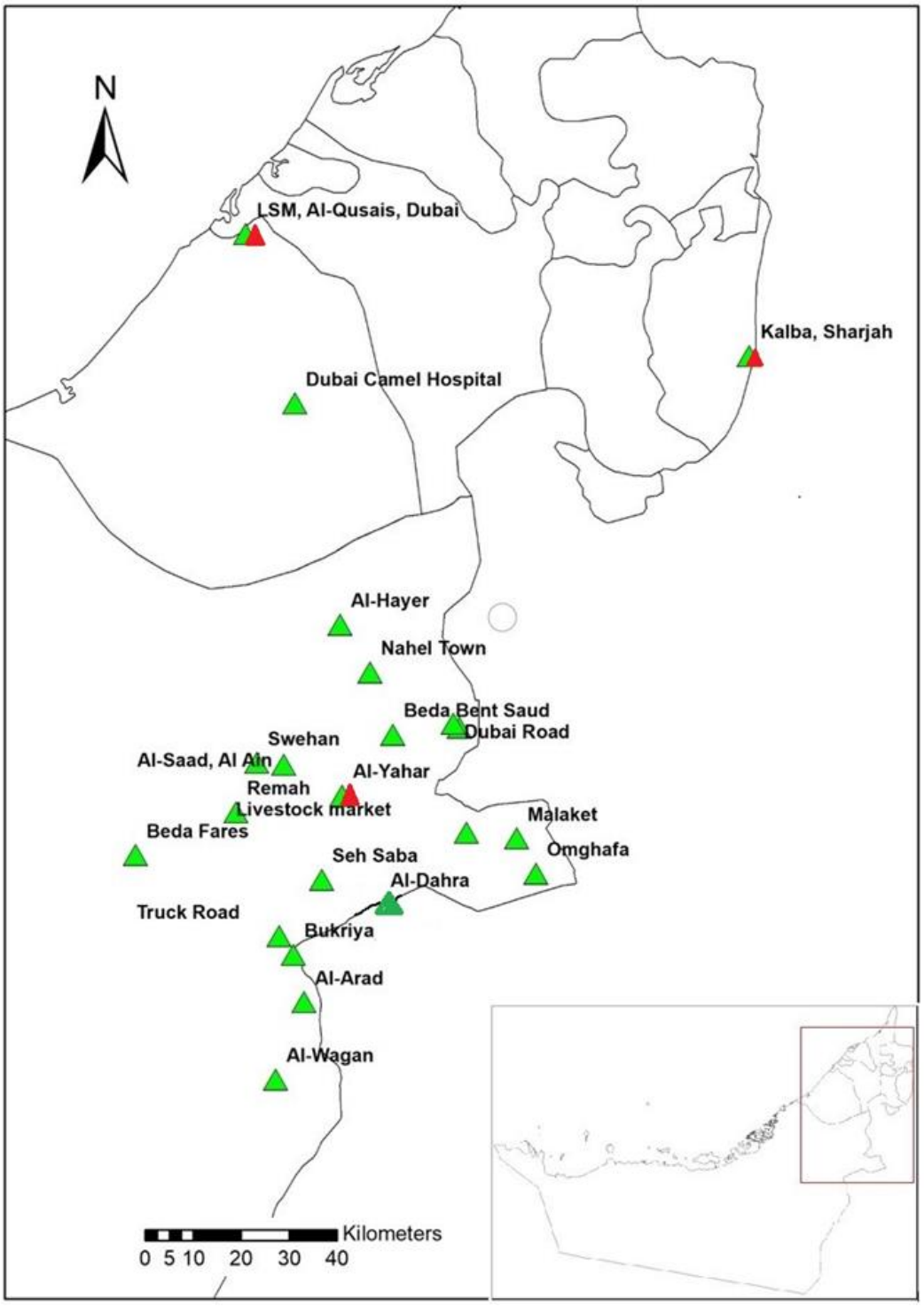

Figure 14: The study area from which tick samples were collected in the UAE. Red triangles show the locations of sheep, cows and goats samples, while green triangles show the locations of camel samples except Livestock Market Al-Qusais, Dubai.

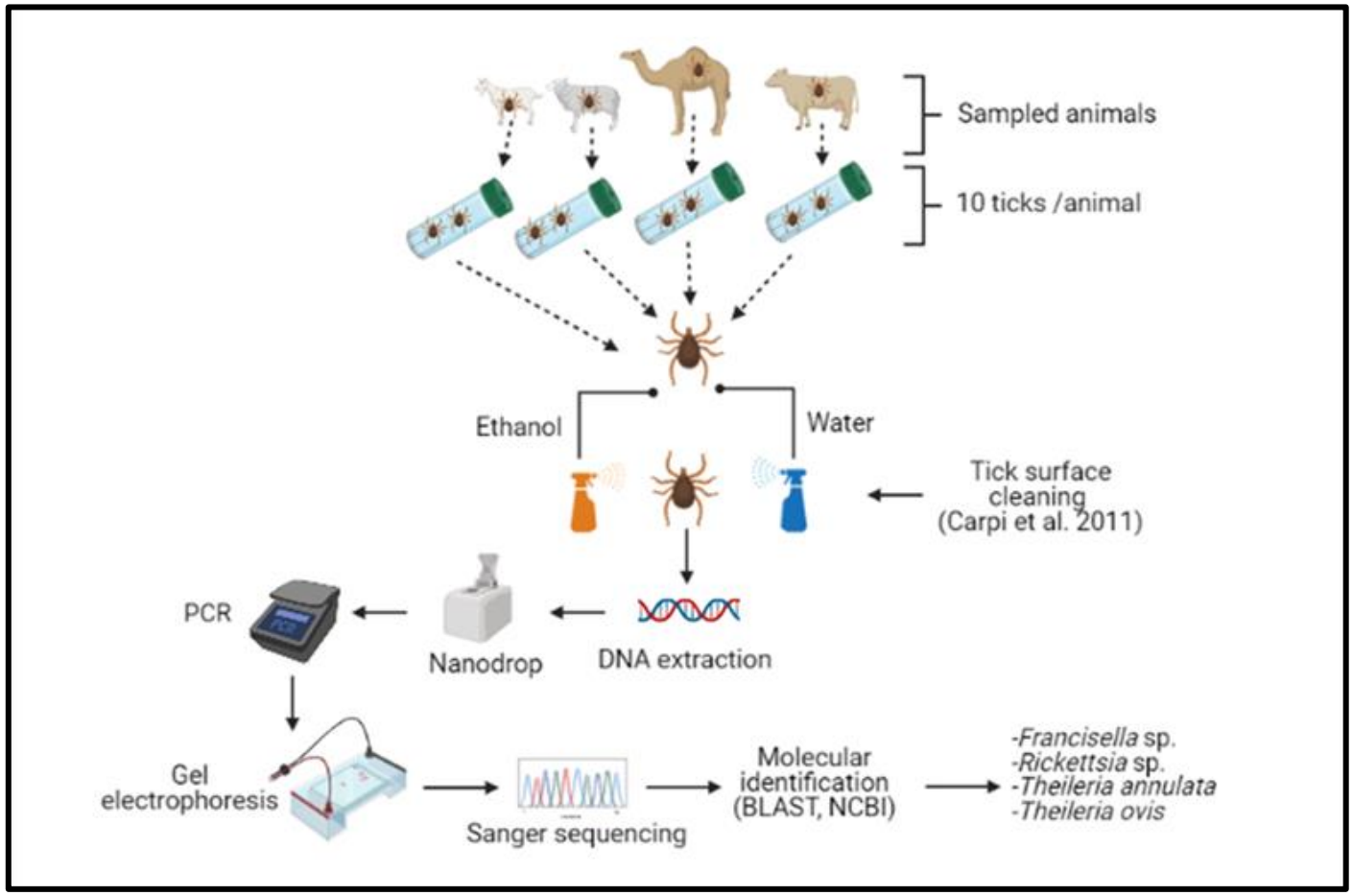

Figure 15: Experimental protocol. *Hyalomma* ticks were collected from animals (camel, cow, goat, and sheep) from Abu Dhabi, Dubai, and Sharjah in the UAE.

2.6.2 Genomic DNA Extraction

As for *H. dromedarii* ticks, DNA was extracted from individual ticks (partially engorged female). However, in the case of *H. anatolicum* ticks, DNA was extracted from a pool of 5 ticks (male) due to their small size. Further, partially engorged females of *H. anatolicum* were not available in all samples. Before the extraction, each tick was washed in 500 µL 70% ethanol followed by 500 µL sterile double-distilled H_2O for five minutes to remove environmental contaminants attached to the tick body (Carpi et al., 2011) and then dried for 10 minutes. Ticks were manually crushed using a plastic pellet pestle inside a sterile 1.5 mL micro-centrifuge tube by using liquid nitrogen. The DNeasy blood and tissue kit (Qiagen, Hilden, Germany) was used for tick genomic DNA extraction, following the protocol of the manufacturer. Extracted DNA samples were stored in a freezer at -80°C.

2.6.3 Polymerase Chain Reaction

PCR tests were performed for *Francisella* sp. detection, with tick genomic DNA using an oligonucleotide primer pair (Table 4) (Duzlu et al., 2016), to amplify 1151 bp of the 16S rRNA gene. Detection of *Rickettsia* sp. was carried out by nested PCR of the *ompA* gene (Blair et al., 2004). The amplification of a 590 bp fragment was obtained in the first PCR and the amplification of a 540 bp fragment was obtained in the second PCR using oligonucleotide primers (Table 4). The detection of *T. annulata* and *T. ovis* was done by PCR tests with tick genomic DNA using the oligonucleotide primer pair amplifying 560 bp of the ssrRNA gene (Beck et al., 2009) (Table 4). For all the above-mentioned microorganisms, each PCR reaction was carried out in 25 µl volume containing 12.5 µL Taq PCR master mix (Qiagen, Hilden, Germany), 1.0 µL

(10 pM) of each primer, 3.0 µL of genomic DNA, and 7.5 µL nuclease-free water. PCR amplifications were carried out in a Swift MaxPro thermo-cycler (ESCO, Singapore) according to cycle conditions given in Table 4. Every PCR included a negative control (no template DNA) to detect any contamination. In addition, a positive control was used to indicate that the primers were properly annealing to the target region on the template DNA. In every PCR, we used filter tips and separate 0.2 mL tubes, rather than a PCR 96-well plate, to avoid aerosol cross-contamination between samples. In addition, PCR reaction tubes of the positive controls were prepared in a separate laboratory, to avoid any chance of contamination.

2.6.4 Agarose Gel Electrophoresis and Amplicon Purification

Products of PCR reactions were visualized using gel electrophoresis on 1.5% agarose gel, stained by ethidium bromide. The bands on the gel were visualized and photographed using a gel documentation system (Major Science, Taiwan). Amplicons of the positive samples which produced the expected band size were purified using a PCR purification kit (Qiagen, Hilden, Germany) following the manufacturer's protocol and saved for DNA sequencing.

Table 4: Primers and cycle conditions used to amplify gene fragments to identify microorganisms.

Pathogen	Target Gene	Primer	Sequence (5′–3′)	Cycle Conditions	Amplicon Size (bp)	Reference
Francisella sp.	16S rRNA	Fr153F0.1 Fr1281R0.1	GCCCATTTGAGGGGGATACC GGACTAAGAGTACCTTTTTGAGT	95°C 4 min 40 cycles: 94°C 30 s 60°C 45 s 72°C 60 s 72°C 20 min	1151	(Duzlu et al., 2016)
Rickettsia sp.	*ompA*	RR 190-70 (1st PCR) RR 190-701(1st PCR) 190-FN1 (nested) 190-RN1 (nested)	ATGGCGAATATTTCTCCAAAA GTTCCGTTAATGGCAGCATCT AAGCAATACAACAAGGTC TGACAGTTATTATACCTC	94°C 1 min 35 cycles: 94°C 30 s 50°C 1 min 68°C 4 min 72°C 20 min	590 540	(Blair et al., 2004)
Theileria sp.	ssrRNA	Pirop-F Pirop-F	GTCTTGTAATTGGAATGATGG CCAAAGACTTTGATTTCTCTC	94°C 2 min 35 cycles: 94°C 30 s 50°C 30 s 72°C 60 s 72°C 7 min	560	(Beck et al., 2009)

2.6.5 DNA Sequencing, Phylogenetic Analysis, and Microorganism Identification

Purified PCR products were sequenced (Sanger sequencing) at the Biology Department sequencing unit, UAE University. Microorganisms were identified based on sequence analysis using the NCBI BLAST analysis tool in the GenBank database. Sequences were submitted in GenBank and received accession numbers (MW537791, MW559557, MW560059, and MW701398). The DNA sequences of this study were compared with known sequences listed in the GenBank nucleotide sequence databases. The obtained sequences were aligned using the MUSCLE program and the phylogenetic trees were constructed through the Maximum Likelihood approach using Kimura 2-parameter method and bootstrap analyses with 1000 replicates in MEGA X 10.0.5 software (Kumar, Stecher, Li, Knyaz, & Tamura, 2018). In each phylogenetic analysis, we chose the most suitable substitution model based on the lowest Bayesian Information Criterion (BIC) scores. Consequently, after the DNA-based molecular identification of the four microorganisms, the infection rate of each microorganism was calculated from the number of samples from each host.

Chapter 3: Results

3.1 Prevalence, Distribution, and Molecular Record of Hard Ticks from Livestock

3.1.1 Tick Identification

Using the morphological features as described by the taxonomic keys, three genera, *Amblyomma, Hyalomma,* and *Rhipicephalus* (Figure 16 and 17) were identified. In genus *Amblyomma*, palp articles 2 are longer than articles 1 and 3 and basis capituli has straight lateral margins, and in genus *Rhipicephalus*, palp articles are all small and basis capituli has distinctly angular lateral margins (making a hexagonal shape of the entire basis capituli) (Figure 16). Whereas in genus *Hyalomma*, basis capituli has medium angular lateral margins (Figure 16).

Four species of ticks in three genera, namely, *H dromedarii*, *H. anatolicum*, *A. lepidum*, and *R. sanguineus* were identified based on morphology. In addition, the species designation was confirmed by the results of DNA sequencing, which was done in the molecular characterization. The primers for *cox1* and *16S* rRNA gene fragments produced the expected band size on the agarose gel.

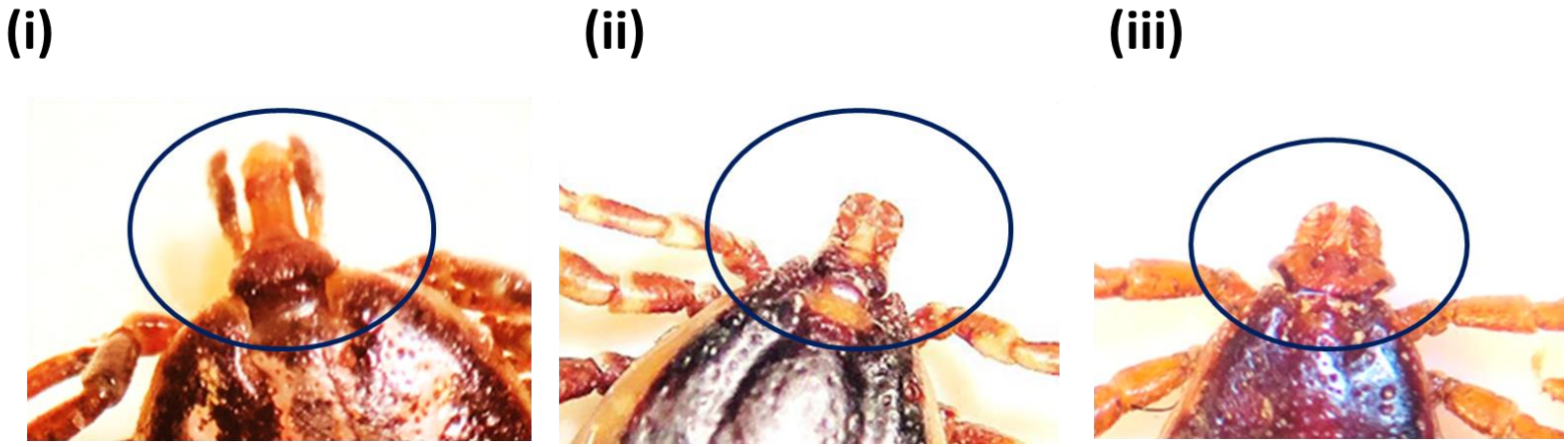

Figure 16: Head of tick genera. (i) *Amblyomma* (ii) *Hyalomma*, (iii) *Rhipicephalus*.

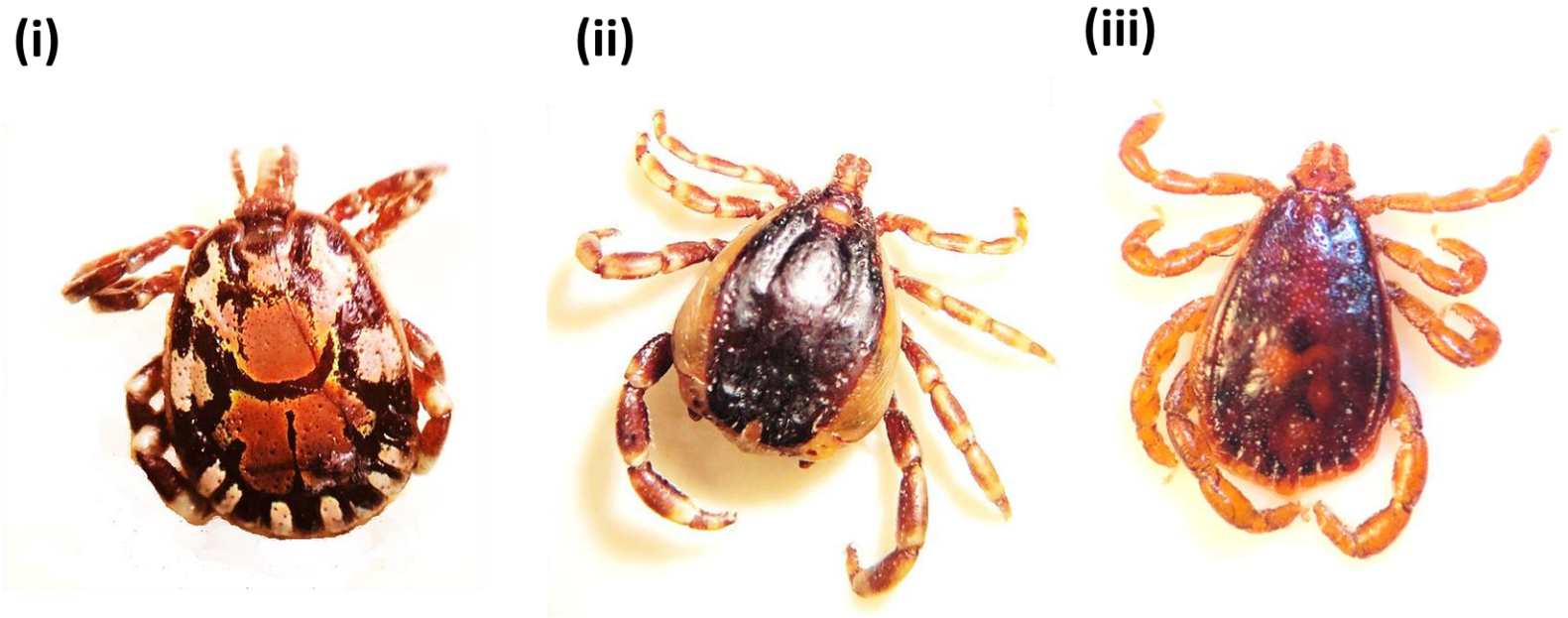

Figure 17: Tick genera. (i) *Amblyomma*, (ii) *Hyalomma*, (iii) *Rhipicephalus*

3.1.1.1 *Hyalomma Dromedarii*

The identification of *H. dromedarii* was confirmed based on the following diagnostic characteristics: the sub-anal plates were aligned outside the adanal plates in male ticks (Figure 18); the central festoon was pale colored; cervical and lateral grooves reached up to 2/3 of the length of conscutum; the marginal grooves were short and furrow-like; the paramedian grooves were well defined and large; the posteromedian groove was reaching parma; the cervical grooves were very deep; the basis capituli dorsal posterior margin was deeply concave; the dorsal prolongation of spiracular plates was long and narrow, and the posterolateral spurs were longer than the posteromedian spur and taper to apices. *Hyalomma dromedarii* was identified from only camels in all three emirates by using *cox1* and *16S* rRNA genes. The species was confirmed based on sequence similarity with GenBank records (Appendix 3). A representative sequences of *H. dromedarii* from camel in this study was submitted in the GenBank (MZ976772). The UAE specimen showed the 99.50% similarity to the sequences of *H. dromedarii* detected from camels in Tunisia (MN960589.1), and Egypt (MG757400.1) with sequence coverage of 95% (Appendix 3).

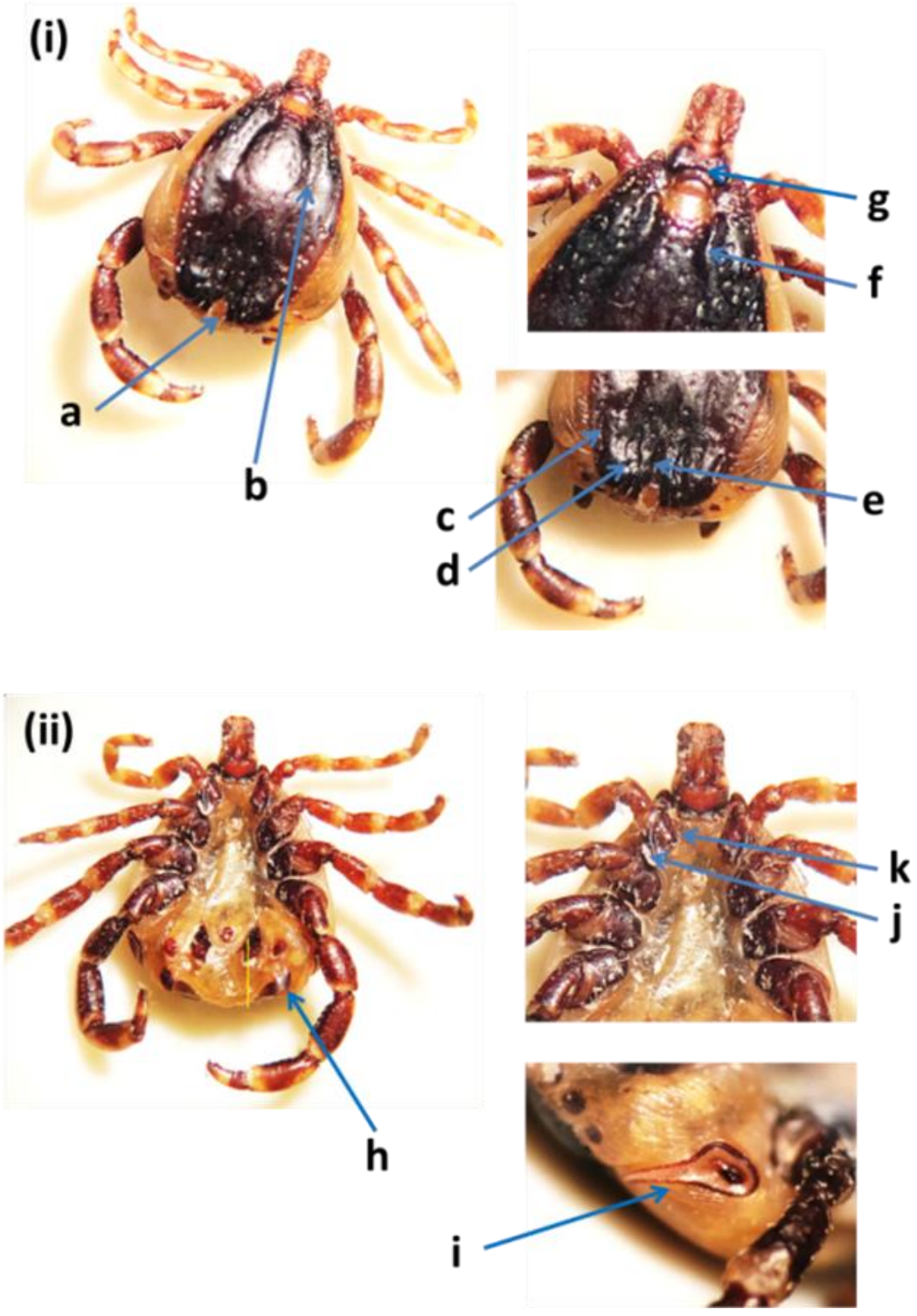

Figure 18: *Hyalomma dromedarii* male from the UAE. (i) dorsal view (a) central festoon (b) cervical and lateral grooves (c) marginal grooves (d) paramedian grooves (e) posteromedian groove (f) cervical grooves (g) basis capituli (ii) ventral view (h) sub-anal plates (i) spiracular plates (j) posterolateral spurs (k) posteromedian spur.

3.1.1.2 *Hyalomma Anatolicum*

Hyalomma anatolicum ticks were small in size (about 2.69 mm in length), oval shape, and reddish-brown in color (Figure 19). Cervical grooves and lateral grooves were shallow and reached 1/2 of the length of conscutum, the posteromedian groove was not reaching the parma. The spurs of coxae I were close together, the medial spur was wider than the lateral, in the form of a triangle. The lateral spur of coxae I was narrow curved. The sub-anal shields were situated on the axis of the adanals. *Hyalomma anatolicum* was identified from camels, cows, sheep, and goats in the three emirates by using *16S* rRNA and *cox1* genes. However, it was not detected in tick samples collected from camels in Dubai and Sharjah. DNA fragments were identified based on sequence similarity with the records of both *16S* rRNA and *cox1* genes from the GenBank (Appendix 4). A representative sequences of *H. anatolicum* from cow, sheep, and goat (MZ976771, MZ976770, and MZ976780) for *16S* rRNA gene and from cow (OK017169) for *cox1* gene were deposited in the GenBank. This sequence was 99.70% identical to the *H. anatolicum* detected from goat in Pakistan (MT800311.1), and 99.39% similar to *H. anatolicum* reported in China (MH459380.1) with sequence coverage of 96%, and 97%, respectively.

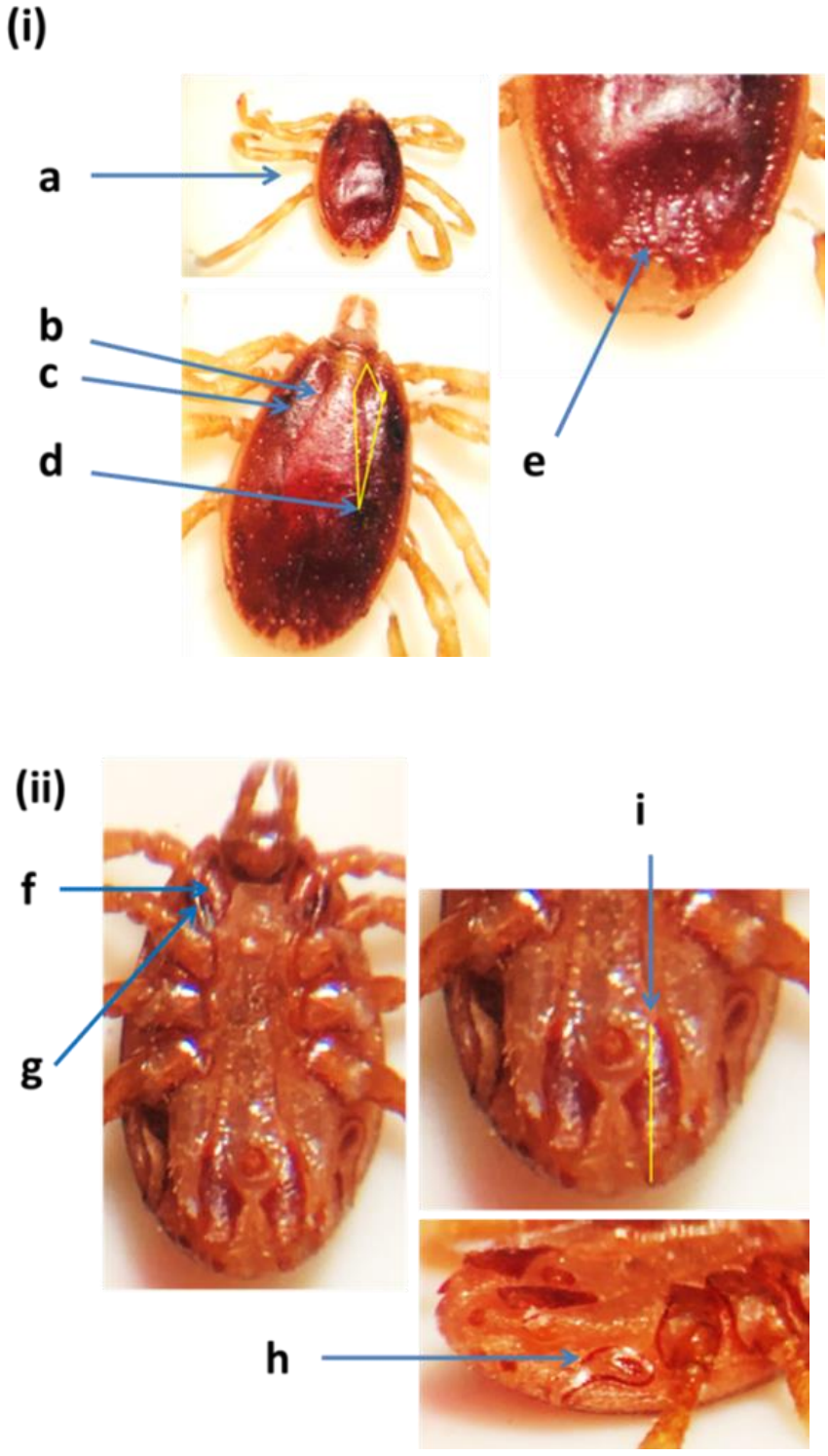

Figure 19: *Hyalomma anatolicum* male from the UAE. (i) dorsal view (a) reddish-brown and oval shape (b)(c) cervical grooves and lateral grooves (d) cervical and lateral grooves length (e) posteromedian groove (ii) ventral view (f) coxae I spurs close together, medial spur in the form of triangle (g) coxae I lateral spur (narrow) (h) spiracle plate (i) subanal shields situated on the axis of the adanals**.**

3.1.1.3 *Amblyomma Lepidum*

Amblyomma lepidum found in the UAE (Figure 20) was a large and ornamented tick with long mouthparts. Primary punctuation size was small and distribution was localized (between the eyes). The enamel color was pink to orange. The posteromedian strip was narrow. Enameling of the festoons was partial (no enamel on central and two outermost festoons). Leg coloration was with the pale ring. Lateral median areas of enamel orientation were large. Eyes were distinctly convex. The length of internal spur of coxae I was short. Coxae II and III were with a broad salient ridge-like spur. The length of external spur of coxae I was median. In addition, *16S* rRNA and *cox1* genes were used to confirm this species. *Amblyomma lepidum* was not detected in livestock tick samples from Abu Dhabi and Sharjah. DNA fragments were identified based on DNA sequence similarity with the records of *cox1* gene from the GenBank (Appendix 5) A representative sequence of *A. lepidum* was deposited in the GenBank (OK001821). This sequence was 99.84% identical to the *A. lepidum* detected from sheep in Israel (KP987775.1), and 99.38% similar to *A. lepidum* reported in Kenya (KT307492.1) with sequence coverage of 93% and 94%, respectively.

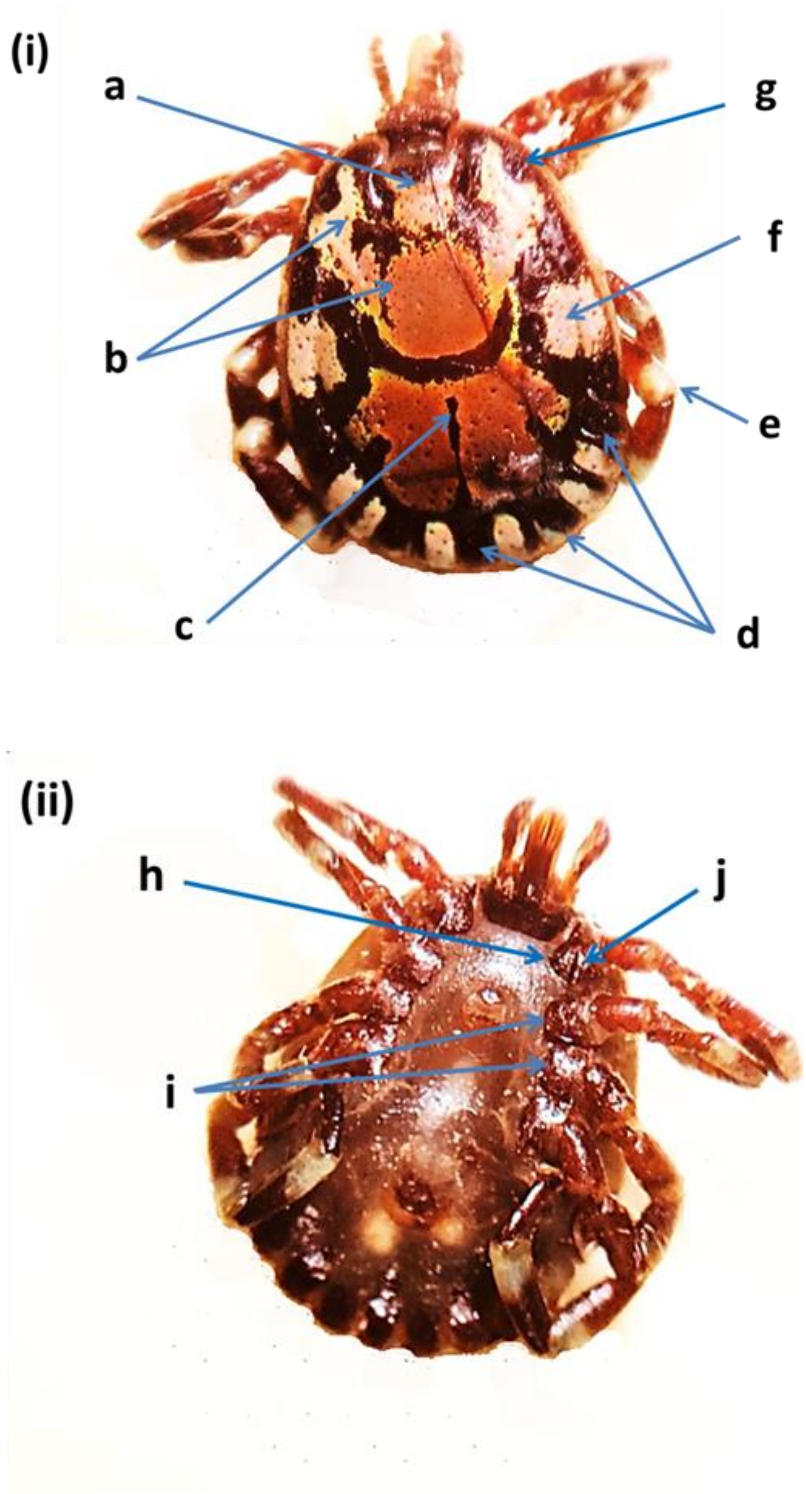

Figure 20: *Amblyomma lepidum* male from the UAE. (i) dorsal view (a) primary punctuation (b) enamel color (c) posteromedian strip (d) festoon enameling (e) leg with pale ring (f) lateral median areas of enamel orientation (g) eyes (distinctly convex) (ii) ventral view (h) coxae I internal spur length (i) coxae II and III (j) coxae I external spur length.

3.1.1.4 *Rhipicephalus Sanguineus*

Rhipicephalus sanguineus is a brown-color tick and was present only on cows from Dubai. Diagnostic features are given in (Figure 21). Marginal lines were heavily punctate. Furthermore, marginal lines were long, deep, and reaching anteriorly almost to eye level. The posterior grooves were comma-shaped and three in numbers, one posteromedian groove, and two posterolateral grooves. The adanal plates were curved but not sickle shaped. Subadanal plates were absent. The spiracle plate had a narrow tail. *Rhipicephalus sanguineus* was confirmed based on sequence similarities with GenBank record (Appendix 6). A representative sequence of *R. sanguineus* from a cow was submitted in the GenBank (MZ976769). This sequence showed 99.03% similarity to the sequences of *R. sanguineus* detected from dogs in India (MG066692.1), 98.56% from dogs in Taiwan (AY883868.1), and 98.33% from dogs in Cuba (KP830114.1) with sequence coverage of 96%, 98%, and 98%, respectively.

Accession numbers of all representative sequences of identified ticks, *H. dromedarii, H. anatolicum, A. lepidum,* and *R. sanguineus* are given in Table 5.

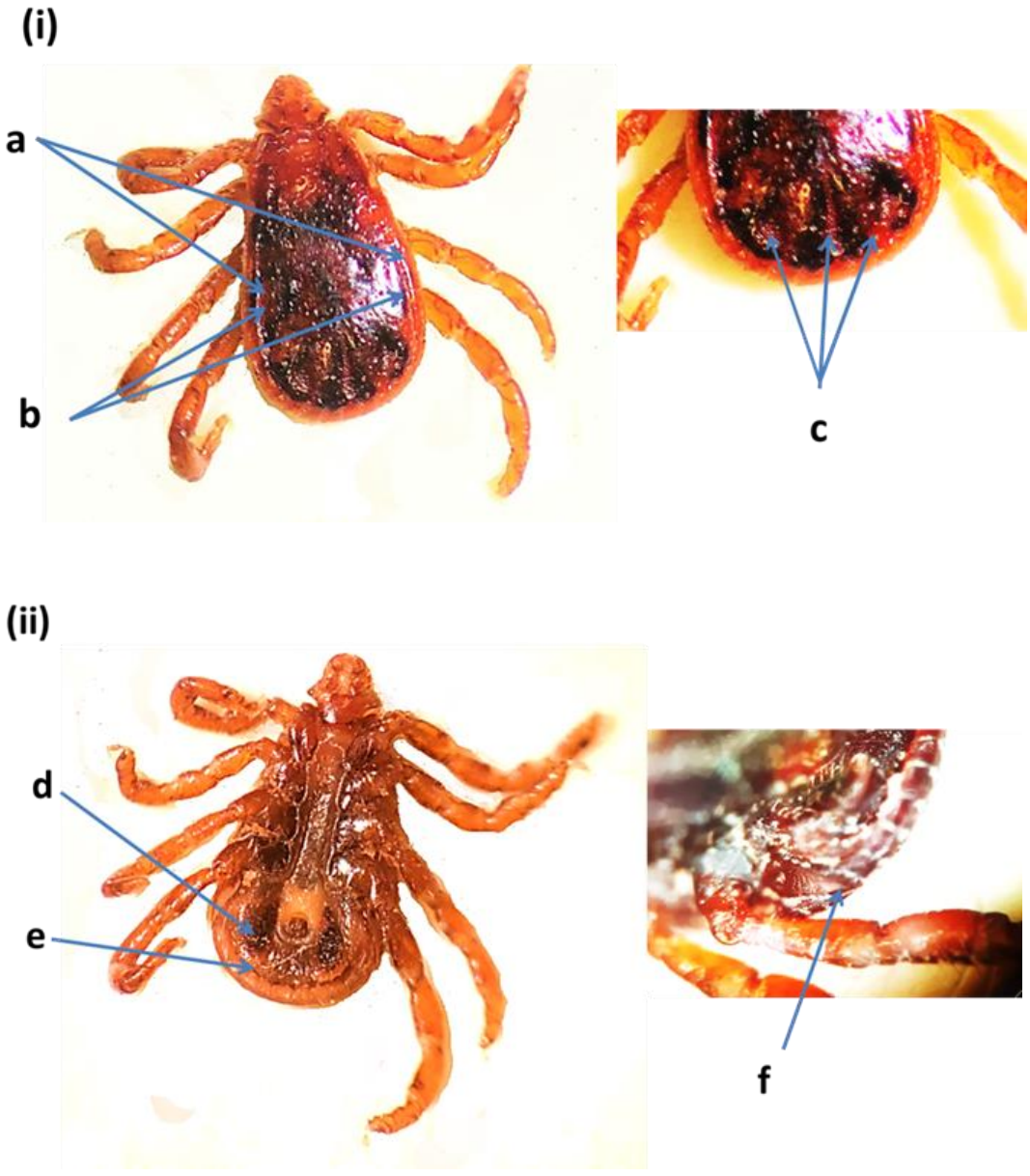

Figure 21: *Rhipicephalus sanguineus* male from the UAE. (i) dorsal view (a) marginal lines (b) marginal lines anteriorly almost to eye level (c) posterior grooves (comma shaped) (ii) ventral view (d) adanal plates (e) subadanal plates absent, (f) spiracle plate.

Table 5: Identity of tick species and percentage similarity value with the reference sequences from the GenBank.

Sample Accession	**Host**	**Location**	**GenBank Reference**		**Identity %**	**Species**
			16S	***cox1***		
MZ976772	Camel	Abu Dhabi	L34306.1	-	99.27	*H. dromedarii*
OK017169	Cow	Dubai	-	MT800311.1	99.70	*H. anatolicum*
MZ976771	Cow	Dubai	MK829042.1	-	99.28	*H. anatolicum*
MZ976770	Sheep	Dubai	MK829042.1	-	99.52	*H. anatolicum*
MZ976780	Goat	Dubai	KC203338.1	-	99.51	*H. anatolicum*
MZ976769	Cow	Sharjah	MG066692.1	-	99.03	*R. sanguineus*
OK001821	Cow	Dubai	-	KP987775.1	99.84	*A. lepidum*

3.1.2 Tick Prevalence

The prevalence of ticks in camels (94%) was very high as compared to cows, sheep, and goats (Figure 22; Table 6) (Appendix 7) (Fisher's Exact test, $p < 0.001$ for all pairwise comparisons). The prevalence of ticks in cows (38%) was also very high as compared to goats (14%) (Fisher's Exact test, $p < 0.001$), however, tick prevalence did not differ significantly between cows (38%) and sheep (37%) (Fisher's Exact test, $p = 1.00$). In addition, the prevalence of ticks in sheep (37.1%) was also very high as compared to goats (14%) (Fisher's Exact test, $p < 0.001$). In relation to the sex of hosts, the prevalence of ticks did not differ significantly between male and female hosts (Fisher's Exact test, $p > 0.05$ for all pairwise comparisons) except in goats, where prevalence was higher in females (25.9%) than males (6.8%) (Fisher's Exact test, $p = 0.036$) (Figure 23; Table 7) (Appendix 8). We did not find any ticks on Australian cows.

The mean intensity of ticks on camels was significantly higher than on sheep (Bootstrap 2-sample t-test, $p < 0.001$). However, the mean intensity of ticks did not differ significantly between camels and cows, and between camels and goats (Bootstrap 2-sample t-test, $p > 0.05$ for all pairwise comparisons). Further, the mean intensity of ticks on cows was significantly higher than on sheep (Bootstrap 2-sample *t*-test, $p < 0.005$) for pairwise comparison. There was no significant difference between mean intensities in pairwise comparisons between cows and goats, and between sheep and goats (Bootstrap 2-sample *t*-test, $p > 0.05$).

Mean tick abundance on camels was significantly higher than on cows, sheep, and goats (Bootstrap 2-sample *t*-test, $p < 0.001$ for all pairwise comparisons). However, there was no difference in mean abundance of ticks on goats and sheep, and

cows and goats (Bootstrap 2-sample *t*-test, $p > 0.05$ for all pairwise comparisons). Mean tick abundance on cows was significantly higher than on sheep (Bootstrap 2-sample *t*-test, $p < 0.01$). *Hyalomma dromedarii* ticks were collected in large numbers only from camels in all three emirates with 94.3% prevalence (Table 7). Whereas, *H. anatolicum* was found in all emirates on all hosts, camels, cows, sheep, and goats. The prevalence of *H. anatolicum* on cows (32.8%) was high as compared to camels, goats, and sheep (Fisher's Exact test, $p < 0.01$ for all pairwise comparisons). However, the prevalence of *H. anatolicum* did not differ significantly between sheep and goats, and between camels and goats (Fisher's Exact test, $p > 0.05$ for all pairwise comparisons). Mean intensity and mean abundance of *H. anatolicum* in all hosts did not differ significantly (Bootstrap 2-sample *t*-test, $p > 0.05$ for all pairwise comparisons), except camels and cows (Bootstrap 2-sample *t*-test, $p < 0.01$). *Amblyomma lepidum* was recorded in cows from Dubai and *R. sanguineus* was recorded in cows from Sharjah, in the least numbers with 0.8% prevalence. The engorged nymphs and engorged female ticks, which were difficult to identify, were included in a category named "others" (Appendix 9).

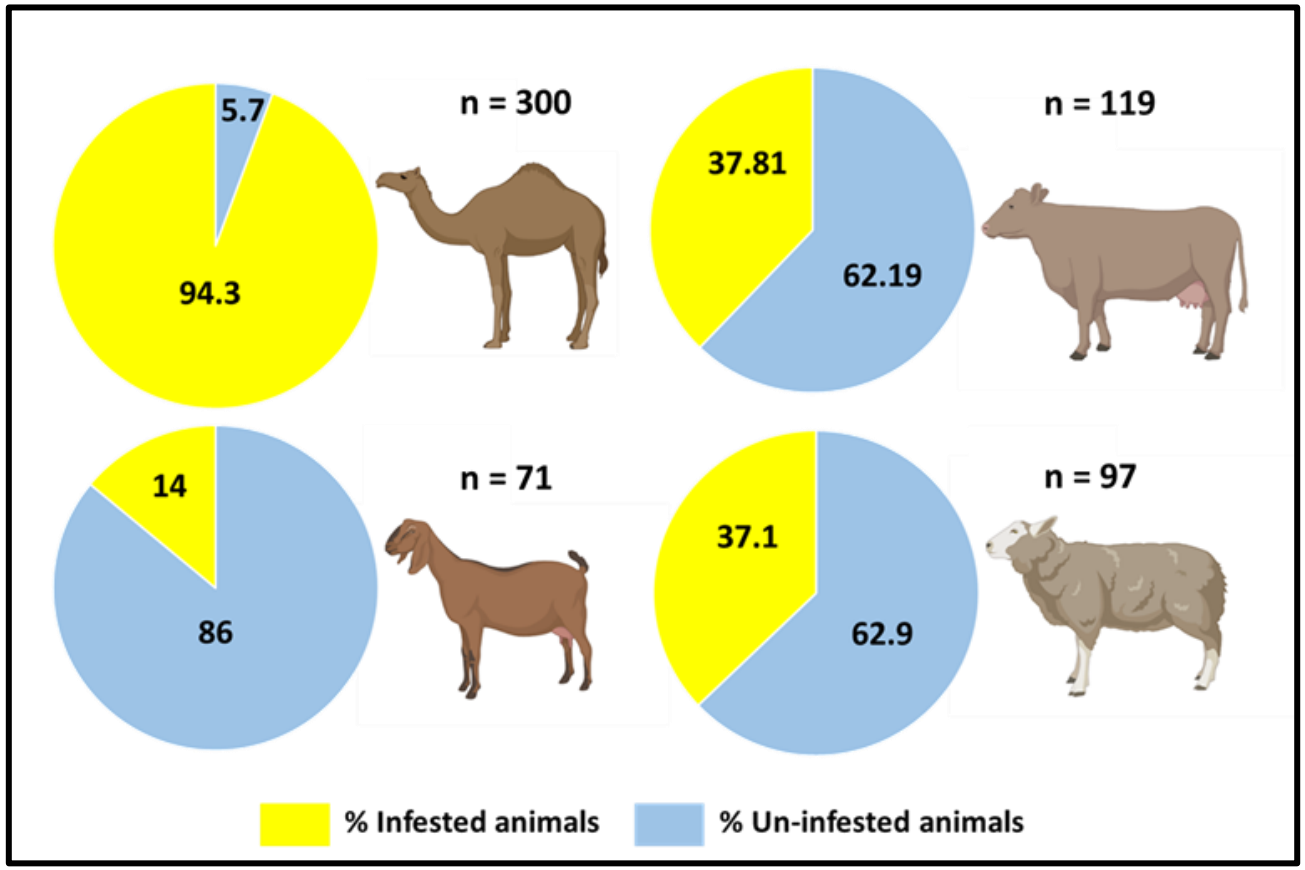

Figure 22: Prevalence of ticks per host (camel, cow, goat, and sheep) in the sampling areas, UAE.

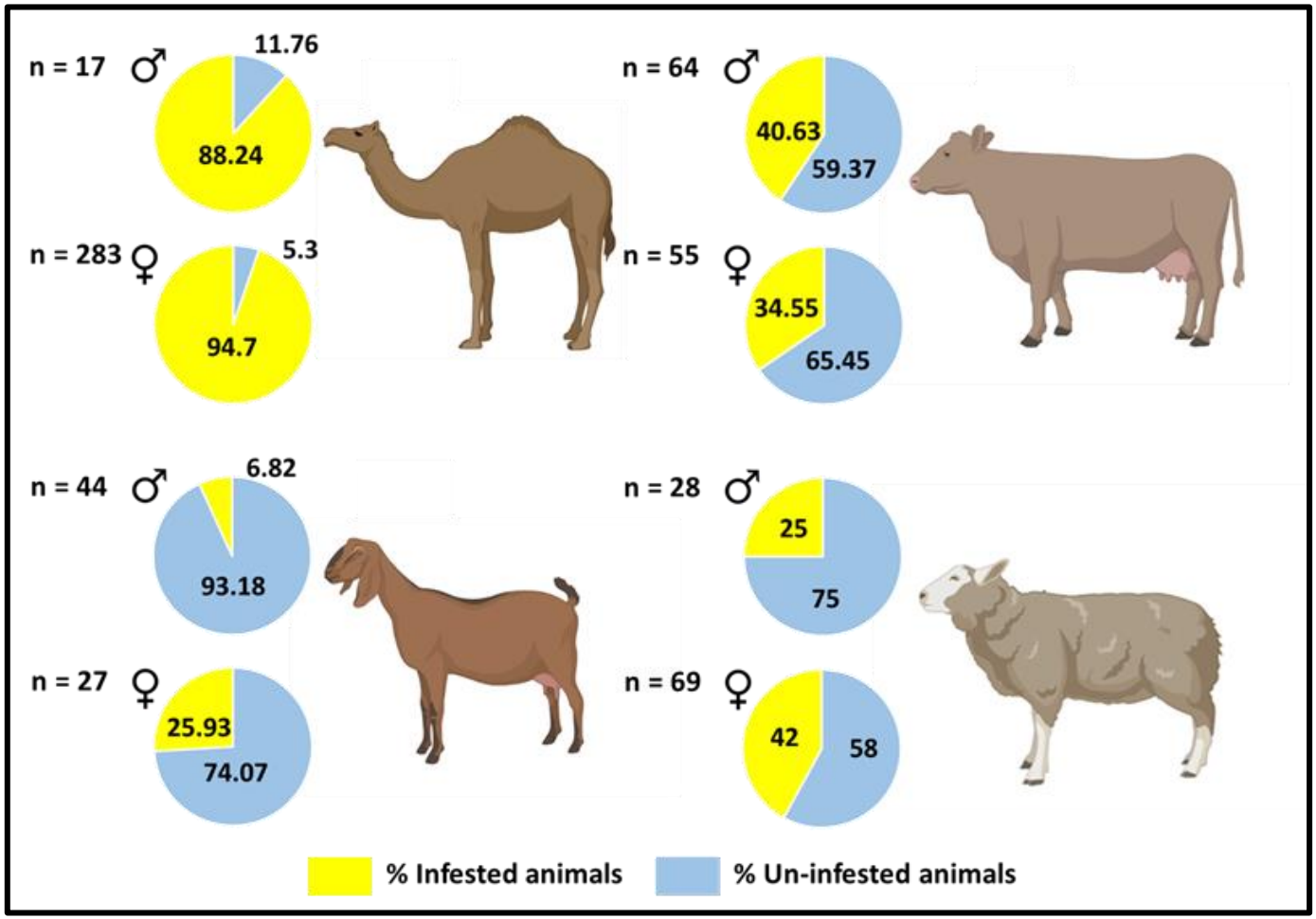

Figure 23: Prevalence of ticks per host (camel, cow, goat, and sheep) in relation to the sex of the host animal.

Table 6: Tick prevalence, mean intensity, mean abundance on camels, cows, sheep, and goats in the sampling areas, UAE.

Hosts	Examined Animals	Infested with Ticks	Prevalence (95% Confidence Level)	Mean Intensity (95% Confidence Level)	Mean Abundance (95% Confidence Level)
Camels	300	283	0.94 (0.91–0.97)	17 (15.23–19.52)	16 (14.39–18.63)
Cows	119	45	0.38 (0.29–0.47)	14.47 (11.18–18.87)	5.47 (3.99–7.87)
Sheep	97	36	0.37 (0.28–0.48)	7.69 (5.69–10.67)	2.85 (1.96–4.36)
Goats	71	10	0.14 (0.07–0.24)	21.9 (10.50–55)	3.08 (1.14–9.08)

Table 7: Number (n) of tick species collected from camels, cows, sheep, and goats in the sampling areas, UAE.

Hosts	**Camels**			**Cows**			**Sheep**			**Goats**		
	P	**MI**	**MA**	**P**	**MI**	**MA**	**P**	**MI**	**MA**	**P**	**MI**	**MA**
H. dromedarii	94.3	16.52	15.58	0	0	0	0	0	0	0	0	0
H. anatolicum	3.7	1.18	0.04	32.8	8.51	2.79	14.4	10.36	1.5	9.9	15.14	1.49
R. sanguineus	0	0	0	0.8	5	0.04	0	0	0	0	0	0
A. lepidum	0	0	0	0.8	2	0.02	0	0	0	0	0	0
Others	8	4.83	0.39	36.1	7.26	2.62	34	4	1.4	14.1	11.7	1.65

P = prevalence, MI = Mean intensity, MA = Mean Abundance.

3.2 Seasonal Population Dynamics of Hyalomma Dromedarii on Camels

A total of 2658 ticks were collected from camels (n = 25) during 12 visits, of which 216 were nymphs. No larvae were found on any of the camels. *Hyalomma dromedarii* lifecycle with field-collected stages is shown in Figure 24. The sex ratio of the ticks (M: F) was calculated that was 1.92: 1. Tick burden was highest in June and was lowest in November (Figure 25). The infestation prevalence was calculated and it was high during the whole study, with a mean of 94.33% (Table 8). Camels were found infested with ticks throughout the year, and the infestation prevalence was 100% from March 2019 to October 2019 (Table 8). The predilection sites of ticks were the humid skin regions of the camel body. Perianal and vulvar regions, udder, and inguinal regions were heavily infested with ticks as compared to the pinna and the chest region (from where fewer ticks were collected). The relative abundance was 17.72 ticks /animal while the mean overall intensity of infestation was 18.52 ticks /animal. In June, the infestation intensity was maximum, 38.32 ticks /animal, and was minimum in November, 12.63 ticks /animal (Table 8). The correlation between monthly average temperature and tick average burdens was found non-significant ($r = 0.3646$, $p = 0.2439$) (Figure 25). Also, the correlation between monthly average relative humidity and tick average burdens was non-significant ($r = -0.54$, $p = 0.0694$) (Figure 25). During March, nymphs were found in maximum percentage however, the numbers reached zero in June and November (Figure 26). As compared to female ticks, male ticks were present in maximum numbers throughout the year on camels (Figure 27). There was a significant difference in tick burden between 12 months ($F = 9.310$, $df = 11, 288$, $p < 0.0001$).

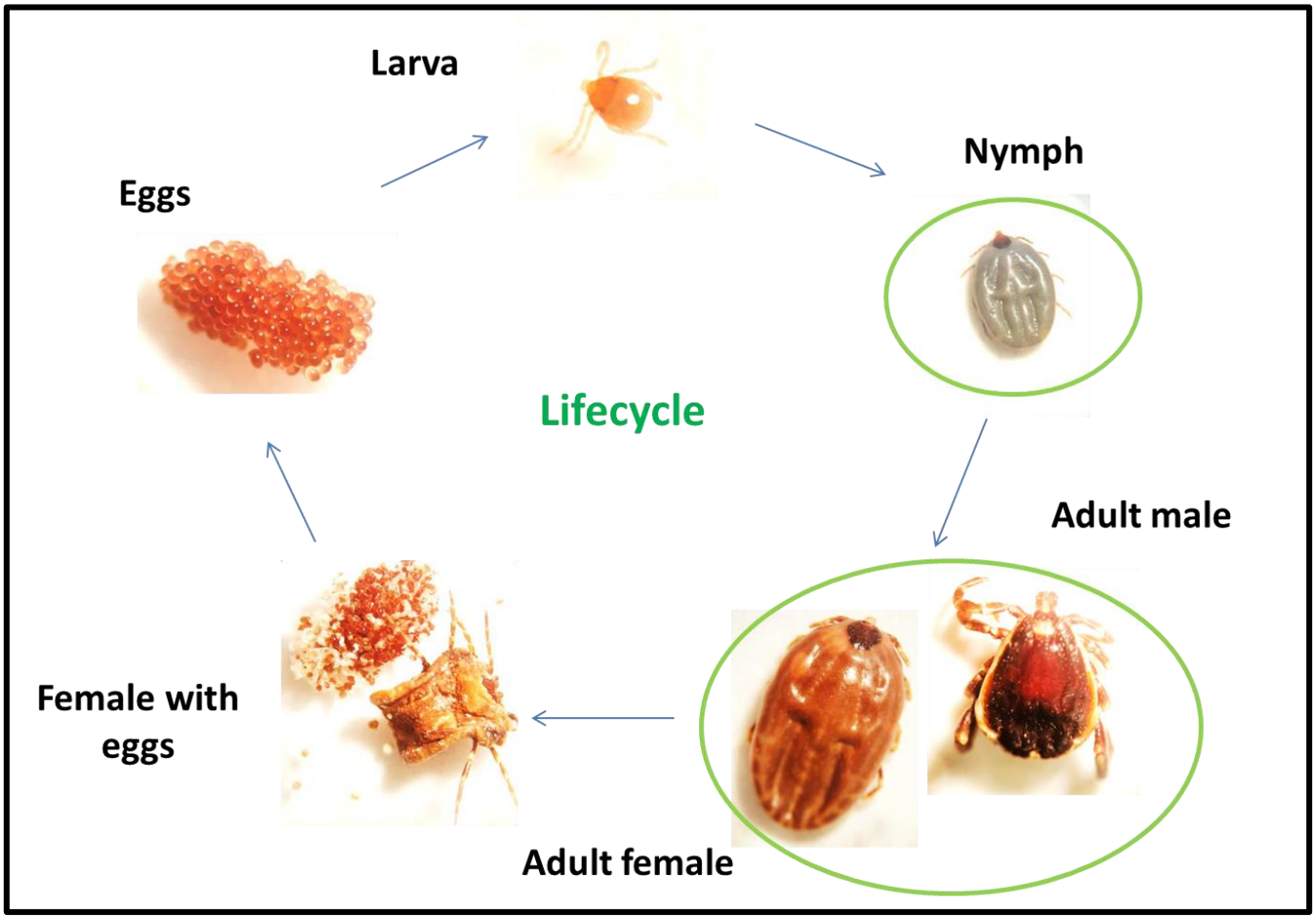

Figure 24: Lifecycle of camel tick, *H. dromedarii*. Field collected stages are in green circles.

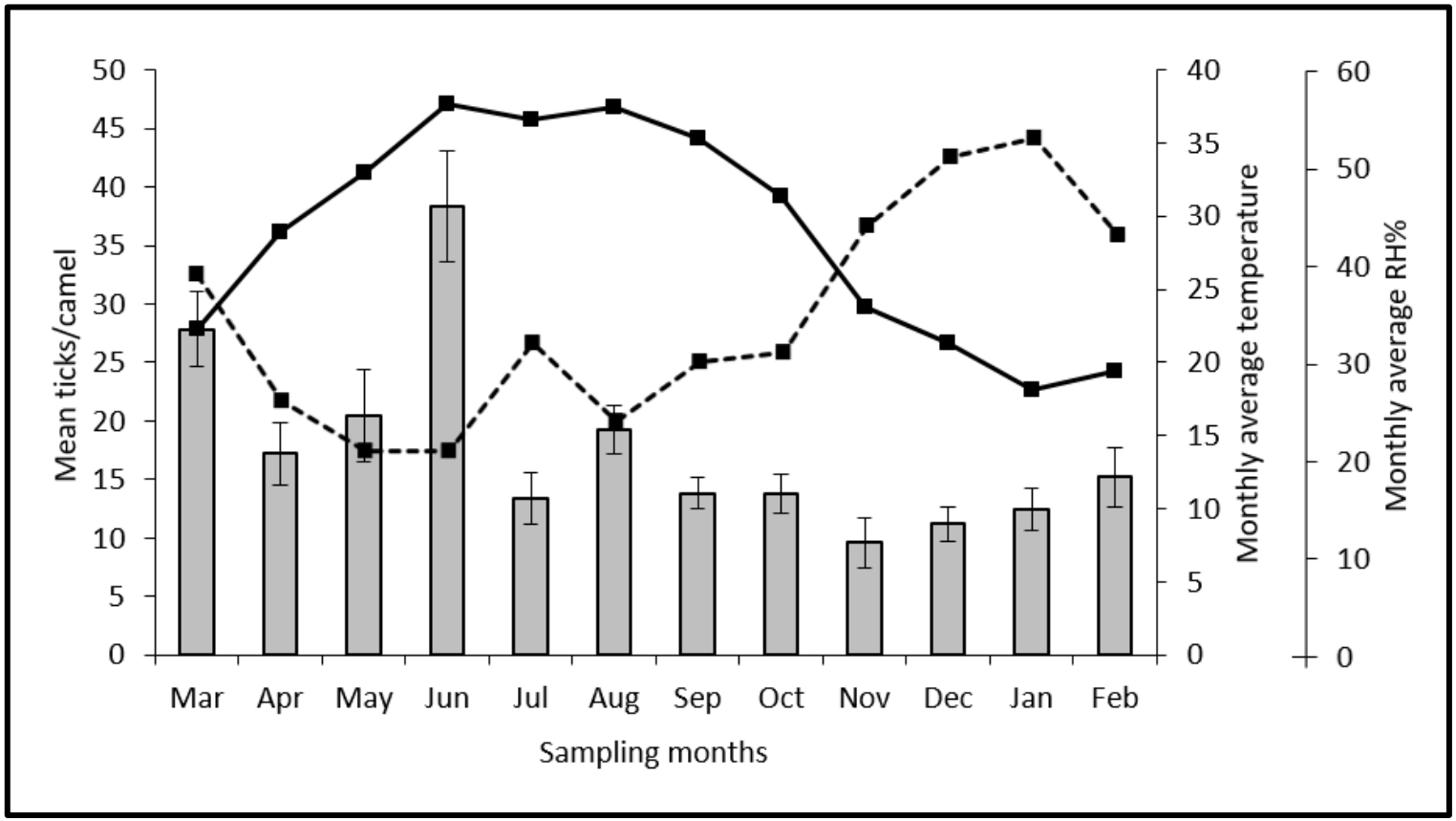

Figure 25: Fluctuation of *H. dromedarii* tick load on camels in Al Ain, UAE. Continuous line: monthly average temperature (°C). Dotted line: monthly average relative humidity (%). Column: mean (±SE) monthly ticks/camel from March 2019 to February 2020.

Table 8: Prevalence and infestation intensity of *H. dromedarii* tick on camels in Al Ain, UAE.

Year-Month	Total examined animals	Total infested animals	No. of ticks	Prevalence %	Infestation intensity
19-March	25	25	696	100	27.84
19-April	25	25	430	100	17.2
19-May	25	25	512	100	20.48
19-June	25	25	958	100	38.32
19-July	25	25	334	100	13.36
19-August	25	25	482	100	19.28
19-September	25	25	346	100	13.84
19-October	25	25	346	100	13.84
19-November	25	19	240	76	12.63
19-December	25	21	280	84	13.33
20-January	25	20	312	80	15.6
20-Feburary	25	23	380	92	16.52

Overall prevalence: 94.33%, Mean infestation intensity: 18.52.

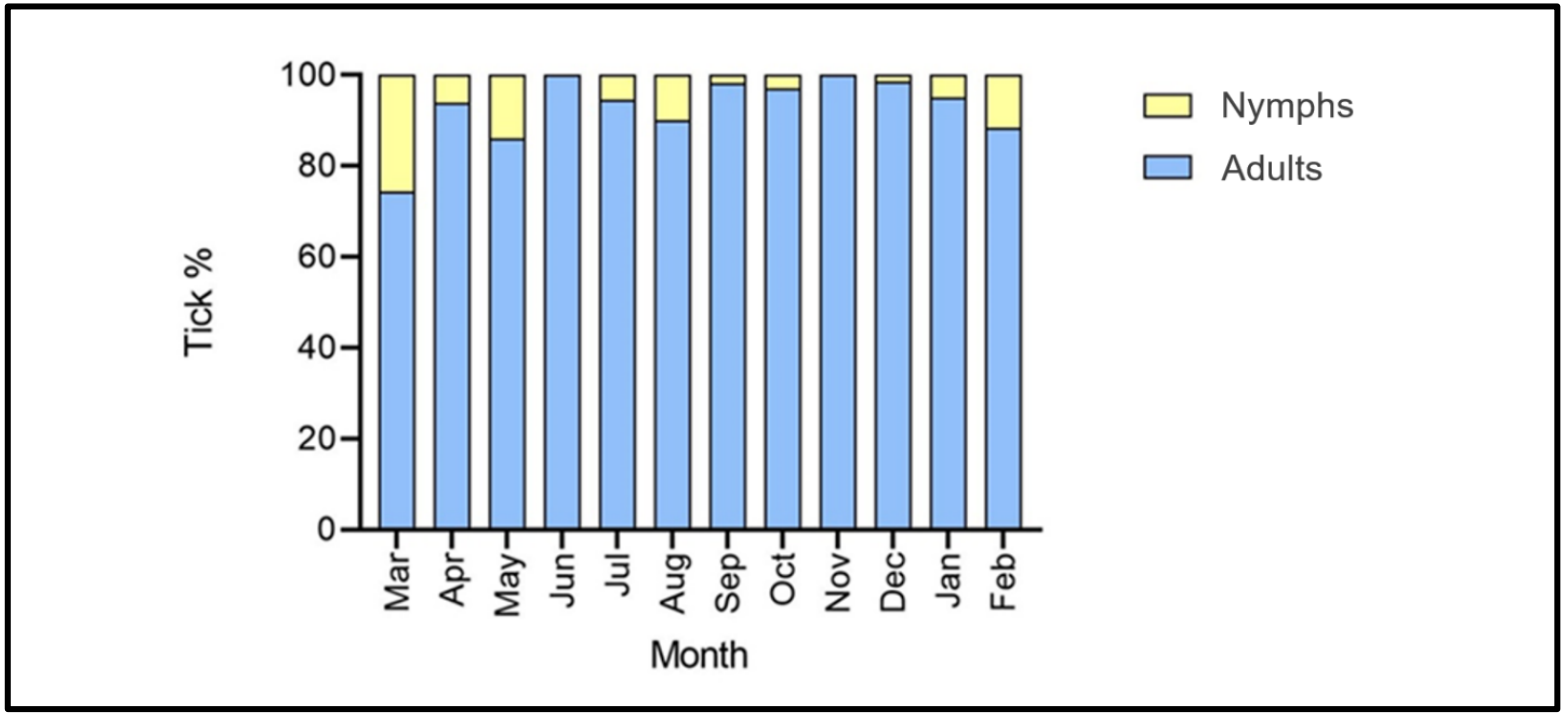

Figure 26: Adult and nymph *H. dromedarii* tick percentages on camels in twelve months (March 2019 to February 2020) in Al Ain, UAE.

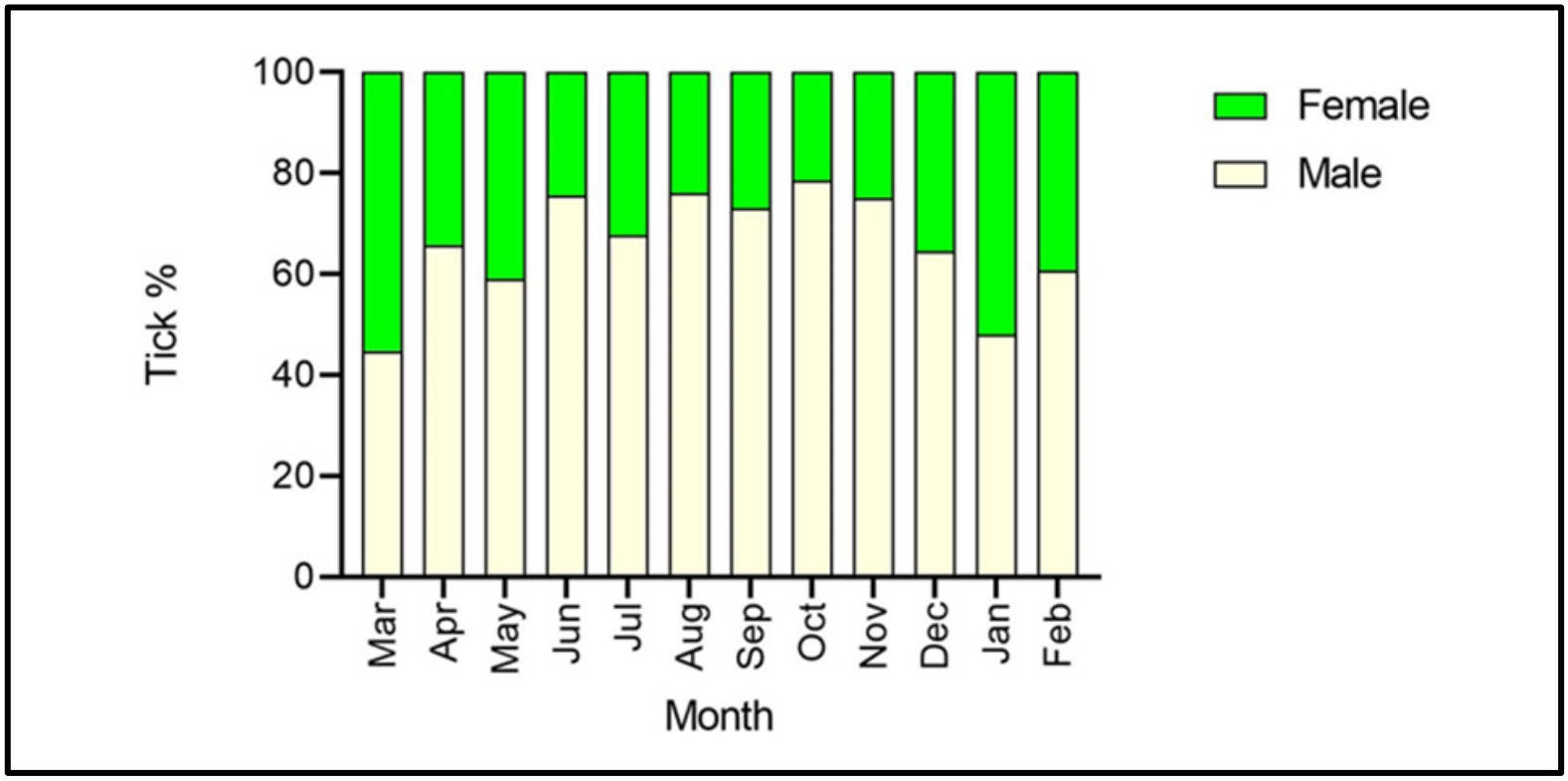

Figure 27: Adult male and female *H. dromedarii* tick percentages on camels in twelve months (March 2019 to February 2020) in Al Ain, UAE.

3.3 Bacterial Communities' Composition and Diversity in Camel Tick, *Hyalomma dromedarii* using Next-Generation Sequencing

3.3.1 Microbial Diversity in 2010

Total read counts, 899,574 (average 89,957 sequences per sample) were obtained, and these formed 371 operational taxonomic units (OTUs, clustered at 97% similarity). Further, these OTUs belonged to 10 phyla, 24 classes, 107 families, and 202 genera from 2010 samples.

3.3.1.1 Relative Abundance of Bacterial Phyla in 2010

Taxonomic profiling of the bacteria established from *H. dromedarii* confirmed seven abundant phyla: *Proteobacteria, Bacteroidetes, Firmicutes, Actinobacteria, Cyanobacteria, Verrucomicrobia,* and *Planctomycetes*. The phylum *Proteobacteria* was detected from all ten locations. Further, it was the most abundant (87.86%) phylum whereas the *Planctomycetes* had the least abundance (0.28%) and was found only at Omghafa (Appendix 10).

3.3.1.2 Relative Abundance of Bacterial Classes in 2010

Out of 24 bacterial classes, 16 classes were abundant namely *Gammaproteobacteria, Flavobacteriia, Actinobacteria, Bacilli, Tissierellia, Bacteroidia, Clostridia, Betaproteobacteria, Alphaproteobacteria, Sphingobacteriia, Negativicutes, Chitinophagia, Planctomycetia, Erysipelotrichia, Deltaproteobacteria,* and *Verrucomicrobiae. Gammaproteobacteria* was recorded as the dominant class in all locations except two, Dwar Al-Shahenat (DS) and Al-Wagan (AW) (Appendix 11). At DS, *Actinobacteria* was more abundant (41.60%) than *Gammaproteobacteria*

(37.03%) and, at AW *Bacilli* was the abundant class (41.90%) as compared to *Gammaproteobacteria* (24.45%).

3.3.1.3 Relative Abundance of Bacterial Families in 2010

Taxonomic assignment showed that 25 bacterial families were more abundant (Appendix 12). The ones with the highest relative abundance were: *Moraxellaceae* (77.52%), *Morganellaceae* (55.82%), *Enterobacteriaceae* (54.63%), *Staphylococcaceae* (38.1%), *Bacillaceae* (37.33%), *Corynebacteriaceae* (36.62%), *Flavobacteriaceae* (26.66%), *Xanthomonadaceae* (24.5%), *Francisellaceae* (11.4%) and *Neisseriaceae* (8%) in all of the sampled locations (Figure 28A).

3.3.1.4 Relative Abundance of Bacterial Genera in 2010

The relative abundance of genera was highly variable in the microbiome of *H. dromedarii* in all locations. *Acinetobacter* (75.66%) and *Corynebacterium* (36.62%) were the two most common genera with high relative abundance. *Proteus* had a relative abundance of 55.82% followed by *Escherichia* (53.13%) and *Staphylococcus* (37.68%). *Flavobacterium*, *Francisella*, *Moraxella*, *Uruburuella*, and *Stenotrophomonas* occurred in moderately low relative abundance (6-25%). In addition, genera including *Enterobacter*, *Comamonas*, *Brevibacterium*, *Helcococcus*, *Facklamia*, *Anaerococcus*, *Ignavigranum*, and *Muribaculum* were all low in terms of relative abundance (1-3.55%) (Figure 29A, Appendix 13).

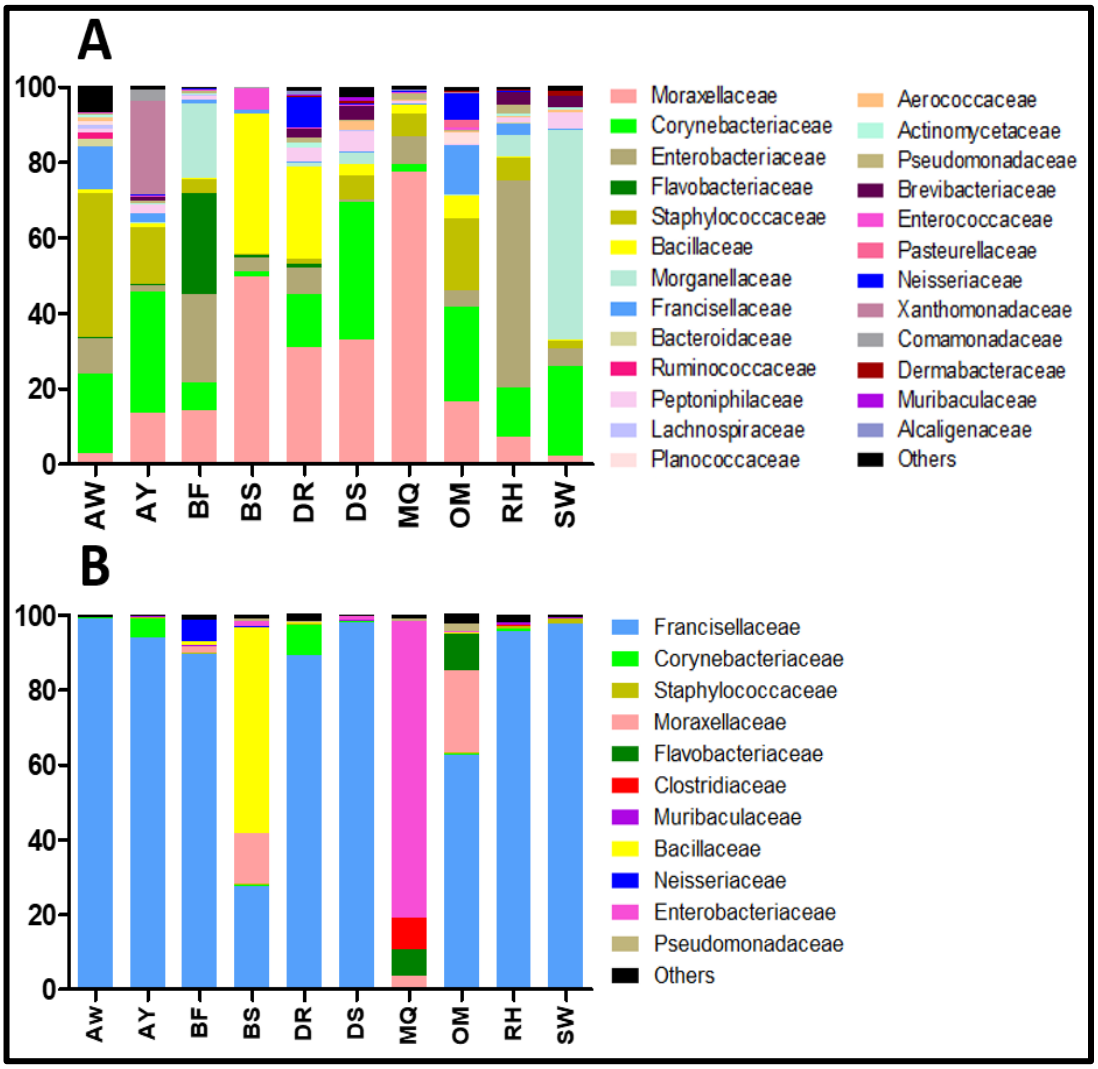

Figure 28: Microbial families detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2010 and 2019. 2010 (A) and 2019 (B). AW Al-Wagan, AY Al-Yahar, BF Bede' Fares, BS Bede'Bent Suod, DR Dubai Road, DS Dwar Al-Shahenat, MQ Malaket, OM Omghafa, RH Remah, SW Swehan.

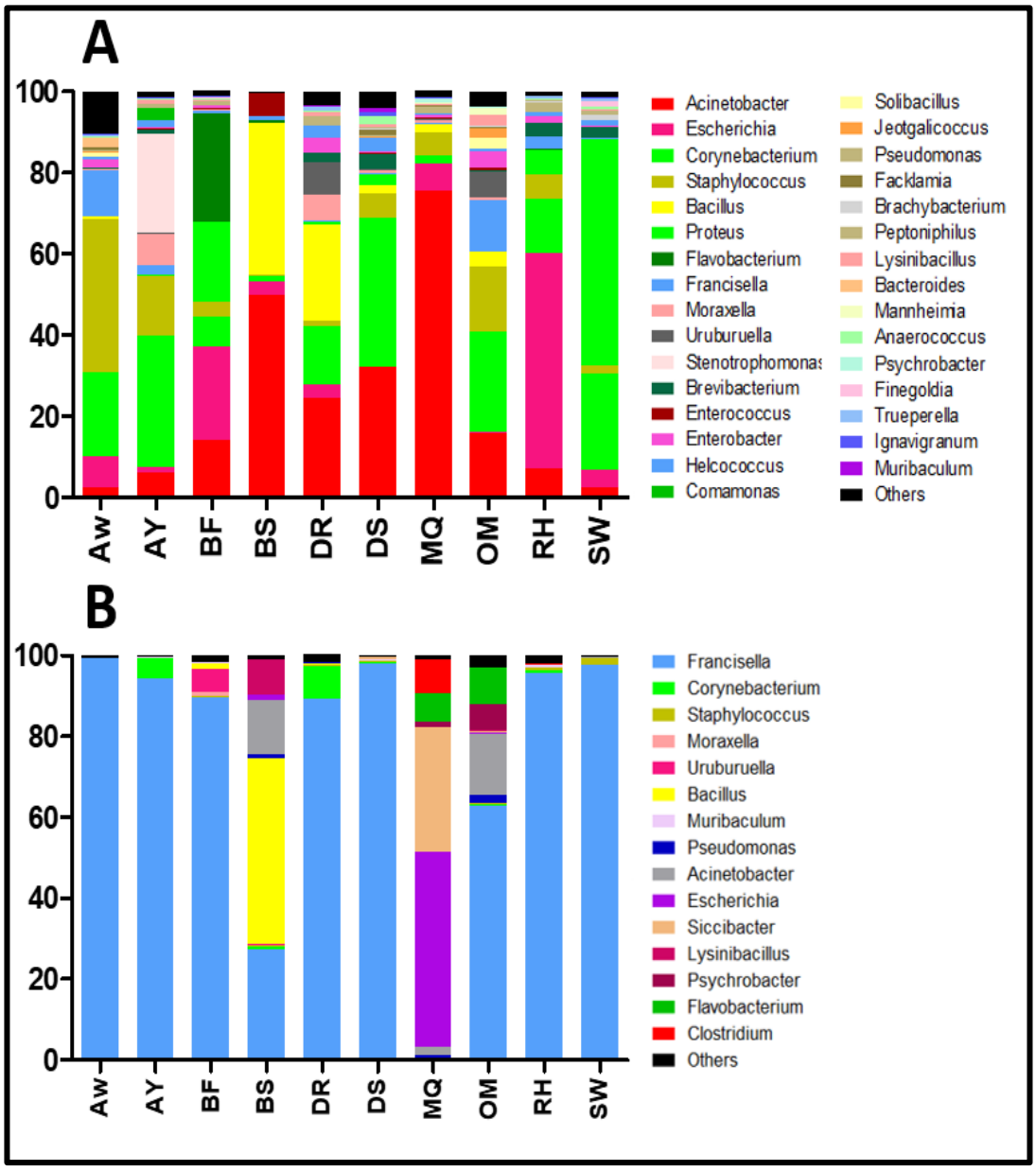

Figure 29: Microbial genera detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2010 and 2019. 2010 (A) and 2019 (B). AW Al-Wagan, AY Al-Yahar, BF Bede' Fares, BS Bede'Bent Suod, DR Dubai Road, DS Dwar Al-Shahenat, MQ Malaket, OM Omghafa, RH Remah, SW Swehan.

3.3.2 Microbial Diversity in 2019

Total 781,452 sequences (average 78,145 sequences per sample) were obtained and these formed 191 unique OTUs belonging to 7 phyla, 18 classes, 65 families, and 109 genera from 2019 samples.

3.3.2.1 Relative Abundance of Bacterial Phyla in 2019

Profiling of the bacteria sampled from *H. dromedarii* species identified seven phyla: *Proteobacteria, Actinobacteria*, *Bacteroidetes, Firmicutes, Cyanobacteria, Verrucomicrobia*, and *Planctomycetes*. *Proteobacteria* was dominant in all locations except one location, Bede'Bent Suod where *Firmicutes* had a high relative abundance (55.54%) as compared to *Proteobacteria* (43.80%) (Appendix 14).

3.3.2.2 Relative Abundance of Bacterial Classes in 2019

Taxonomic profiling revealed 14 bacterial classes with high abundance including *Gammaproteobacteria, Actinobacteria, Bacilli, Bacteroidia, Clostridia, Betaproteobacteria, Tissierellia, Flavobacteriia, Alphaproteobacteria, Thermoleophilia, Cytophagia, Sphingobacteriia, Negativicutes,* and *Verrucomicrobiae*. The composition of the classes indicated that *Gammaproteobacteria* was the dominant bacterial class in all locations, except one, Bede'Bent Suod, where *Bacilli* had a high relative abundance (55.52%) as compared to *Gammaproteobacteria* (43.21%). Other bacterial classes with high relative abundance include *Betaproteobacteria* and *Actinobacteria* (Appendix 15).

3.3.2.3 Relative Abundance of Bacterial Families in 2019

Overall, *Francisellaceae, Corynebacteriaceae, Staphylococcaceae, Moraxellaceae, Flavobacteriaceae, Clostridiaceae, Muribaculaceae, Bacillaceae, Neisseriaceae, Enterobacteriaceae,* and *Pseudomonadaceae* were the predominant families (Figure 28B, Appendix 16). *Enterobacteriaceae* had the highest relative abundance of 79.22% while *Bacillaceae* had a high relative abundance of 54.74%. However, *Francisellaceae* was dominant in all locations, with the highest relative abundance of up to 99.1% in one location. *Muribaculaceae* was found from most of the locations, however with low relative abundance.

3.3.2.4 Relative Abundance of Bacterial Genera in 2019

The dominant bacterial genus was *Francisella*. It was recorded in all locations, comprising up to 99.1% of relative abundance in one location (Figure 29B, Appendix 17). The *Escherichia* showed a relative abundance of 48.41% followed by *Bacillus,* which had a relative abundance of 45.84% while *Siccibacter* had the highest relative abundance of 30.81%. *Corynebacterium* was recorded in all locations; however, the *Clostridium* and *Flavobacterium* were recorded in samples from only two locations. Though *Staphylococcus* was recorded in all locations it was in a low relative abundance (Figure 29B).

3.3.3 Richness and Evenness of Microbes

The richness of tick microbiota (richness of taxa among the samples) associated with *H. dromedarii* in 2010 samples (371 OTUs) was higher compared to 2019 samples (191 OTUs). Principal Coordinates Analysis showed that Coordinates

1, 2, and 3 accounted for over 84% of the variation (based on cumulative Eigenvalues) and the first two coordinates accounted for over 78% of the variation. Furthermore, there was a clear separation among the microbial communities between years except for one site (Malaket, MQ) (Figure 30). Samples from this site in 2019 had a microbial community more closely aligned with the microbial communities from 2010. However, the microbial community of the same site in 2010 was completely different (Figure 30). The richness of genera differed significantly between years with higher richness recorded in 2010 (22.9 in 2010 versus 8.3 in 2019, two sample paired *t*-test, $p < 0.0001$). The Shannon Wiener index did not differ significantly between years (1.71 in 2010 versus 1.53 in 2019, two sample paired *t*-test, $p > 0.05$). In contrast, the Index of Evenness was significantly higher in 2010 (0.26 in 2010 versus 0.59 in 2019; two sample paired *t*-test, $t = 6.27$, $p = 0.0001$). The Index of Dominance (D) was not significantly different between years (0.27 in 2010 versus 0.27 in 2019; two sample paired *t*-test, $p > 0.05$).

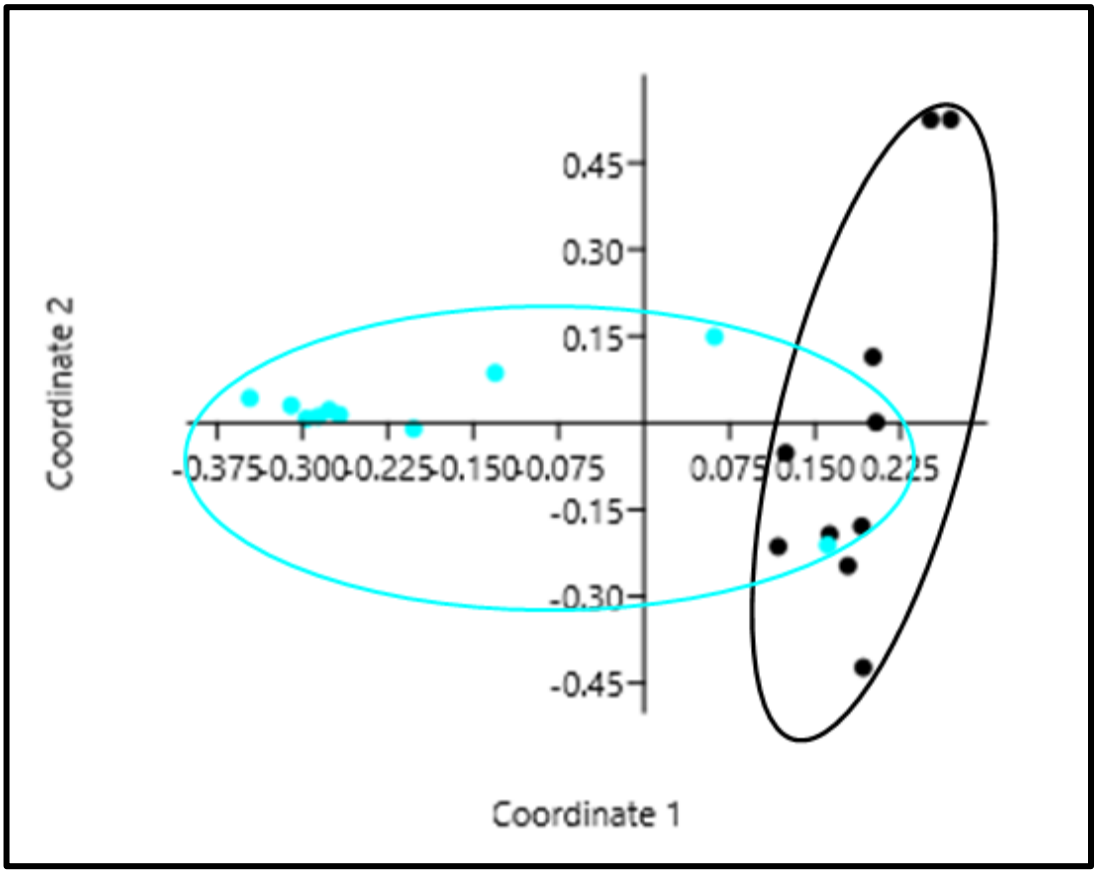

Figure 30: Principal Coordinates Analysis (PCoA) showing microbial diversity between years 2010 and 2019. 2010 (black circles) and 2019 (blue circles).

3.3.4 Associations between Bacterial Genera

Pearson's correlation coefficients (*r*) indicated that many bacterial genera were significantly correlated (Figure 31, Appendix 18). *Francisella* was significantly negatively correlated with *Acinetobacter*, *Corynebacterium*, and *Escherichia*. *Bacillus* was significantly positively correlated with *Lysinibacillus*. *Staphylococcus* and was positively correlated with *Corynebacterium*. *Escherichia* was significantly positively correlated with *Pseudomonas* and *Moraxella* was correlated with *Uruburuella*. *Acinetobacter*, *Francisella*, and *Escherichia* were significant predictors of many

bacterial genera (Table 9). *Acinetobacter* counts were significantly predicted by *Corynebacterium, Escherichia, Francisella, Lysinibacillus, Moraxella, Pseudomonas,* and *Psychrobacter*. Similarly, *Escherichia* counts were significantly predicted by *Acinetobacter, Corynebacterium, Francisella, Lysinibacillus, Moraxella, Pseudomonas,* and *Psychrobacter.* On the other hand, *Francisella* counts were significantly predicted by *Acinetobacter* and *Escherichia*.

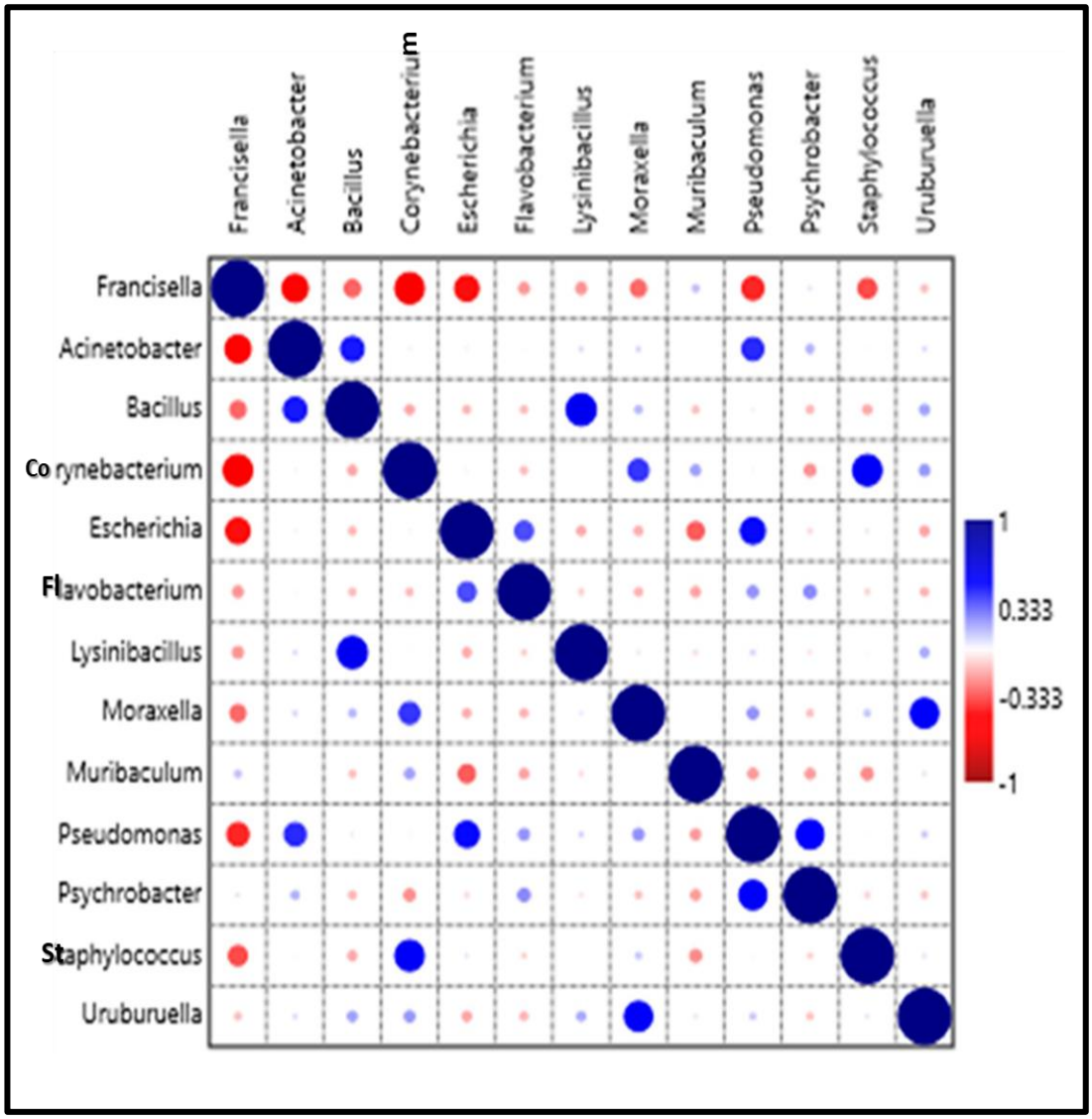

Figure 31: Pearson's correlation coefficients indicating associations between bacterial genera. Showing significantly positive interactions (large dark blue circles) and significantly negative interactions (large red circles).

Table 9: Significant Multiple Regression models that were retained after backward selection.

$Y_{Acinetobacter} = \beta_0 + \beta_{Corynebacterium} + \beta_{Escherichia} + \beta_{Francisella} + \beta_{Lysinibacillus} + \beta_{Moraxella} + \beta_{Pseudomonas} + \beta_{Psychrobacter} + \varepsilon$ ($F_{(7,12)}$=7.02, p = 0.0005)
$Y_{Escherichia} = \beta_0 + \beta_{Acinetobacter} + \beta_{Corynebacterium} + \beta_{Francisella} + \beta_{Lysinibacillus} + \beta_{Moraxella} + \beta_{Pseudomonas} + \beta_{Psychrobacter} + \varepsilon$ ($F_{(7,12)}$=17.26, p<0.0001)
$Y_{Francisella} = \beta_0 + \beta_{Acinetobacter} + \beta_{Escherichia} + \varepsilon$ ($F_{(2,17)}$ = 7.21, p<0.005)

3.4 Tick-Borne Microorganisms and their Prevalence in *Hyalomma* Ticks Collected from Livestock

3.4.1 Tick Identification

All ticks were identified as ixodid (Acari: Ixodidae). Ticks collected from cows, sheep, and goats were identified as *H. anatolicum*, based on morphological features, whereas ticks collected from camels were identified as *H. dromedarii*.

3.4.2 Detection of *Francisella*

Francisella sp. DNA was detected (using PCR by amplifying 16S rRNA gene), in *H. dromedarii* ticks from Abu Dhabi. A positive sample of *Francisella* sp. showed a band of 1151-bp on 1.5% agarose gel stained with ethidium bromide. I found total of thirty samples positive with *Francisella* sp. based on DNA sequences similarity with the records in GenBank (Appendix 19). From the current study, a representative sequence was deposited in the GenBank (accession number MW560059). The representative sequence was 98.59% identical to the *Francisella* sp. endosymbionts of *Amblyomma paulopunctatum* (MN998649.1), *Dermacentor auratus* (JQ764629.1), and *Ornithodoros moubata* (AB001522.1) in GenBank (Appendix 19). A phylogenetic tree was prepared (Figure 32) for *Francisella* sp. of this study with the sequences from the GenBank that were showing the highest similarity to it. The *Francisella* sp. sequence of this study formed a well-defined branch, which was supported by a significant bootstrap value. Furthermore, *Francisella* sp. was not detected in *H. anatolicum* ticks collected from cows, sheep, and goats.

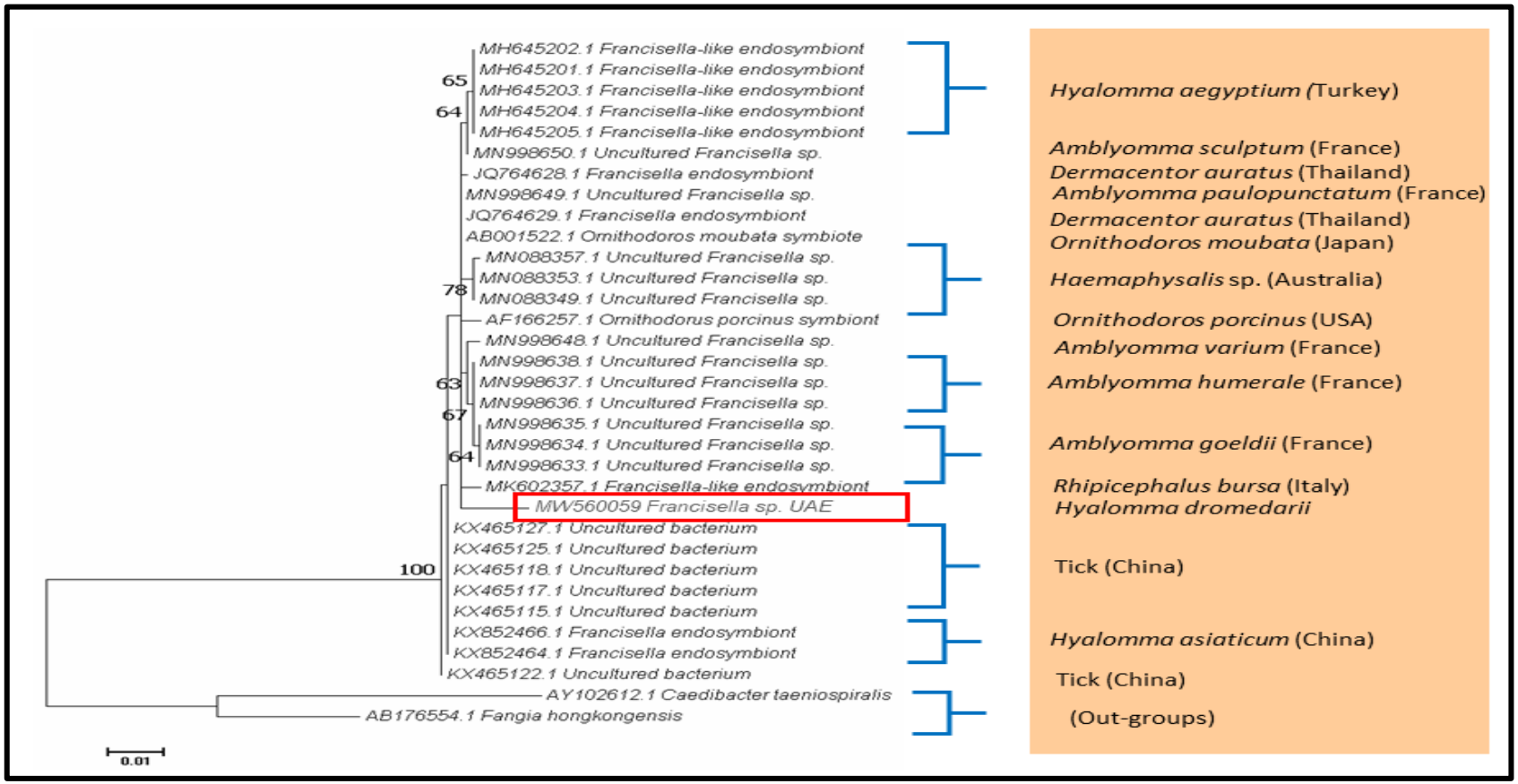

Figure 32: Maximum Likelihood tree based on the 16S rRNA gene showing the phylogenetic relationship of *Francisella* sp.. Detected in *H. dromedarii* from camels in Abu Dhabi, with reference sequences from the GenBank database. *Caedibacter taeniospiralis* and *Fangia hongkongensis* were used as out-groups. Column shows host names.

3.4.3 Detection of *Rickettsia*

Uncultured *Rickettsia* sp. DNA has only been detected in *H. dromedarii* ticks (using nested PCR by amplifying the *omp*A gene) collected from camels in Abu Dhabi. A positive sample of Uncultured *Rickettsia* sp. showed a band of 540-bp on 1.5% agarose gel stained with ethidium bromide. Uncultured *Rickettsia* sp. was identified based on DNA sequences similarity with the records of the *omp*A gene in the GenBank (Appendix 20). From the present study, a representative sequence of Uncultured *Rickettsia* sp. was deposited in the GenBank (accession number MW701398). This sequence was 99.8% identical to the *Candidatus* Rickettsia andeanae detected in *Amblyomma parvum* from Brazil (KY628370.1), *Amblyomma tigrinum* from Brazil (KX434737.1), and *Amblyomma maculatum* from USA (KX158267.1). Also, this sequence of the present study was 99.8% identical to Uncultured *Rickettsia* sp. detected in *Amblyomma parvum* from Brazil (MK522488.1), and *H. dromedarii* from UAE (KF156874.1). A phylogenetic tree (Figure 33) was constructed using highly similar GenBank sequences of *Rickettsia* sp. to this study showed that the UAE sample was in a cluster of *Candidatus* Rickettsia andeanae and Uncultured *Rickettsia* sp., supported by a high bootstrap value. Uncultured *Rickettsia* sp. was not detected in *H. dromedarii* and *H. anatolicum* collected from Dubai and Sharjah.

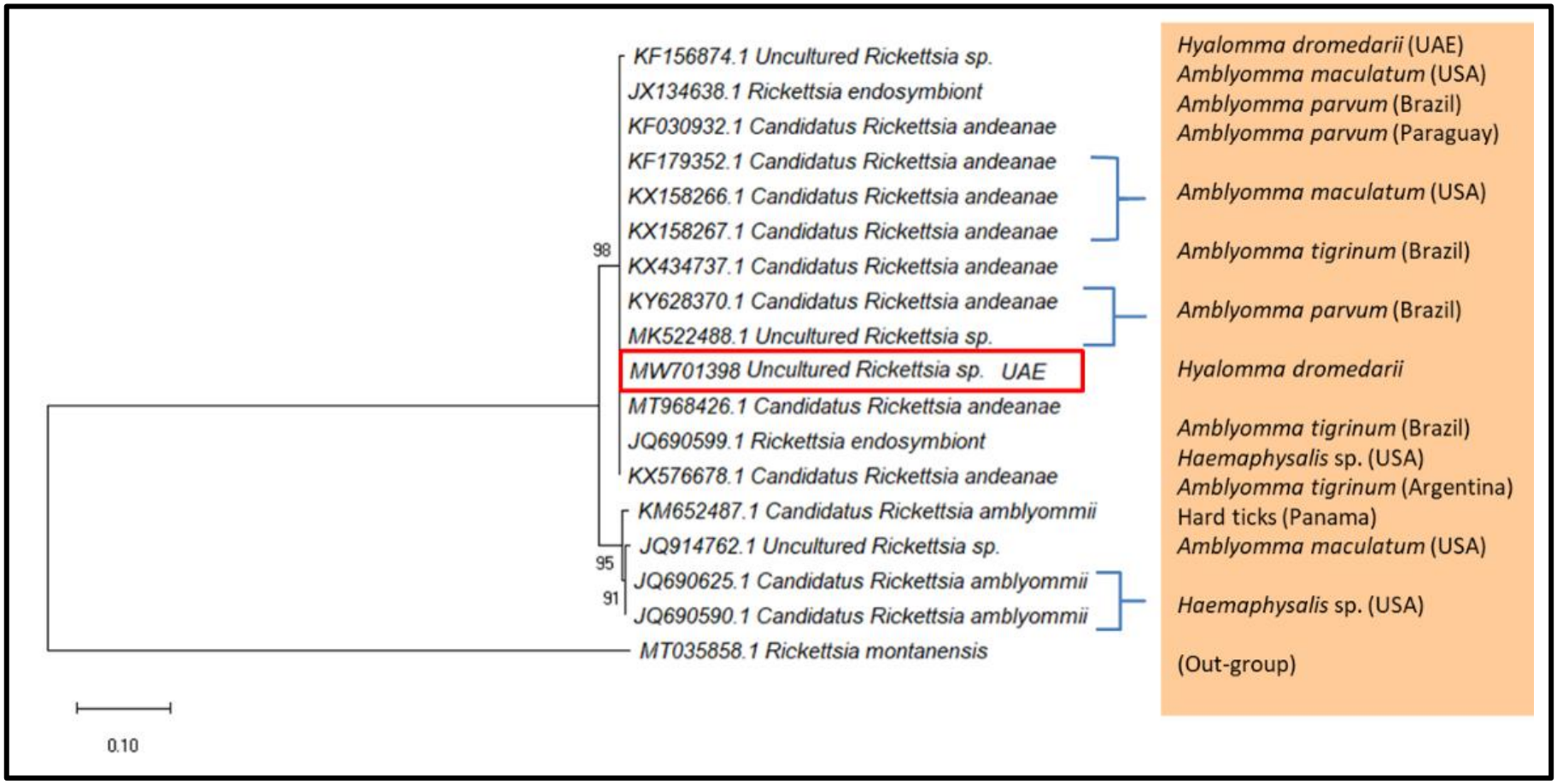

Figure 33: Maximum Likelihood tree based on the *omp*A gene showing the phylogenetic relationship of *Rickettsia* sp.. Detected in *H. dromedarii* from camels in Abu Dhabi, with reference sequences from the GenBank database. *Rickettsia montanensis* was used as an out-group. Column shows host names.

3.4.4 Detection of Piroplasmids

Two piroplasmids were detected in the present study, *T. annulata* and *T. ovis*. The DNA of both piroplasmids were detected (using PCR by amplifying the ssrRNA gene), in *H. anatolicum* ticks collected from livestock in Sharjah. *Theileria annulata* was detected in *H. anatolicum* ticks collected from cows whereas *T. ovis* was detected in ticks collected from goats. A positive sample of *T. annulata* showed a strong band of 560-bp on 1.5% agarose gel stained with ethidium bromide while a positive sample of *T. ovis* showed a light band of 560-bp on agarose gel. Fragments of the ssrRNA gene were identified as *T. annulata* and *T. ovis* based on DNA sequences similarity with the records in the GenBank (Appendix 21 and 22). A sequence of *T. annulata* was deposited in the GenBank with the accession number MW537791 and the one of *T. ovis* with accession number MW559557. Sequences of *T. annulata* were 99.62% identical to the *T. annulata* detected in cattle, *Bos taurus* (MT341858.1), ruminants (MT318160.1), and ticks (MN227669.1) (Appendix 17). Through phylogenetic analysis of *T. annulata* (Figure 34), which was performed by using GenBank sequences of high similarity to *T. annulata*, the UAE sequences appeared in a cluster of *T. annulata* samples detected from Italy, Pakistan, and Egypt. Similarly, sequences of *T. ovis* from the UAE were 99.81% identical to the *T. ovis* detected in cattle, *Bos grunniens* (MN394810.1) from China, Tibetan sheep (MN394809.1) from China, and sheep from Iraq (MN712508.1), and Egypt (MN625886.1). The phylogenetic tree (Figure 35) showed that the UAE species was in a cluster of *T. ovis* samples detected from Iraq and Egypt. Finally, piroplasmids were not detected in *H. dromedarii* and *H. anatolicum* from Abu Dhabi and Dubai.

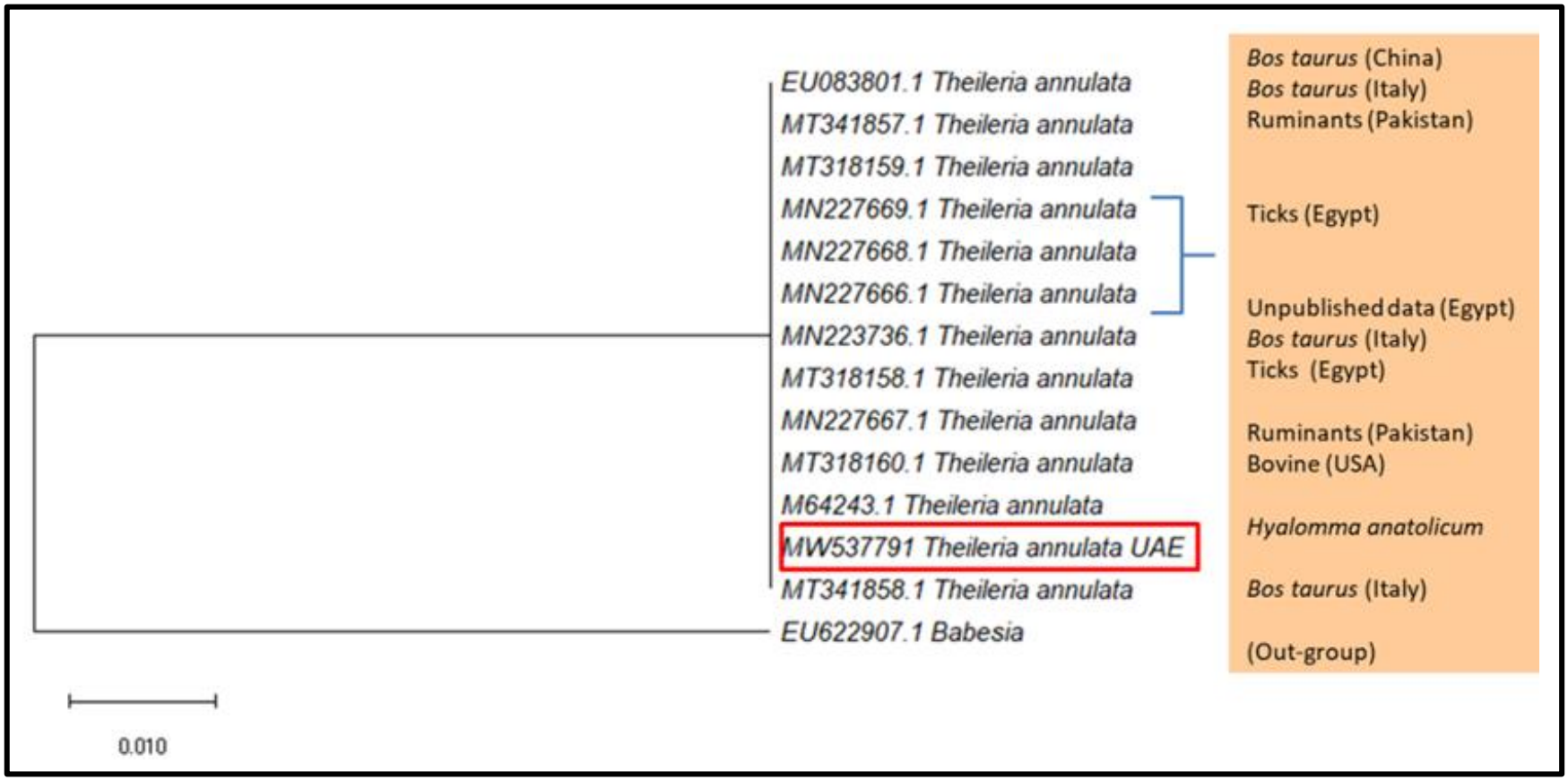

Figure 34: Maximum Likelihood tree based on the ssrRNA gene showing the phylogenetic relationship of *T. annulate.* Detected in *H. anatolicum* collected from cows in Sharjah, with reference sequences from the GenBank database. *Babesia major* was used as an out-group. Column shows host names.

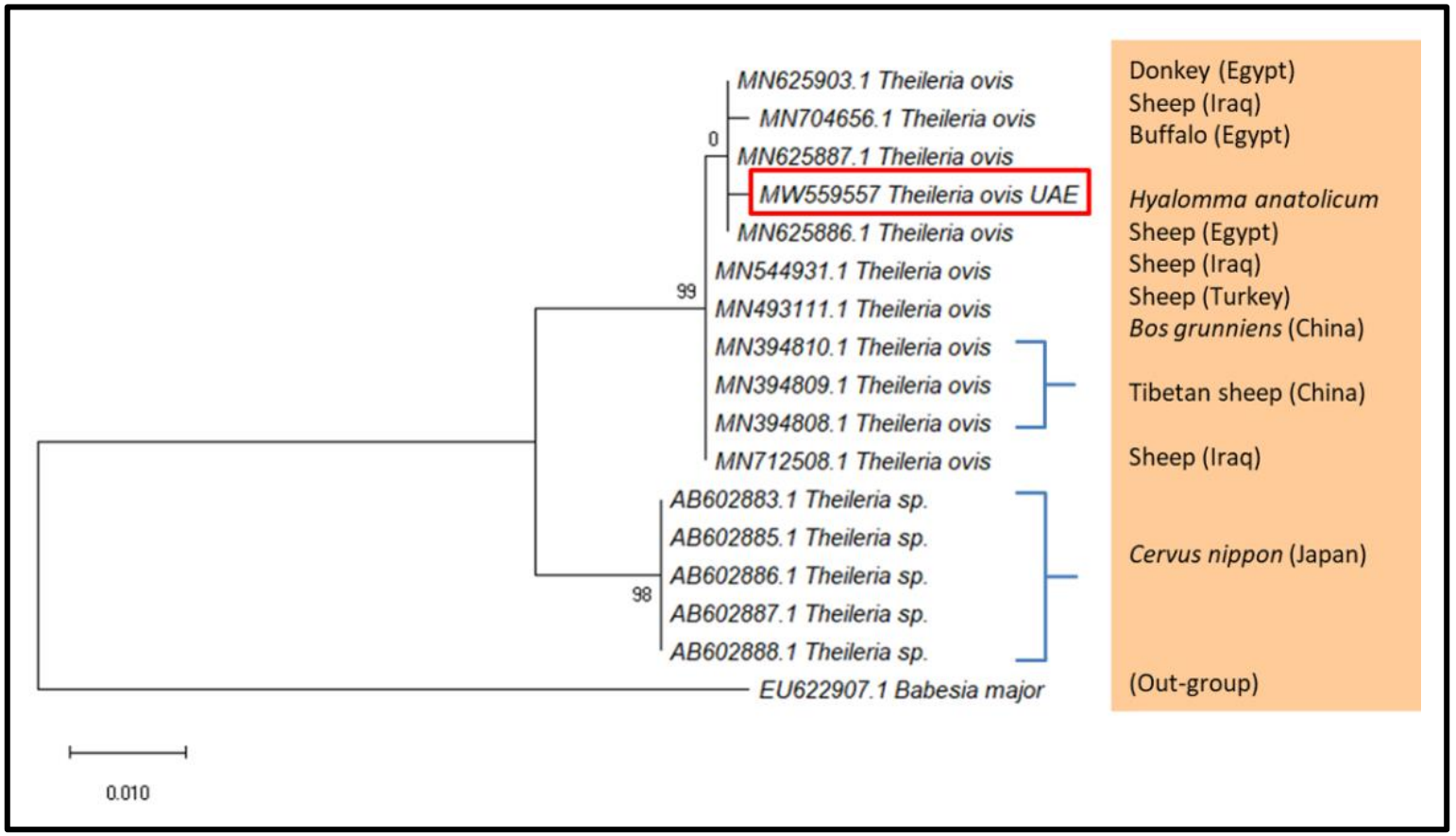

Figure 35: Maximum Likelihood tree based on the ssrRNA gene showing the phylogenetic relationship of *T. ovis.* Detected in *H. anatolicum* collected from goats in Sharjah, with reference sequences from the GenBank database. *Babesia major* was used as an out-group. Column shows host names.

3.4.5 PCR-based Infection Rates of Tick-borne Microbes

Tick-borne microorganisms were detected using PCR in 39 out of 562 DNA samples extracted from ticks, with an overall infection rate of 6.94%. In Abu Dhabi, *H. dromedarii* collected from camels were infected with the *Francisella* sp. (5.81%) and Uncultured *Rickettsia* sp. (1.36%). However, these microbes were not detected in tick samples from Dubai and Sharjah. In Sharjah, *H. anatolicum* ticks collected from cows were found positive with *T. annulata* (4.55%) whereas *H. anatolicum* collected from goats in Sharjah were found positive with *T. ovis* (10%). No microorganism was detected in the ticks collected from sheep in Abu Dhabi, Dubai, and Sharjah. Ticks collected from camels had a higher rate of infection compared with ticks collected from cows, sheep, and goats (Table 10). In the current study, no Babesia species were detected in ticks collected from the sampled animals. In addition, no microbe was detected in ticks collected from camels, cows, sheep, and goats in Dubai.

Table 10: Infection rate of bacteria and piroplasmids in ticks collected from livestock in UAE.

Hosts	**Tick species**	**Number of animals**	**Number of samples**	**Number of positive samples (total infection rate)**			
				Francisella sp.	*Rickettsia* sp.	*T. annulata*	*T. ovis*
Camel	*H. dromedarii*	518	516	30 (5.81)	7 (1.36)	0 (0)	0 (0)
Sheep *	*H. anatolicum*	70	14	0 (0)	0 (0)	0 (0)	0 (0)
Goat *	*H. anatolicum*	34	10	0 (0)	0 (0)	0 (0)	1 (10)
Cow*	*H. anatolicum*	26	22	0 (0)	0 (0)	1(4.55)	0 (0)
Total		648	562	30	7	1	1

*ticks were pooled into given numbers of samples.

Chapter 4: Discussion

4.1 Prevalence, Distribution, and Molecular Record of Hard Ticks from Livestock

4.1.1 Tick Identification

The study aimed to identify ticks collected from livestock and measure their prevalence on camels, cows, sheep, and goats. Tick identification is a significant step to develop ticks and tick-borne diseases management strategies. There are limited studies on ticks in the UAE. Consequently, more studies are required to generate morphological and genetic databases for the UAE tick species. In this study, the presence of four tick species from livestock, *H. dromedarii, H. anatolicum, A. lepidum,* and *R. sanguineus* was confirmed through morphological and molecular characterization of the ticks. Additionally, the current work provides the first molecular record of *H. anatolicum, A. lepidum* and *R. sanguineus* from the UAE along with the morphological description. *Rhipicephalus sanguineus* has often been reported from cattle in Iraq and Pakistan (Atif, Khan, Iqbal, Ali, & Ullah, 2012; Hasson, 2012; Sajid, Iqbal, Khan, Muhammad, & Khan, 2009) and are presumably associated with dogs near farms or animal markets. The four ticks species reported in this study can be distinguished from each other very easily; however, some notable similarities might cause taxonomic confusion, underscoring the need for molecular identification. Some of the distinctive features of *R. sanguineus* collected and identified in this study include the presence of posterior grooves as comma-shaped which are three in numbers, and adanal plates were curved. In addition, the subadanal plates were absent (McCarthy, 1967; Walker et al., 2003) as compared to other *Rhipicephalus* species. *Hyalomma dromedarii* was larger than *H. anatolicum* and the sub-anal plates were aligned outside

the adanal plates in male ticks (Walker et al., 2003). *Hyalomma anatolicum* was reddish-brown in color and sub-anal shields situated on the axis of the adanals (Apanaskevich, 2003; Walker et al., 2003). *Amblyomma lepidum* identified in the current work was a large and ornamented tick with pink to orange enamels and long mouthparts. The posteromedian strip was narrow which separates it from *Amblyomma gemma* (Robinson, 1962; Walker et al., 2003) in which the posteromedian stripe is broad (Walker et al., 2003). Although diagnostic morphological characters are used to identify ticks to species level, the quality, and level of engorgement of ticks could pose challenges in identification. Thus, molecular methods could improve or confirm morphological identification, especially for species that are difficult to identify. There have been previous records of *H. dromedarii*, *H. anatolicum, A. lepidum,* and *R. sanguineus* infestations in neighboring countries, present study findings shed light on the need for conducting joint projects on tick species that are common in countries sharing borders. Further studies are also required to document the occurrence of more tick species in the UAE and to better understand their possible role in the epidemiology of tick-borne diseases in the desert ecosystem.

4.1.2 Tick Prevalence

No significant difference was found in tick prevalence between male and female hosts except goats, where more females were infested compared to males. However, previously males (cattle) were found more infested (63.4%) than females (60.9%) in Nigeria (Musa et al., 2014). Some studies recorded that the female cattle infestation rate was slightly higher (46.5%) than males (45%) (Ghafar, Gasser, Rashid, Ghafoor, & Jabbar, 2020). In sheep, females were reported heavily infested (100%) tick prevalence as compared to male sheep in Cameroon (Malla, Payne, & Cedric,

2021). The selection of male and female hosts could be related to host behavior or odors and this observation requires further study to determine why, in some hosts, ticks show a preference for a particular sex.

4.1.2.1 *Hyalomma Dromedarii* Prevalence

Camels were heavily infested (94.3%) with *H. dromedarii* compared to all animals and these results concurred with our previous studies in the UAE (Al-Deeb & Muzaffar, 2020). *Hyalomma dromedarii* is known to harbor a variety of microbes with some serious tick-borne bacterial (Alreshidi, 2020; Elbir et al., 2019) and protozoan pathogens (Al-Deeb et al., 2015) in the MENA region including the UAE.

4.1.2.2 *Hyalomma Anatolicum* Prevalence

Hyalomma anatolicum was found on all hosts in the sampling areas of the UAE, suggesting that the same tick species in the three emirates was probably associated with unrestricted livestock trade and the movement of animals between emirates (Damian, Damas, Wensman, & Berg, 2021). Tick prevalence, mean intensity and mean abundance varied among the four animal hosts (camel, cows, sheep, and goats) and this is most likely due to host preference in this species. Furthermore, *H. anatolicum* was the most prevalent tick species in cows (32.8%) as compared to other tick species. The present study results are similar to previous studies which reported a high prevalence of *H. anatolicum* in cattle (63.1%) (Aktas, Dumanli, & Angin, 2004) and livestock (38.83%) including cattle, sheep, and goats (Biglari et al., 2018) as compared to other tick species. In addition, another study reported the highest prevalence of *H. anatolicum* in cattle and sheep (Hasson, 2012). *Hyalomma anatolicum* in sheep was also found as the most prevalent tick species (14.4%)

followed by goats (9.9%). In contrast, in Cameroon (Malla et al., 2021), *Boophilus geygei* was reported as the most dominant species in goats. Such differences in the results may be due to different geographic areas, the genetic makeup of host as well as tick species distribution in various regions. In the current study, the prevalence of *H. anatolicum* was found on all hosts except camels and this could be attributed to farming conditions and resistance of this tick species to acaricides. This tick is the competent vector of CCHF (Choubdar, Karimian, Koosha, & Oshaghi, 2021) and poses a serious threat to livestock as well as humans who may get exposed to tick bites in the livestock industry. In the UAE, this species was found as a carrier of CCHF (Khan et al., 1997), *T. annulata* and *Theileria ovis*.

4.1.2.3 *Rhipicephalus Sanguineus* Prevalence

Rhipicephalus sanguineus or the kennel tick was identified from cattle tick samples in Sharjah with very low prevalence (0.8%) and the results of the present study are comparable to records from Iraq where *R. sanguineus* prevalence was 0.09% from cattle and sheep during the investigation of monthly tick distribution (Hasson, 2012). In addition, *R. sanguineus* was recorded with low prevalence (7.52%) on cattle (Atif et al., 2012) and bovine species (13%) (Sajid et al., 2009) from Pakistan. The higher prevalence of this tick on livestock could be associated with a generally higher abundance of stray dogs in those regions (Atif et al., 2012; Sajid et al., 2009). In the UAE, dogs are not present as feral or stray populations and possibly this can prevent the buildup of these ticks in such populations.

4.1.2.4 *Amblyomma Lepidum* Prevalence

Amblyomma lepidum was found only on cattle with low prevalence (0.8%) from Dubai. This is a common tick species on livestock in Sudan, Ethiopia, Somalia, Uganda, Kenya, and Tanzania (Walker et al., 2003). It has the potential to cause bovine theileriosis and heart-water in livestock by transmitting bacterial and protozoan pathogens (Dinkisa, 2018; Walker et al., 2003). Though it was found in the lowest prevalence, it is a very important species that could pose a risk to livestock if allowed to build up in numbers.

4.1.3 Future Research

Although the current work has reported the presence and distribution of four tick species, a well-documented distribution of ticks in the entire UAE is still needed. The presence of the aforementioned four tick species is of medical relevance because some of them are known vectors of diseases such as CCHF and spotted fever group (SFG) *Rickettsia*. Further studies are required because pathogens carried by these ticks can infect both livestock and people and consequently identification and surveillance of tick species is crucial to avoid or manage future tick-borne diseases incidences in the UAE.

4.2 Seasonal Population Dynamics of *Hyalomma dromedarii* on Camels

Camel tick, *H. dromedarii* has economic significance because this species is an ectoparasite of domestic animals and a competent vector of a variety of tick-borne pathogens. *Hyalomma* tick species can survive in inclement environments and is affected by extreme temperature, humidity, and host condition (Hoogstraal, 1956). These survival factors play a huge role in allowing these ticks to thrive in an

environment where few or no others species can survive. The United Arab Emirates is the home of a large population of camels, therefore, this tick species may create a problem in the UAE. In the MENA region, *H. dromedarii* is one of their major tick species.

In the present study, fluctuation in numbers of *H. dromedarii* was assessed during 12 months under common camel breeding and management practices in the study area. Results showed that *H. dromedarii* ticks were found on the camels throughout the year despite a monthly application of an acaricide by the farm owner. This study shows that infested camels are under continuous parasitic pressure throughout the year and consequently they may suffer from blood loss and probable tick-borne disease infections. Furthermore, the constant presence of *H. dromedarii* ticks on camels even after application of an acaricide may indicate that either they developed acaricide resistance as a result of repeated exposure to the same chemical or there is an inadequate acaricide application. In both scenarios, further studies are required for investigations. In the present study, the peak activity of adult *H. dromedarii* was observed in June 2019, which is consistent with the findings of Gharbi, Ettaı, et al. (2013) from Tunisia and Benchikh-Elfegoun et al. (2007) from Algeria (Benchikh-Elfegoun, Benakhla, Bentounsi, Bouattour, & Piarroux, 2007) where a peak activity of adult *Hyalomma scupense* on cattle was reported in June. The peak activity of *H. dromedarii* in June could be due to the change in farming practices in the summer months. In these months, camels sit in shades, and more water is provided to keep them hydrated, this could lead to more tick infestation due to aggregation behavior of hosts and high humidity. In addition, *H. dromedarii* remained active throughout the year, confirming previous results in Egypt and Tunisia (Gharbi, Moussi, et al., 2013; Van Straten & Jongejan, 1993). Furthermore, *H. dromedarii* ticks

were reported with high prevalence (94.33%) in the present study, which is similar to records from Egypt (95.6%) (Van Straten & Jongejan, 1993) and UAE (98%) in 2010-2011 (Al-Deeb & Muzaffar, 2020). *Hyalomma* ticks lifecycle may be greatly prolonged under unfavorable climatic conditions, or shortened under optimum conditions (Hoogstraal, 1956). Previously, it was found in some studies that total monthly tick burdens were positively correlated with several abiotic factors, for instance, monthly mean minimum temperature, monthly mean maximum temperature, and the number of sunny days while negatively correlated with relative humidity (Gharbi, Moussi, et al., 2013). Pearson's correlation test showed that there is a weak positive correlation ($r = 0.36$) between tick loads and monthly average temperature and also there is a moderate negative correlation ($r = -0.54$) between tick loads and relative humidity. Based on the p values ($p = 0.2439$, $p = 0.0694$; respectively) of the test, the correlation is not significant ($p > 0.05$). However, results showed that there was a significant difference in tick burden between the months. The relationship between abiotic factors and tick burdens is known for different tick species although this has not been demonstrated in arid areas where high temperatures and low relative humidity significantly influence tick dynamics. Though ticks were present during all months of the year, these abiotic factors may influence tick populations, but do not reduce their activity (Gharbi, Moussi, et al., 2013).

No larvae were found on camels during the 12 month sampling period. Moreover, nymphs were found in very low numbers and it is assumed that *H. dromedarii* in the UAE has a two-host lifecycle, and probably the larval and nymphal stages feed on alternate hosts for instance birds, reptiles, or small mammals. The host availability, host size, host density as well as microclimatic factors in the environment of the host (especially in captivity) may alter the natural life cycle of *Hyalomma* ticks

(Hoogstraal, 1956). Immature stages of these ticks often feed on birds, rodents, and hares that are important reservoirs of pathogens especially viruses and rickettsiae (Hoogstraal, 1956). *Hyalomma dromedarii* can behave like a three, two, or one-host species (Hoogstraal, 1956; Walker et al., 2003) and it is believed that the two-host life cycle is the most common for this species (Hoogstraal et al., 1981). *Hyalomma dromedarii* fed on rabbits under laboratory conditions behaved as a two-host tick (Alahmed & Kheir, 2003). However, it was found that *H. dromedarii* was usually a three-host tick, and became a two-host tick when density on the host was high (Ouhelli, 1994). Camels are the principal hosts of the adult ticks, which also parasitize other domestic ungulates, such as cattle, sheep, buffaloes, horses, donkeys, and goats (Apanaskevich et al., 2008; Hoogstraal, 1956; Hoogstraal et al., 1981; Pomerantzev, 1959). The occasional records of adults from dogs, hyenas, ostrich, lizards, and humans were also reported (Apanaskevich et al., 2008; Hoogstraal et al., 1981). In addition, *H. dromedarii* is the only species of the genus *Hyalomma* in which the immature stages can use both small and large mammals as hosts. Nymphs and larvae both may use the same species of large animals (especially camels) as the adults (Diab et al., 2006). However, the immature stages can also parasitize rodents, leporids, and hedgehogs, as well as birds and reptiles (Apanaskevich et al., 2008; Hoogstraal, 1956; Hoogstraal et al., 1964). This was confirmed in one study on ticks of wildlife from Saudi Arabia where seventeen *Hyalomma* nymphs were collected from Arabian spiny mouse (*Acomys dimidiatus*) and nine nymphs were collected from Sundevall's jird (*Meriones crassus*) which were later molted to *H. dromedarii* and *H. impeltatum* (Al-Khalifa, Diab, Al-Asgah, Hussein, & Khalil, 2006). Based on the design and available resources, the focus of the current study was to collect ticks only from camels (the main host). Therefore, results shed light on a major part of the life cycle of *H.*

dromedarii and specifically the damaging stages (adult and nymphs), which cause economic damage to the camel industry as a result of blood loss and disease transmission.

Males tick population was dominant throughout the year on camels as compared to females. Previously, in Saudi Arabia, males were recorded in larger numbers as compared to females of *H. dromedarii* during a study of tick infestation on livestock (Diab et al., 2006). The engorged female of *H. dromedarii* probably burrows a few centimeters below the ground surface to find favorable microhabitats for egg deposition and to protect the eggs and emerging larvae against high temperature and low humidity during the dry season (Alahmed & Kheir, 2003). This is also consistent with our assumption that larvae and nymphs feed on small mammals since these burrows are often occupied by rodents or insectivores. Future studies in the UAE need to focus on these two stages (egg and larvae) of *H. dromedarii* to understand the full life cycle and to identify secondary hosts. Moreover, the seasonal production of eggs (laying locations and dates) needs to be investigated for tick management strategies. This study results also revealed that engorged females were encountered in almost every sampling date throughout the year. However, in December 2019, some live engorged females were kept in plastic vials (ventilated) in the laboratory until they laid eggs. Therefore, it is assumed that eggs are laid throughout the year given the high abundance of engorged ticks year-round.

In Al-Ain and many other places in the UAE, it is very common that camels are kept with sheep and goats in the same area on the farm and are only separated by a small net fence. This was the situation of the farm on which the current study was conducted. In the desert , occasionally, camels are left to graze freely for grooming and eating different desert plants. When *H. dromedarii* and *H. impeltatum* ticks are

highly prevalent on camels and these animals subsequently are allowed to graze together with a large number of sheep, there is a risk of ticks finding alternative hosts and becoming established (Jacquiet et al., 1994). Previously, it was reported that in areas where camels and cattle coexist, *H. dromedarii* might act as a vector of *T. annulata* (Jacquiet et al., 1994). In Tunisia, where camels shared common pastures with cattle (Hniche, 2006), there was a frequent infestation of ticks originating from camels to other livestock such as cattle. This resulted in a higher tick burden and possibly an altered tick distribution on cattle. For example, *H. dromedarii* was the dominant tick (82.09%) infesting cattle, followed by *H. impeltatum*, *H. marginatum, H. scupense*, and only one individual of *Rhipicephalus sanguineus* (Hniche, 2006). *Hyalomma dromedarii* is a vector of many viral, bacterial and protozoan pathogens. Many genera of viruses have been isolated from *H. dromedarii,* namely Crimean-Congo Hemorrhagic Fever Virus, Kadam Virus, Dera Ghazi Khan Virus, and Dhori Virus. Additionally, this tick species serves as a vector for bacterial pathogen transmission such as *C. burnetti*, *T. camelensis*, and *T. annulata* (Hoogstraal et al., 1981). Therefore, it is essential to characterize the pathogenic organisms associated with ticks, given the high densities of camels and ticks in the region. In the UAE, this is 1st study on *Hyalomma dromedarii* population dynamics and could help in tick management strategies, by providing knowledge of tick population fluctuation in different months.

4.3 Bacterial Communities' Composition and Diversity in Camel Tick, *Hyalomma dromedarii* Using Next-Generation Sequencing

Camel ticks can carry and transmit potential pathogens (Al-Deeb et al., 2015; Bowman & Nuttall, 2008; Bratton & Corey, 2005; de la Fuente et al., 2008; Gayle & Ringdahl, 2001; Jongejan & Uilenberg, 2004). Microbial diversity in ticks plays a

significant role in pathogen transmission, vector competence (Burgdorfer, Brinton, & Hughes, 1973; Vilcins, Old, & Deane, 2009), and tick reproductive fitness (Zhong, Jasinskas, & Barbour, 2007). Tick-borne pathogens can significantly decrease the production of camel milk and meat and may affect the racing breeds. We found a diverse array of pathogens in *H. dromedarii* ticks, highlighting the reservoir potential of this tick species for significant pathogens.

4.3.1 Microbial Diversity

4.3.1.1 Relative Abundance of Bacterial Phyla

The patterns of bacterial phyla in the current study were consistent with findings of Elbir et al. (2019) where the *Proteobacteria* was the most abundant followed by *Firmicutes* and *Actinobacteria*. In addition, results were consistent with the results of Thapa et al. (2019), who also found *Proteobacteria* with the highest relative abundance across all baseline *Ixodes scapularis* males and female ticks under different temperatures in the USA. In similar studies, the bacterial phylum *Proteobacteria* was reported to be the most dominant (89%) in the microbiota of whole *Amblyomma tuberculatum* ticks infesting the gopher tortoise (Budachetri, Gaillard, Williams, & Mukherjee, 2016) and (83.39%) in bacterial communities associated with *A. maculatum* (Budachetri et al., 2014). Moreover, *Proteobacteria* was found to be the dominant phylum followed by *Actinobacteria* and *Firmicutes* in *Ixodes ricinus* ticks on sheep in Northern Italy (Carpi et al., 2011). Overall, these findings indicate that *Proteobacteria* is a very common phylum, which exists in different tick species on different animal hosts.

4.3.1.2 Relative Abundance of Bacterial Classes

Bacterial classes (16 classes in 2010 and 14 in 2019) observed in this study were comparable to Khoo et al. (2016), where the taxonomic composition of tick samples indicated that the abundant bacterial class was *Gammaproteobacteria* along with *Alphaproteobacteria*, *Actinobacteria*, *Bacilli* and *Deltaproteobacteria*, which represented 80% to 99% of the population in each of the samples. However, Karim et al. (2017) documented predominantly only six classes namely *Bacilli, Gammaproteobacteria, Betaproteobacteria, Clostridia, Alphaproteobacteria*, and *Actinobacteria,* after profiling the bacteria sampled from tick species collected from various livestock. In another study, the bacterial DNA sequences of *Alphaproteobacteria* and *Gammaproteobacteria* types were abundant in *Ixodes persulcatus, Ixodes pavlovskyi,* and *Dermacentor reticulatus* samples with 30.2% and 60.8% average occurrence, respectively (Kurilshikov et al., 2015). Generally, these studies in addition to the current study show that the above-mentioned common bacterial classes have a wide geographical distribution occurring in ticks from the UAE, Malaysia, Pakistan, and Russia.

4.3.1.3 Relative Abundance of Bacterial Families

Patterns of abundance of bacterial families in this study differed from the findings of Karim et al. (2017) who found *Oxalobacteraceae, Staphylococcaceae, Clostridiaceae, Enterobacteriaceae, Coxiellaceae, Rickettsiaceae, Streptococcaceae,* and *Lactobacillaceae* as the predominant microbial families in tick samples that were not *H. dromedarii*. Some of this variation can be explained in light of quantitative and qualitative differences in microbial communities between hosts (Khoo et al., 2016).

Enterobacteriaceae was the most abundant bacterial family in *R. microplus* ticks collected from cattle whereas *Rickettsiaceae*, *Oxalobacteraceae*, and *Micrococcaceae* were abundant in the *R. turanicus* ticks infesting goats (Karim et al., 2017). Furthermore, the results of this study differ from Ravi et al., 2019 who reported four bacterial families: *Coxiellaceae, Francisellaceae, Rickettsiaceae*, and *Anaplasmataceae* in *H. dromedarii*, *R. sanguineus*, and *Haemaphysalis concinna*. The differences among families could be attributed to host-specific factors. The existence of common families among different tick hosts may indicate that these bacterial families are generalists and may not require a very specific host internal environment. Results of the present study are partly consistent with the findings of Kurilshikov et al. (2015) who found *Francisellaceae* as the most abundant family in *Dermacentor reticulatus* ticks and the *Moraxellaceae* in *Ixodes persulcatus*. In addition, Budachetri et al. (2014) reported that *Francisellaceae* and *Enterobacteriaceae* were the prevalent bacterial families in *A. maculatum* ticks. Based on these findings, *Francisellaceae* and *Enterobacteriaceae* coexist in *H. dromedarii* and *A. maculatum* suggesting that they thrive under similar conditions and microbial interactions inside the host. In general, the composition of microbial families can be affected by external and stochastic factors, which contribute to producing high or low diversity inside each individual tick. Although in this study, the diversity in microbial families within *H. dromedarii* was explained, the environmental and host-related factors, which might shape this complex microbial ecosystem, were not focused. Further, it is assumed that certain interactions among microorganisms inside *H. dromedarii* result in the dominance of some families over the others.

4.3.1.4 Relative Abundance of Bacterial Genera

The use of the *16S* rRNA gene for the identification of a broad range of clinically relevant bacterial pathogens is a good tool to assess microbial communities. However, short-read sequencing platforms which target different regions of 16S rRNA do not provide good taxonomic resolution when compared to sequencing the entire gene (Johnson et al., 2019). This implies that 16S rRNA gene-based identification is reliable up to the genus-level. In addition, getting full-length or near full-length 16S sequences is crucial for making confident genus level taxonomic placements. Therefore, the genus level identifications presented in the current study are provided as preliminary baseline data, which may require further confirmation. Results indicated that *Acinetobacter* and *Corynebacterium* were the two most abundant genera detected in the microbiota of *H. dromedarii* from all locations with high sequence ratios among a total of 31 genera in 2010 samples with more than 1% or some with 1% sequence reads at different locations. Other abundant genera included *Escherichia, Proteus, Staphylococcus, Bacillus, Flavobacterium*, and *Stenotrophomonas*. However, the *Francisella* was the dominant genus (99.1%) in 2019 among all 15 abundant genera. *Bacillus, Escherichia, Siccibacter,* and *Acinetobacter* were the other predominant genera at different locations in 2019 samples. The genus *Francisella* was confirmed previously in *H. dromedarii* ticks from Palestine (Ravi et al., 2019), Saudi Arabia (Elbir et al., 2019), and UAE.

4.3.2 Associations between Bacterial Genera

Significant associations between *Acinetobacter*, *Escherichia* and *Francisella,* and between these three genera and several other genera were found. Little is known

about *Francisella* and their associations with ticks. It seems that many diverse bacterial genera co-exist with tick-borne pathogens (Bonnet, Binetruy, Hernández-jarguín, & Duron, 2017) and endosymbiotic forms could increase the colonization potential of pathogenic forms (Aivelo, Norberg, & Tschirren, 2019). *Francisella* appears to occur in many ticks species, most commonly in mutualistic forms (Bonnet et al., 2017). However, phylogenetic similarities between mutualistic and pathogenic *Francisella* suggest periodic and perhaps even frequent shifts from non-pathogenic forms (Bonnet et al., 2017; Narasimhan & Fikrig, 2015). Nonetheless, the co-occurrence of non-pathogenic and pathogenic bacteria may not always result in genetic transformations (Greay et al., 2018), suggesting that multiple factors could influence pathogenicity in tick microbiota. Moreover, the constant occurrence of the *Francisella* indicates a systemic association between arthropods and this bacterial genus. The current study shows negative associations between *Francisella*, *Acinetobacter*, and *Escherichia* which may indicate possible suppressive effects of the former on the latter two genera. On the other hand, the positive association between *Acinetobacter* and a broad range of bacterial genera also deserves further consideration. Many species of *Acinetobacter* are known to be pathogenic, while others are considered commensal and even part of the normal flora of animals (van der Kolk, Endimiani, Graubner, Gerber, & Perreten, 2019).

Francisella was reported in 2010 and 2019, however, was found with the highest abundance (99%) in 2019. This finding is consistent with the overall change in bacterial communities experienced in 2019, with the general rise in *Francisella*. If future studies confirm the presence of the pathogenic species of the genus *Francisella* in the UAE, this could be a potential emerging disease pathogen in the country and may affect the people who are closely working with the camels such as workers at

farms and slaughterhouses, veterinary hospitals and research centers. Therefore, the above-mentioned bacterial genera need further confirmation through PCR-based approaches.

4.4 Tick-Borne Microorganisms and their Prevalence in *Hyalomma* Ticks Collected from Livestock

Disease detection is the most important step in programs that safeguard human or animal health (Cunningham et al., 2017). Early detection of pathogens is crucial in curtailing their spread and subsequently in reducing the risk of exposure and possible outbreaks (Cunningham et al., 2017). This study data revealed the presence of two bacterial and two piroplasmid species in local ticks infesting several animal hosts. *Hyalomma* ticks contained four microbes, namely, *Francisella* sp., *Rickettsia* sp., *T. annulata*, and *T. ovis*.

4.4.1 Detection of *Francisella* and its Prevalence

High infection (5.81%) of *Francisella* sp. was found in the *H. dromedarii* ticks collected from camels in Abu Dhabi. These findings are comparable to infection rates of *Francisella* spp. (4.7%) in *H. dromedarii* from camels in Egypt (Ghoneim et al., 2017). However, the molecular identification of *Francisella* sp. in the present study aligned with *Francisella*–like endosymbionts rather than with any known pathogenic *Francisella* spp., which agrees with reports from Egypt (Ghoneim et al., 2017). In addition, the *Francisella* sp. of this study was closely related to *Francisella* endosymbiont recorded in *A. paulopunctatum* (MN998649.1) (Binetruy et al., 2020), *D. auratus* (JQ764629.1), and *O. moubata* (AB001522.1). It should be pointed out that previously the genus *Francisella* has been reported with a very high prevalence (99.1%) in *H. dromedarii* ticks from camels in the UAE. Therefore, future studies in

the UAE should use species-specific primers to determine whether *Francisella* sp. is pathogenic or a *Francisella*–like endosymbiont.

4.4.2 Detection of *Rickettsia* and its Prevalence

Rickettsia sp. in *H. dromedarii* ticks collected from camels in Abu Dhabi in this study was closely related to *Cand.* R. andeanae recorded in *Amblyomma* ticks from Brazil and the USA. In addition, it was also 99.8% identical to uncultured *Rickettsia* sp. detected previously in *Amblyomma* and *Hyalomma* ticks from Brazil and the UAE, respectively. Many *Rickettsia* species exist in ticks, although their pathogenicity has not been determined (Piotrowski & Rymaszewska, 2020). Since ticks may serve as vectors as well as reservoirs of rickettsiae in nature, this constitutes a risk factor for *Rickettsia* transmission in livestock and humans (Piotrowski & Rymaszewska, 2020). Spotted fever group (SFG) rickettsiae have at least 30 distinct genotypes in 15 species currently recognized as pathogens in humans (Delgado-de la Mora et al., 2019; Paddock et al., 2004). Recently, *R. parkeri* (that causes spotted fever rickettsiosis in humans) and *Cand.* R. andeanae were reported widely in several tick species across wide geographic regions (Delgado-de la Mora et al., 2019; Lee et al., 2018; Noden, Roselli, & Loss, 2020). Although *Cand.* R. andeanae do not seem to cause human infections (Paddock et al., 2015), the high prevalence of *Cand.* R. andeanae in ticks might interfere with the development of *R. parkeri* and limit its distribution (Noden et al., 2020). Therefore, there is a need to better quantify the dynamics among various spotted fever group *Rickettsia* species within their tick hosts to determine how their interactions contribute towards the epidemiology of rickettsioses in human and animal hosts (Paddock et al., 2015). Furthermore, ticks are known to engage in symbiotic associations with at least 10 different genera of maternally inherited bacteria (Duron

et al., 2017). Ticks develop close interactions with beneficial symbionts that provide essential B vitamins and other co-factors required for survival and reproduction (Binetruy et al., 2020; Bonnet et al., 2017; Duron et al., 2017). The coexistence of *Francisella* and *Rickettsia* in ticks on camels from Abu Dhabi in this study highlights the need to characterize the interactions between diverse microbes in ticks (Binetruy et al., 2020).

4.4.3 Detection of Piroplasmids and their Prevalence

Theileriosis has a large economic impact at the global level due to losses in the livestock industry (Bilgic et al., 2019). Better control measures like immunization with a live attenuated vaccine have been effectively used to control theileriosis (Bilgic et al., 2019). Many *Theileria* species have been reported across the MENA region. In this study, *Theileria* spp. was found in Sharjah only and in low prevalence. This may be due to differences in breeds of livestock, farming conditions, and frequency of acaricide application amongst the different Emirates. *Theileria ovis* is reported here for the first time in *H. anatolicum* ticks from goats in the UAE. The genotype was identical to *T. ovis* in cattle and sheep from Iraq and Egypt, suggesting that our genotype could be a geographically widespread variant. *Theileria annulata* detected in cattle in this study was identical to previously identified genotypes from the UAE (Al-Deeb et al., 2015) and clustered with *T. annulata* from Italy, Pakistan, and Egypt, again suggesting that the variant was widespread. The prevalence of *Theileria* in livestock from Oman (Al-Fahdi et al., 2017) and Saudi Arabia is also comparable to our findings (Alanazi et al., 2021). Furthermore, the highest prevalence of *Theileria* infections occur in *H. anatolicum* compared to *H. excavatum*, *H. scupense* and *H. marginatum*, suggesting that *H. anatolicum* may be the main vector of theileriosis

(Aktas et al., 2004). Thus, the Arabian Peninsula could be a region where theileriosis may become endemic in the future. The presence of Malignant Ovine Theileriosis (MOT) in Oman indicates that mixed species infections are associated with pathogen density regulation (presumably through within-host interactions), resulting in lower mortality (Awad et al., 2020). The role of mixed infections of *Theileria* pathogens in the epidemiology of ovine theileriosis is required to be investigated for a control strategy and improved clinical outcome (Awad et al., 2020). Overall, the findings of the current study highlight that tick-vectored microorganisms continue to be detected repeatedly in the UAE because of the increasing livestock industry and associated tick vectors. Thus, there is a need for annual large-scale disease screening programs. Furthermore, it may be suggested that detailed investigations of the abundance and diversity of these piroplasm pathogens and their mixed infections in vector populations (ticks) need to be performed all over the UAE. In addition, continuous surveillance is imperative to maintain the good health of livestock and for the early detection of disease catastrophes in ruminants.

Chapter 5: Conclusion

Overall, this study provides new data in four aspects of ticks and tick-borne diseases research in the UAE, with a combination of multiple disciplines in the field of biology such as tick ecology, taxonomy, and molecular biology by using genomics and genetic approaches. First, it provides the prevalence, distribution, and molecular records of the four hard tick species collected from the livestock in the UAE. The tick species infesting domestic animals (camel, cattle, sheep, and goat) in the study area were *A. lepidum*, *H. anatolicum*, *H. dromedarii*, and *R. sanguineus*. The presence of different groups of tick species reveals the possible biological diversity of hard ticks present in the UAE desert ecosystem and livestock markets. Therefore, this work will be a milestone to avoid the emergence or re-emergence of future tick-borne diseases associated with these tick species by their continued monitoring and surveillance. Second, this study provides the first camel tick population dynamics assessment. Further, it reported the actual developmental stages of camel tick which parasitizes camels throughout the year in the UAE. Results showed that *H. dromedarii* ticks have a constant presence on camels without any temporal gaps, which specifies the magnitude of the tick problem and the probable lack of success of chemical control. Based on current research data, it is suggested that tick management strategies may be implemented from March to June, which is the time of the tick population peak. Third, the present study advances the knowledge about the microbial communities in *H. dromedarii* ticks in the UAE. It provides clear evidence that the microbiota of *H. dromedarii* is rich and diverse with the potential of harboring pathogenic bacteria, which pose a serious health risk to camels and people. 16S rRNA gene-based sequencing, presented in the current study, gives phylum, class, and family

identifications and sheds light on the microbial diversity in *H. dromedarii* in general. In addition, it provides baseline genus identification considering some of the limitations of 16S rRNA gene-based sequencing. Therefore, it is suggested that further investigation of the microbial ecology of the *H. dromedarii* is required and it also calls for a deeper understanding of how some species of its microbiota become dominant over time especially the pathogenic ones. The results of this study set the stage for further screening and detection of pathogenic species that pose serious health risks to camels and humans. Fourth, molecular detection of microbes in *Hyalomma* ticks was done through PCR based approaches. The main finding was that *H. dromedarii* ticks collected from camels had *Francisella* sp. (5.81%) and *Rickettsia* sp. (1.36%), whereas *H. anatolicum* ticks collected from cows were found to be positive for *T. annulata* (4.55%). Moreover, *H. anatolicum* ticks collected from goats were positive for *T. ovis* (10%). This study suggests large-scale screening of these microbes from livestock in all emirates of the UAE for assessment of their future threat to public health and livestock industry.

Future research is needed on ticks' systematics; their prevalence and distribution in the UAE. It is also mandatory to study the interaction between microorganisms associated with livestock ticks, their prevalence, and ecological inferences to understand tick-borne zoonotic diseases' emergence and re-emergence, and their epidemiology towards management strategies.

5.1 Recommendations

5.1.1 Lack of Published Record of Research Data in Arab Countries

Published data on ticks and tick-borne pathogens in some Arab countries is limited, despite the presence of animals. One possibility is that there might be no tick

infestations in the country, which is very unlikely, knowing how prevalent the ticks are in the region. The second possibility might be that there is no research conducted on ticks, which could be due to different problems that need to be investigated and fixed. Whatever the case might be, the concerned authorities in each country should encourage tick-related research and provide necessary resources especially financial support.

5.1.2 Need of Mutual Collaboration

In the MENA region, some tick species are common in neighboring countries that share joint borders. There is a need for a call for mutual collaboration among such countries to study and stop tick cross-border movement. In addition, the success of any tick control program in one country is always going to be reliant on good collaboration from the country on the other side of the border. Otherwise, it will serve as a tick reservoir from which ticks continue to cross into the border and reestablish infestations. In order to enhance tick control efforts, inter-country research projects should be established and supported by inter-country funding. This is very important for the management of tick species that are common in more than one country. Moreover, establishing a central collection of tick specimens and a repository for DNA and RNA samples extracted from different tick species in each country can facilitate and enhance the research on ticks. Consequently, tick management will be more successful over time. In addition, the collaboration among research teams in different countries will be more successful and effective.

5.1.3 Need to Create Awareness through Workshops and Conferences

It is essential to organize awareness creation workshops to ensure that the reporting of ticks and tick-borne pathogens is an ongoing practice for people dealing with animals and that this always needs to be done in a proper and timely manner. Because some animal care providers do not fully appreciate the importance of publishing research results or reporting the presence of tick and tick-borne pathogens as long as proper treatment is given to affected animals.

5.1.4 Animal Trade (Import and Export) Regulations

Animal trade (import and export) regulations concerning border inspections of animals for the presence of ticks and tick-borne pathogens should be standardized among all Arab countries. This practice can eliminate, or at least minimize, any infiltration of ticks into a new country as a result of lenient inspection on some points of entry.

5.1.5 Tick Surveillances Programs

There is a need for conducting comprehensive tick surveillances (qualitative and quantitative) in each Arab country to know tick species and hosts. The results should be coupled with tick species mapping to determine the geographical distribution of tick infestations and the hot spots in each country. Latest mapping software and global positioning system data should be utilized.

5.1.6 Reference Laboratory

It is vital to establish a standard reference laboratory in each Arab country to identify the tick-borne pathogens, that will serve as an information resource and point of contact at the national, regional, and international levels.

5.1.7 Cutting-Edge Research Protocols

There is a need to encourage the use of cutting-edge research protocols for studying and identifying ticks and tick-borne pathogens with an emphasis on the use of molecular tools and next-generation sequencing. Because the ixodid tick species reported in this review have veterinary and medical importance. The increase in tick-borne diseases has been attributed to a range of factors that include habitat fragmentation, changes in host communities, human travel and trade, and climate change. Ending critical gaps in tick-borne diseases ecology research would significantly improve our ability to forecast the location and timing of hot spots of these infectious diseases and to target control efforts at the most important phase of the transmission cycle. Efficient prevention and control require an understanding of ecology and human behavior.

5.1.8 Acaricide Resistance

The presence of ticks after acaricide application on the treated animals suggested that ticks had developed somehow acaricide resistance or this was likely because immature stages of ticks had fed on different hosts.. Thus, genomic approaches may help us to understand acaricide resistance by unraveling the molecular mechanisms conferring them. The resistance emerges when an acaricide is used intensively to control ticks, which may select for mutations in the genes encoding

detoxification enzymes, Glutathion-S-Transferases, Esterases, and Mixed function oxidases. This aspect needs to be investigated.

5.2 Research Implications

5.2.1 Strategies for Prevention and Control of Infectious Diseases

One health approach is used to tackle zoonotic diseases by considering all components including environmental, domestic/wild animals, and human factors. The success of this multi-disciplinary approach has been driven by combining the field sciences with analytical approaches and laboratory science. Challenges remain, however. Tick-borne disease management is likely best achieved through integrated public health, veterinary medicine, animal management, and ecological approaches. Health approaches are also mandatory at the policy and governance levels and these become successful and cost-effective if developed and implemented by all relevant parties including ecologists, entomologists/parasitologists, conservation biologists, policy-makers, and experts from veterinary and medical professions.

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List of Publications

PhD Dissertation Publications

Perveen, N., Muzaffar, S. B., & Al-Deeb, M. A. (2021). Prevalence, Distribution, and Molecular Record of Four Hard Ticks from Livestock in the United Arab Emirates. Insects, 12(11), 1016. https://doi.org/10.3390/insects12111016

Perveen, N., Muzaffar, S. B., & Al-Deeb, M. A. (2021). Four tick-borne microorganisms and their prevalence in *Hyalomma* ticks collected from livestock in United Arab Emirates. *Pathogens*, *10*(8), 1005. https://doi.org/10.3390/pathogens10081005

Perveen, N., Muzaffar, S. B., & Al-Deeb, M. A. (2021). Ticks and Tick-Borne Diseases of Livestock in the Middle East and North Africa: A Review. *Insects*, *12*(1), 83. https://doi.org/10.3390/insects12010083

Perveen, N., Muzaffar, S. B., Vijayan, R., & Al-Deeb, M. A. (2020). Microbial communities associated with the camel tick, *Hyalomma dromedarii*: 16S rRNA gene-based analysis. *Scientific reports*, *10*(1), 1-11. https://doi.org/10.1038/s41598-020-74116-7

Perveen, N., Muzaffar, S. B., & Al-Deeb, M. A. (2020). Population dynamics of *Hyalomma dromedarii* on camels in the United Arab Emirates. *Insects*, *11*(5), 320. https://doi.org/10.3390/insects11050320

Non-PhD Dissertation Publications

Perveen, N., Muzaffar, S. B., Vijayan, R., & Al-Deeb, M. A. (2021). Microbial composition in Hyalomma anatolicum collected from livestock in the United Arab Emirates using next-generation sequencing. *Parasites and Vectors* (Accepted).

Perveen, N., Muzaffar, S. B., & Al-Deeb, M. A. (2021). Exploring human-animal host interactions and emergence of COVID-19: evolutionary and ecological dynamics. Saudi *Journal of Biological Sciences*, 28, 1417–1425. https://doi.org/10.1016/j.sjbs.2020.11.077

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Appendices

Appendix 1

Tick collection sites and distribution of tick species on livestock in the study area, in 2019, 2020 and 2021, in the United Arab Emirates.

Sr. No.	**Farm Locations**	**Camels (n=300)**			**Cows (n=119)**			**Sheep (n=97)**			**Goats (n=71)**			***Hyalomma dromedarii**			****Hyalomma anatolicum**			*****Amblyomma lepidum**			******Rhipicephalus sanguineus**		
Abu Dhabi		2019	20	21	2019	20	21	2019	20	21	2019	20	21	2019	20	21	2019	20	21	2019	20	21	2019	20	21
1.	Al-Yahar	5 (6)	-	-	-	-	-	15 (30)	-	-	-	-	-	58	-	-	104	-	-	-	-	-	-	-	-
2.	Remah	5 (5)	-	-	-	-	-	-	-	-	-	-	-	33	-	-	-	-	-	-	-	-	-	-	-
3.	Al-Hayer	5 (5)	-	-		-	-		-	-	-	-	-	122	-	-	-	-	-	-	-	-	-	-	-
4.	Nahel	18 (21)	-	-	-	-	-	-	-	-	-	-	-	740	-	-	-	-	-	-	-	-	-	-	-
5.	DR	29 (35)	-	-	0 (14)	-	-	-	-	-		-	-	347	-	-	-	-	-	-	-	-	-	-	-
6.	BBS	6 (6)	-	-	-	-	-	-	-	-	-	-	-	73	-	-	-	-	-	-	-	-	-	-	-
7.	Swehan	16 (16)	-	-	-	-	-	-	-	-	0 (15)	-	-	217	-	-	-	-	-	-	-	-	-	-	-
8.	Al-Saad	16 (16)	10 (10)	-		-	-		-	-	-	-	-	200	98	-	7	-	-	-	-	-	-	-	-
9.	Beda Fares	17(17)	-	-	-	-	-		-	-	-	-	-	102	-	-	-	-	-	-	-	-	-	-	-
10.	LSM,AlAin	9 (11)	-	-	-	-	-	11 (30)	-	-	0 (15)	-	-	114	-	-	90	-	-	-	-	-	-	-	-
11.	TR	20 (20)	-	-	-	-	-	-	-	-	-	-	-	148	-	-	-	-	-	-	-	-	-	-	-
12.	Seh Saba	3 (3)	-	-	-	-	-	-	-	-	-	-	-	9	-	-	-	-	-	-	-	-	-	-	-
13.	Malaket	12 (12)	-	-	-	-	-	-	-	-	-	-	-	408	-	-	-	-	-	-	-	-	-	-	
14.	Omghafa	15 (15)	8(8)	-	-	-	-	-	-	-	-	-	-	364	61	-	-	-	-	-	-	-	-	-	-
15.	Al-Dahra	19 (19)	9(9)	-		-	-	-	-	-	-	-	-	497	123	-	6	-	-	-	-	-	-	-	-
16.	Bukriya	15 (15)	-	-	-	-	-	-	-	-	-	-	-	245	-	-	-	-	-	-	-	-	-	-	
17.	Al-Arad	10 (10)	-	-	-	-	-	-	-	-	-	-	-	144	-	-	-	-	-	-	-	-	-	-	-
18.	Al-Wagan	18 (18)	-	-	-	-	-	-	-	-	-	-	-	312	-	-	-	-	-	-	-	-	-	-	-
S. total		265 (277)			0 (14)			26 (60)			0 (30)			4415			207			0			0		
Dubai																									
19.	DCH	17 (20)			-	-	-	-	-	-	-	-	-	248	-	-	-	-	-	-	-	-	-	-	-
20.	LSMQusais	-			43(93)			9 (30)			9 (35)						230	-	64	-	-	2			
S. total		17 (20)			43 (93)			9 (30)			9 (35)			248			294			2			0		

Sharjah																							
21.	Kalba	1 (3)			2 (12)			1 (7)			1 (6)			11			76	-	-	-	-	-	5	-	-
S. total		1 (3)			2 (12)			1 (7)			1 (6)			11			76			-			5		
Total		283(300)			45 (119)			36 (97)			10 (71)			4674			577			2			5		

DR=Dubai road, BBS=Bede Bent Saud, TR=Truck road, LSM=Livestock market, DCH=Dubai camel hospital
In parentheses, total number of examined animals are given
**Hyalomma dromedarii*=all ticks collected from camels, ***Hyalomma anatolicum*= ticks collected from cows, sheep and goats
****Amblyomma lepidum*= ticks collected from cows, *****Rhipicephalus sanguineus*= ticks collected from cows

Appendix 2

Prevalence of microbes in *Hyalomma* ticks in UAE.

Sr. No.	**Farm locations**	**Samples (Camels)**	**Samples (Cows)**	**Samples (Sheep)**	**Samples (Goats)**	**Total samples**	***Francisella* sp. (positive samples)**	***Rickettsia* sp. (positive samples)**	***Theileria annulata* (positive samples)**	***Theileria ovis* (positive samples)**
Abu Dhabi										
1.	Al-Foah, UAEU	2	-	-	-	2	1	-	-	-
2.	Beda Bent Saud	6	-	-	-	6	1	-	-	-
3.	Livestock Market, Al Ain	11	-	4(36)	-	15	1	-	-	-
4.	Nahel Town	24	-	-	-	24	2	2	-	-
5.	Omghafa	84	-	-	-	84	3	1	-	-
6.	Truck Road	20	-	-	-	20	4	-	-	-
7.	Al-Wagan	30	-	-	-	30	5	1	-	-
8.	Dubai Road	70	0(15)	-	-	70	4	1	-	-
9.	Bukriya	15	-	-	-	15	5	2	-	-
10.	Al-Saad	53	-	-	-	53	4	-	-	-
11.	Al-Hayer	5	-	-	-	5	-	-	-	-
12.	Swehan	24	-	-	-	24	-	-	-	-
13.	Nabagh	15	-	-	-	15	-	-	-	-

14.	Al-Dahra	50	-	-	0(30)	50	-	-	-	-
15.	Al-Yahar	6	-	-	-	6	-	-	-	-
16.	Malaket	12	-	-	-	12	-	-	-	-
17.	Remah	5	-	-	-	5	-	-	-	-
18.	Beda Fares	17	-	-	-	17	-	-	-	-
19.	Seh Saba	3	-	-	-	3	-	-	-	-
20.	Bilayat	2	-	-	-	2	-	-	-	-
21.	Al-Arad	44	-	-	-	44	-	-	-	-
Sub-total		498(498)	0	4(36)	0	502(534)	30	7		
Dubai										
22.	Dubai Camel Hospital	17(17)	-	-	-	17	-	-	-	-
23.	Al-Qusais Market, Dubai	-	20(20)	9(9)	9(9)	38	-	-	-	-
Sub-total		17(17)	20(20)	9(9)	9(9)	55(55)				
Sharjah										
24.	Kalba, Sharjah	1(3)	2(6)	1(25)	1(25)	5	-	-	1	1
Sub-total		1(3)	2(6)	1(25)	1(25)	5(59)	-	-	1(detected in ticks from cows)	1(detected in ticks from goats)
Total		516(518)	22(26)	14(70)	10(34)	562(648)	30	7	1	1

Numbers in parenthesis represent the number of animals.

Appendix 3

Molecular identification of *Hyalomma dromedarii* from camels in Abu Dhabi, UAE based on DNA similarity between *16S* rRNA gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**
Mitochondrion *Hyalomma dromedarii* 16S ribosomal RNA (16S rRNA) gene	L34306.1	99.27	98	0.0		
Hyalomma dromedarii voucher HyT85 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN960589.1	99.50	95	0.0	Camel	Tunisia
Hyalomma dromedarii 16S ribosomal RNA gene, partial sequence; mitochondrial	MG757400.1	99.50	95	0.0	Camel	Egypt
Hyalomma dromedarii voucher HyT1 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN960579.1	99.49	94	0.0	Camel	Tunisia
Hyalomma dromedarii 16S ribosomal RNA gene, partial sequence; mitochondrial	MG972372.1	98.53	96	0.0	Camel	Saudi Arabia
Hyalomma dromedarii isolate Gebal_92 16S ribosomal RNA gene, partial sequence; mitochondrial	KY512798.1	99.49	93	0.0	*Dipodillus dasyurus*	Egypt
Hyalomma dromedarii isolate Gharaba _13/11 16S ribosomal RNA gene, partial sequence; mitochondrial	KY512796.1	99.49	93	0.0	Camel	Egypt
Hyalomma dromedarii isolate MATRUH-EGY 16S ribosomal RNA gene, partial sequence; mitochondrial	MF946465.1	98.99	94	0.0	Camel	Egypt
Hyalomma dromedarii isolate Marsa-Matruh 16S ribosomal RNA gene, partial sequence; mitochondrial	KY945490.1	98.99	94	0.0	Camel	Egypt
Hyalomma dromedarii isolate Arbaein_15/3 16S ribosomal RNA gene, partial sequence; mitochondrial	KY512797.1	99.23	93	0.0		Egypt
Hyalomma dromedarii strain ST6HD2 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MH569481.1	99.74	91	0.0	Camel	Saudi Arabia
Hyalomma dromedarii voucher HyT48 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN960587.1	99.23	93	0.0	Camel	Tunisia
Hyalomma dromedarii voucher HyT7 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN960580.1	99.48	91	0.0	Camel	Tunisia

Hyalomma dromedarii strain ST3HD35 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MH569480.1	99.48	91	0.0	Camel	Saudi Arabia
Hyalomma dromedarii strain ST2HD44 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MH569479.1	99.48	91	0.0	Camel	Saudi Arabia
Hyalomma dromedarii strain ST5HD50 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MH569478.1	99.48	91	0.0	Camel	Saudi Arabia
Hyalomma dromedarii strain ST1HD46 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MH569477.1	99.48	91	0.0	Camel	Saudi Arabia
Hyalomma dromedarii strain ST4HD14 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MH569476.1	99.48	91	0.0	Camel	Saudi Arabia
Hyalomma dromedarii isolate Hdrom4 16S ribosomal RNA gene, partial sequence; mitochondrial	KU130425.1	99.74	90	0.0		Senegal
Hyalomma dromedarii isolate Hdrom2 16S ribosomal RNA gene, partial sequence; mitochondrial	KU130423.1	99.74	90	0.0		Pakistan
Hyalomma dromedarii isolate Hdrom3 16S ribosomal RNA gene, partial sequence; mitochondrial	KU130424.1	99.47	90	0.0		Saudi Arabia
Hyalomma dromedarii isolate KNC106 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN394434.1	99.21	90	0.0	Camel	Nigeria
Hyalomma dromedarii voucher MT1 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MT895170.1	99.73	89	0.0	Camel	Kenya
Hyalomma dromedarii voucher MT120 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MT895169.1	99.73	89	0.0	Camel	Kenya
Hyalomma dromedarii isolate Hdrom1 16S ribosomal RNA gene, partial sequence; mitochondrial	KU130422.1	99.21	90	0.0		Iraq
Hyalomma dromedarii isolate KNC19 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN394429.1	100.00	88	0.0	Camel	Nigeria
Hyalomma dromedarii voucher HyT41 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN960585.1	97.95	93	0.0	Camel	Tunisia
Hyalomma dromedarii isolate KNC21 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN394430.1	100.00	86	0.0	Camel	Nigeria
Hyalomma dromedarii isolate KNC47 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN394431.1	99.45	86	0.0	Camel	Nigeria
Hyalomma somalicum isolate Hsoma1 16S ribosomal RNA gene, partial sequence; mitochondrial	KU130472.1	98.15	90	0.0		Somalia

Hyalomma dromedarii isolate KNC13 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN394427.1	98.90	87	0.0	Camel	Nigeria
Hyalomma dromedarii isolate KNC101 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MN394433.1	99.72	85		Camel	Nigeria
Hyalomma dromedarii isolate Hydr1 16S ribosomal RNA gene, partial sequence; mitochondrial	KT391055.1	97.17	91			Israel

Appendix 4

Molecular identification of *Hyalomma anatolicum* from cows in Dubai, UAE based on DNA similarity between *cox1* gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**
Hyalomma anatolicum anatolicum isolate COX1	MT800311.1	99.70	96	0.0	Goat	Pakistan
Hyalomma anatolicum voucher AC9 cytochrome oxidase subunit 1 (COI) gene	MH459380.1	99.39	97	0.0	Cattle	China
Hyalomma anatolicum anatolicum isolate 3old cytochrome oxidase subunit I (Cox1) gene	KP792577.1	99.85	95	0.0	Buffalo	India
Hyalomma anatolicum isolate GY44-2 cytochrome oxidase subunit I (COI) gene	MN853167.1	99.24	97	0.0	Cattle	China
Hyalomma anatolicum isolate GY43-1 cytochrome oxidase subunit I (COX1) gene	MN841463.1	99.24	97	0.0	Cattle	China

Hyalomma anatolicum voucher AC5 cytochrome oxidase subunit 1 (COI) gene	MH459377.1	99.24	97	0.0	Cattle	China
Hyalomma anatolicum anatolicum cytochrome oxidase subunit I (COI) gene	KJ912622.2	99.85	95	0.0	Cattle	India
Hyalomma anatolicum anatolicum isolate Gansu cytochrome oxidase subunit I (COI) gene	JQ737067.1	99.24	97	0.0	Cattle	China
Hyalomma anatolicum isolate PAK5 cytochrome c oxidase subunit 1 (cox1) gene	MK462197.1	99.69	95	0.0	Cattle	Pakistan
Hyalomma anatolicum isolate XJ-TLF-Han-2019001 cytochrome c oxidase subunit I (COX1) gene	MW221948.1	99.24	96	0.0	Cattle	China
Hyalomma anatolicum isolate XJ074 cytochrome c oxidase subunit I gene	KF583577.1	99.39	96	0.0	Cattle	China
Hyalomma anatolicum voucher ACc cytochrome oxidase subunit 1 (COI) gene	MH459383.1	99.09	97	0.0	lab rearing	China
Hyalomma anatolicum isolate PACA-83 cytochrome c oxidase subunit 1 (cox1) gene	MK462202.1	99.54	95	0.0	Cattle	Pakistan
Hyalomma anatolicum isolate PACA-116 cytochrome c oxidase subunit 1 (cox1) gene	MK462200.1	99.54	95	0.0	Cattle	Pakistan
Hyalomma anatolicum isolate PACA-88 cytochrome c oxidase subunit 1 (cox1) gene	MK462199.1	99.54	95	0.0	Cattle	Pakistan
Hyalomma anatolicum isolate PAK6 cytochrome c oxidase subunit 1 (cox1) gene,	MK462198.1	99.54	95	0.0	Cattle	Pakistan
Hyalomma anatolicum voucher TK0G9 cytochrome oxidase subunit 1 (CO1) gene	MH648685.1	99.69	94	0.0	Cattle	Bangladesh

Appendix 5

Molecular identification of *Amblyomma lepidum* from cows in Dubai, UAE, based on DNA similarity between *cox1* gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**

Amblyomma lepidum cytochrome c oxidase subunit 1 (COX1) gene, partial cds; mitochondrial	KP987775.1	99.84	93	0.0	*Ovis aries*	Israel
Amblyomma lepidum voucher 19634-AlepB10 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial	KT307492.1	99.38	94	0.0		Kenya
Amblyomma cohaerens isolate AET3 cytochrome c oxidase subunit 1 (cox1) gene, partial cds; mitochondrial	MN150170.1	88.47	91	0.0	*Bos taurus*	Ethiopia
Amblyomma cohaerens isolate AET4 cytochrome c oxidase subunit 1 (cox1) gene, partial cds; mitochondrial	MN150171.1	88.31	91	0.0	*Bos taurus*	Ethiopia
Amblyomma testudinarium voucher AMMS-AF-2-1 cytochrome c oxidase subunit I-like (COI) gene, partial sequence; mitochondrial	HM193893.1	86.39	97	0.0		China
Amblyomma pattoni voucher AMMS-AP-2 cytochrome c oxidase subunit I-like (COI) gene, partial sequence; mitochondrial	HM193876.1	86.43	96	0.0		China
Amblyomma hebraeum isolate 190 cytochrome c oxidase subunit I (COX1) gene, partial cds; mitochondrial	MT549815.1	86.65	93	0.0		China
Amblyomma hebraeum isolate 200 cytochrome c oxidase subunit I (COX1) gene, partial cds; mitochondrial	MT549816.1	86.34	93	0.0		China
Amblyomma testudinarium isolate TWKL-Amt1 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	KX712282.1	86.15	94	0.0		Taiwan
Amblyomma testudinarium isolate TWKL-Amt3 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	KX712284.1	86.00	94	0.0		Taiwan
Amblyomma testudinarium isolate TWKL-Amt2 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	KX712283.1	86.00	94	0.0		Taiwan
Amblyomma variegatum isolate 1 cytochrome c oxidase subunit I (COX1) gene, partial cds; mitochondrial	MT549807.1	85.49	93	0.0		China

Amblyomma variegatum isolate 11 cytochrome c oxidase subunit I (COX1) gene, partial cds; mitochondrial	MT549808.1	85.34	93	0.0		China
Amblyomma pattoni voucher AMMS-AP-5 cytochrome c oxidase subunit I-like (COI) gene, partial sequence; mitochondrial	HM193875.1	85.69	91	0.0		China
Amblyomma scalpturatum isolate scalpturatumN2921 cytochrome oxidase subunit 1 (COX1) gene, partial cds; mitochondrial	MH513238.1	84.98	93	0.0		French Guiana
Amblyomma scalpturatum isolate scalpturatumN4222 cytochrome oxidase subunit 1 (COX1) gene, partial cds; mitochondrial	MH513239.1	84.83	93	0.0		French Guiana
Amblyomma variegatum isolate ANI1 cytochrome c oxidase subunit 1 (cox1) gene, partial cds; mitochondrial	MN150168.1	85.17	91	0.0	*Canis familiaris*	Nigeria

Appendix 6

Molecular identification of *Rhipicephalus sanguineus* from cows in Sharjah, UAE based on DNA similarity between *16S* rRNA gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**
Rhipicephalus sanguineus clone TVM small subunit ribosomal RNA gene, partial sequence; mitochondrial	MG066692.1	99.03	96	0.0	Dog	India
Rhipicephalus sanguineus strain 7 16S ribosomal RNA gene, partial sequence	AY883868.1	98.56	98	0.0	Dog	Taiwan
Rhipicephalus sanguineus isolate 3 16S ribosomal RNA gene, partial sequence; mitochondrial	MH765331.1	98.79	97	0.0	Goat	India
Rhipicephalus sanguineus isolate Bejucal 16S ribosomal RNA gene, partial sequence; mitochondrial	KP830114.1	98.33	98	0.0	Dog	*Cuba*

Rhipicephalus sanguineus 16S ribosomal RNA gene, partial sequence; mitochondrial	KC170744.1	98.33	98	0.0	Dog	*Thailand*
Rhipicephalus sanguineus isolate InDRE large subunit ribosomal RNA gene, partial sequence; mitochondrial	MT322611.1	98.33	98	0.0	Dog	Mexico
Rhipicephalus sanguineus strain 13 16S ribosomal RNA gene, partial sequence	AY883871.1	98.33	98	0.0	Dog	Taiwan
Rhipicephalus sanguineus strain 12 16S ribosomal RNA gene, partial sequence	AY883870.1	98.33	98	0.0	Dog	Taiwan
Rhipicephalus sanguineus strain 40 16S ribosomal RNA gene, partial sequence	AY883880.1	98.33	98	0.0	Dog	Taiwan
Rhipicephalus sanguineus strain 5 16S ribosomal RNA gene, partial sequence	AY883866.1	98.09	98	0.0	Dog	Taiwan
Rhipicephalus sanguineus isolate 2 large subunit ribosomal RNA gene, partial sequence; mitochondrial	MT026919.1	99.01	95	0.0	Mammals	Colombia
Rhipicephalus sanguineus strain 2 16S ribosomal RNA gene, partial sequence	AY883863.1	98.09	98	0.0	Dog	Taiwan
Rhipicephalus sanguineus isolate Colombia-Leticia 16S ribosomal RNA gene, partial sequence; mitochondrial	MF351600.1	98.09	98	0.0	Domestic animals	Colombia
Rhipicephalus sanguineus isolate Brazil-Minas Gerais 16S ribosomal RNA gene, partial sequence; mitochondrial	MF351603.1	98.31	97	0.0	Domestic animals	Colombia
Rhipicephalus sanguineus strain 32 16S ribosomal RNA gene, partial sequence	AY883878.1	97.86	98	0.0	Dog	Taiwan
Rhipicephalus sanguineus isolate Colombia-Saldana 16S ribosomal RNA gene, partial sequence; mitochondrial	MF351583.1	98.53	96	0.0	Domestic animals	Colombia
Rhipicephalus sanguineus isolate Colombia-Ibague16S ribosomal RNA gene, partial sequence; mitochondrial	MF351580.1	98.53	96	0.0	Domestic animals	Colombia

Rhipicephalus sanguineus haplotype A 16S ribosomal RNA gene, partial sequence; mitochondrial	KC476294.1	99.01	94	0.0	Dog	Nigeria
Rhipicephalus sanguineus isolate Colombia-Puerto Salgar 16S ribosomal RNA gene, partial sequence; mitochondrial	MF351595.1	98.07	97	0.0	Domestic animals	Colombia
Rhipicephalus sanguineus isolate Colombia-Cali(39-34V3C1) 16S ribosomal RNA gene, partial sequence; mitochondrial	MF351591.1	98.07	97	0.0	Domestic animals	Colombia
Rhipicephalus sanguineus isolate Colombia-Yopal 16S ribosomal RNA gene, partial sequence; mitochondrial	MF351588.1	98.07	97	0.0	Domestic animals	Colombia
Rhipicephalus sanguineus isolate IqRam 16S ribosomal RNA gene, partial sequence; mitochondrial	KT382453.1	99	94	0.0	Dog	Iraq
Rhipicephalus sanguineus isolate RSRC3 16S ribosomal RNA gene, partial sequence; mitochondrial	JX997392.1	98.52	95	0.0	Lab.colony	Brazil
Rhipicephalus sanguineus isolate RSJ1 16S ribosomal RNA gene, partial sequence; mitochondrial	JX997391.1	98.52	95	0.0	Lab.colony	Brazil
Rhipicephalus sanguineus 16S ribosomal RNA gene, partial sequence; mitochondrial	KU198404.1				Dog	Egypt
Rhipicephalus sanguineus isolate Orkun-RS314 small subunit ribosomal RNA gene, partial sequence; mitochondrial	KR870984.1				Dog	Turkey

Appendix 7

Prevalence of ticks in camels, cows, sheep and goats in the United Arab Emirates[a].

Hosts	Type				Total
	Camels N (%)	Cows N (%)	Sheep N (%)	Goats N (%)	
Examined animals	300	119	97	71	587
Infested with ticks	283 (94.3)	45 (37.81)	36 (37.1)	10 (14)	374 (63.71)

[a] Number of infested animals/Number of examined animal X 100.

Appendix 8

Prevalence of ticks in camels, cows, sheep and goats in relation to sex of animals in the United Arab Emirates[b].

Host	Type							
	Camels N (%)		**Cows N (%)**		**Sheep N (%)**		**Goats N (%)**	
	Male	Female	Male	Female	Male	Female	Male	Female
Examined animals	17	283	64	55	28	69	44	27
Infested with ticks	15 (88.24)	268 (94.7)	26 (40.63)	19 (34.55)	7 (25)	29 (42)	3 (6.82)	7 (25.93)

[b] Number of infested animals/Number of examined animal X 100.

Appendix 9

Number (N) of tick species collected from camels, cows, sheep and goats in the United Arab Emirates.

Tick species	Hosts				Total
	Camels (N)	**Cows (N)**	**Sheep (N)**	**Goats (N)**	
Amblyomma lepidum	0	2	0	0	2
Hyalomma anatolicum	13	317	145	102	577
Hyalomma dromedarii	4674	0	0	0	4674
Rhipicephalus sanguineus	0	5	0	0	5
Others	116	327	132	117	692
Total	4803	651	277	219	5950

Appendix 10

Microbial phyla (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2010.

Phylum	**BF**	**BS**	**DR**	**DS**	**MQ**	**RH**	**SW**	**OM**	**AW**	**AY**
Actinobacteria	7.40	1.51	18.25	41.60	2.25	17.66	29.06	26.12	21.68	33.84
Bacteroidetes	26.95	0.87	1.11	1.30	0.37	0.71	0.60	0.26	3.82	0.46
Chloroflexi	0.00	0.00	0.00	0.02	0.00	0.00	0.00	0.00	0.00	0.00
Cyanobacteria	0.00	0.00	0.07	0.00	0.00	0.00	0.00	0.00	1.15	0.07
Deinococcus-Thermus	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02
Firmicutes	6.15	43.38	30.19	17.98	9.52	8.05	6.92	30.74	47.27	19.26
Gemmatimonadetes	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.03	0.00	0.00
Planctomycetes	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.28	0.00	0.00
Proteobacteria	59.47	54.21	50.15	37.97	87.86	73.57	63.28	42.53	25.21	46.30
Verrucomicrobia	0.00	0.00	0.00	0.41	0.00	0.00	0.13	0.00	0.21	0.00

Appendix 11

Microbial classes (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2010.

Class	**BF**	**BS**	**DR**	**DS**	**MQ**	**RH**	**SW**	**OM**	**AW**	**AY**
Actinobacteria	7.40	1.51	18.24	41.60	2.25	17.66	29.06	26.12	21.62	33.84
Alphaproteobacteria	0.07	0.00	0.10	0.36	0.08	0.14	0.05	0.26	0.48	0.07
Bacilli	4.52	43.36	26.24	11.77	8.89	6.43	2.58	30.07	41.90	16.69
Bacteroidia	0.07	0.07	0.39	0.99	0.13	0.48	0.38	0.06	3.43	0.22
Betaproteobacteria	0.15	0.04	8.87	0.58	0.52	0.42	0.13	6.63	0.12	3.35
Chitinophagia	0.01	0.00	0.00	0.30	0.00	0.00	0.00	0.08	0.00	0.00
Clostridia	0.53	0.02	0.21	0.62	0.12	0.05	0.05	0.13	3.95	0.31
Coriobacteriia	0.00	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.06	0.00
Cytophagia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.00
Deinococci	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02
Deltaproteobacteria	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.15	0.00
Erysipelotrichia	0.00	0.00	0.00	0.12	0.04	0.00	0.00	0.05	0.02	0.00
Flavobacteriia	26.66	0.80	0.72	0.00	0.01	0.23	0.09	0.11	0.39	0.23

Fusobacteriia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Gammaproteobacteria	59.25	54.17	41.18	37.03	87.25	73.01	63.10	35.65	24.45	42.89
Gemmatimonadetes	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.00
Longimicrobia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.03	0.00	0.00
Negativicutes	0.05	0.00	0.08	0.00	0.00	0.09	0.00	0.00	0.17	0.00
Oligoflexia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00
Planctomycetia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.28	0.00	0.00
Sphingobacteriia	0.21	0.00	0.00	0.00	0.22	0.00	0.13	0.00	0.00	0.00
Thermomicrobia	0.00	0.00	0.00	0.02	0.00	0.00	0.00	0.00	0.00	0.00
Tissierellia	1.05	0.00	3.65	5.47	0.47	1.48	4.29	0.50	1.23	2.27
Verrucomicrobiae	0.00	0.00	0.00	0.41	0.00	0.00	0.13	0.00	0.21	0.00

Appendix 12

Microbial families (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2010.

Family	**AW**	**AY**	**BF**	**BS**	**DR**	**DS**	**MQ**	**OM**	**RH**	**SW**
Moraxellaceae	2.84	13.43	14.27	49.72	31.02	33	77.52	16.62	7.1	2.31
Corynebacteriaceae	21	32.23	7.25	1.42	14.2	36.62	2.04	25	13.28	23.82
Enterobacteriaceae	9.51	1.8	23.54	3.47	7	0.5	7.46	4.3	54.63	4.54
Flavobacteriaceae	0.22	0.23	26.66	0.8	0.72	0	0.01	0.04	0.23	0.1
Staphylococcaceae	38.1	15	3.74	0.28	1.5	6.42	5.84	19.3	6.01	1.81
Bacillaceae	1	1.03	0.37	37.33	24.5	3	2.54	6.06	0.1	0.3
Morganellaceae	0	0.3	19.73	0	1	3	0	0.02	6.03	55.82
Francisellaceae	11.4	2.33	0.86	1	0.34	0.2	0.33	13	2.88	0.33
Bacteroidaceae	2.3	0.12	0	0	0	0	0	0	0.28	0.04
Ruminococcaceae	1.4	0	0	0	0	0	0	0	0	0
Peptoniphilaceae	1.13	2.24	1.05	0	3.6	5.4	0.47	0.5	1.48	4.3
Lachnospiraceae	1.12	0	0.44	0.02	0	0.31	0.05	0.05	0.03	0.01
Planococcaceae	1.04	0	0	0	0	0.01	0.14	3	0	0.03

Aerococcaceae	1	0.23	0.1	0	0.1	2.41	0.25	0.3	0.28	0.4
Actinomycetaceae	0.4	0.3	0.1	0	1.1	0	0	0.22	0.52	0.8
Pseudomonadaceae	0.4	0.54	0.8	0.02	1.6	0.5	1.7	0.1	2.32	0.1
Brevibacteriaceae	0.2	1.1	0.1	0	2.3	3.6	0.05	0.1	3.42	2.76
Enterococcaceae	0.19	0.4	0.28	5.67	0.12	0.17	0.11	0.8	0.05	0.04
Pasteurellaceae	0.03	0	0.01	0	0.21	0.16	0.1	2	0	0
Neisseriaceae	0.01	0.34	0.13	0	8	0.42	0.32	7	0.18	0
Xanthomonadaceae	0	24.5	0.07	0	0.03	0.02	0	0.02	0	0
Comamonadaceae	0	3	0	0.04	0	0	0.01	0	0.09	0
Dermabacteraceae	0	0.19	0.02	0	0.21	0.43	0.01	0.5	0.32	1.25
Muribaculaceae	0	0	0.07	0	0.23	1	0.13	0	0	0.15
Alcaligenaceae	0	0	0.02	0	1	0.16	0.19	0	0.15	0.13
Others	6.71	0.69	0.39	0.23	1.22	2.67	0.73	1.07	0.62	0.96

Appendix 13

Microbial genera (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2010.

Genus	**Aw**	**AY**	**BF**	**BS**	**DR**	**DS**	**MQ**	**OM**	**RH**	**SW**
Acinetobacter	2.5	5.92	14.2	49.72	24.55	32.08	75.66	15.81	7.09	2.31
Escherichia	7.52	1.57	23	3.43	3.36	0	6.41	0.11	53.13	4.37
Corynebacterium	20.73	32.23	7.25	1.42	14.17	36.62	2.04	24.88	13.28	23.82
Staphylococcus	37.68	14.73	3.66	0.28	1.48	6.1	5.82	16.11	6	1.79
Bacillus	0.85	0.17	0.06	37.32	23.44	2.04	2	3.6	0.06	0.01
Proteus	0	0.26	19.73	0	0.78	2.73	0	0.02	6.03	55.82
Flavobacterium	0	0	26.65	0.8	0	0	0	0.04	0.23	0.09
Francisella	11.38	2.33	0.86	0.97	0.34	0.19	0.33	12.73	2.88	0.33
Moraxella	0.2	7.47	0.05	0	6.48	0.81	0.68	0.51	0.01	0.01
Uruburuella	0.01	0.34	0.13	0	7.94	0.42	0.32	6.43	0.18	0
Stenotrophomonas	0	24.5	0.04	0	0	0	0	0.02	0	0

Brevibacterium	0.19	1.05	0.07	0	2.25	3.55	0.05	0.1	3.42	2.76
Enterococcus	0.19	0.35	0.28	5.67	0.12	0.17	0.11	0.78	0.05	0.04
Enterobacter	1.98	0.2	0.47	0.03	3.62	0.45	0.68	4.12	1.47	0.17
Helcococcus	0.52	1.86	0.16	0	3.03	3.35	0.46	0.5	1.04	1.29
Comamonas	0	3	0	0	0	0	0.01	0	0.09	0
Solibacillus	1.04	0	0	0	0	0	0.14	2.61	0	0
Jeotgalicoccus	0.26	0.1	0.08	0	0	0.27	0	2.49	0	0.02
Pseudomonas	0.39	0.45	0.77	0.02	1.56	0.48	1.7	0.1	2.32	0.09
Facklamia	0.56	0	0.03	0	0	1.33	0.1	0.25	0	0
Brachybacterium	0	0.19	0.02	0	0.21	0.43	0.01	0.47	0.32	1.25
Peptoniphilus	0.16	0.2	0.23	0	0.42	0.16	0	0	0.18	1.18
Lysinibacillus	0.02	0.87	0.27	0.01	1.02	0.71	0.54	2.46	0.01	0.23
Bacteroides	2.3	0.12	0	0	0	0	0	0	0.28	0.04
Mannheimia	0	0	0.01	0	0.2	0.16	0.1	1.89	0	0
Anaerococcus	0.32	0.18	0.39	0	0.13	1.84	0	0	0.16	0.77
Psychrobacter	0.14	0.04	0	0	0	0.1	1.18	0.3	0	0
Finegoldia	0.03	0	0.26	0	0	0.03	0	0	0.08	1.05
Trueperella	0.36	0.25	0.03	0	1.05	0	0	0	0.52	0.67
Ignavigranum	0.07	0.22	0.08	0	0.08	1.04	0.14	0	0.22	0.39
Muribaculum	0	0	0.07	0	0.23	1	0.13	0	0	0.15
Others	10.6	1.4	1.15	0.33	3.54	3.94	1.39	3.67	0.95	1.35

Appendix 14

Microbial phyla (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2019.

Phylum	**AW**	**AY**	**BF**	**BS**	**DR**	**DS**	**MQ**	**OM**	**RH**	**SW**
Actinobacteria	0.26	5.04	0.19	0.45	7.90	0.28	0.31	0.53	0.82	0.07
Bacteroidetes	0.13	0.21	0.58	0.15	0.37	0.21	7.01	10.26	1.17	0.43
Cyanobacteria	0.02	0.00	0.00	0.00	0.08	0.00	0.00	0.00	0.30	0.00
Firmicutes	0.13	0.37	1.44	55.54	1.02	0.38	9.54	1.13	1.63	1.59
Planctomycetes	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Proteobacteria	99.26	94.14	97.08	43.80	90.35	99.12	83.14	88.07	95.77	97.69
Verrucomicrobia	0.00	0.00	0.00	0.05	0.14	0.00	0.01	0.00	0.31	0.00

Appendix 15

Microbial classes (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2019.

Class	**AW**	**AY**	**BF**	**BS**	**DR**	**DS**	**MQ**	**OM**	**RH**	**SW**
Actinobacteria	0.26	5.04	0.17	0.45	7.90	0.28	0.31	0.53	0.82	0.07
Alphaproteobacteria	0.01	0.00	0.08	0.11	0.00	0.00	0.01	0.01	0.00	0.08
Bacilli	0.07	0.21	1.44	55.52	0.61	0.30	0.70	0.85	1.23	1.52
Bacteroidia	0.12	0.20	0.58	0.13	0.37	0.21	0.05	0.09	1.17	0.31
Betaproteobacteria	0.01	0.02	5.71	0.48	0.04	0.06	0.01	0.26	0.00	0.00
Chitinophagia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.11
Clostridia	0.05	0.10	0.00	0.02	0.27	0.08	8.70	0.10	0.40	0.07
Cytophagia	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.00
Erysipelotrichia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Flavobacteriia	0.02	0.01	0.00	0.00	0.00	0.00	6.89	9.83	0.00	0.00
Gammaproteobacteria	99.24	94.12	91.29	43.21	90.31	99.06	83.12	87.81	95.77	97.61
Hydrogenophilalia	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Negativicutes	0.00	0.00	0.00	0.00	0.00	0.00	0.07	0.00	0.00	0.00
Phycisphaerae	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Sphingobacteriia	0.00	0.00	0.00	0.01	0.00	0.00	0.07	0.34	0.00	0.00
Thermoleophilia	0.00	0.00	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Tissierellia	0.00	0.06	0.00	0.00	0.13	0.00	0.07	0.18	0.00	0.00
Verrucomicrobiae	0.00	0.00	0.00	0.05	0.14	0.00	0.01	0.00	0.31	0.00

Appendix 16

Microbial families (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2019.

Family	Aw	AY	BF	BS	DR	DS	MQ	OM	RH	SW
Francisellaceae	99.1	94.1	89.5	27.4	89.3	98	0.2	62.7	95.5	97.6
Corynebacteriaceae	0.16	5.03	0.11	0.44	7.9	0.26	0.28	0.33	0.79	0.07
Staphylococcaceae	0.07	0.2	0.28	0.28	0.41	0.23	0.06	0.38	0.59	1.4
Moraxellaceae	0.04	0.06	1.61	13.51	0.04	0	3.07	21.61	0.28	0.04
Flavobacteriaceae	0.02	0.01	0	0	0	0	6.9	9.83	0	0
Clostridiaceae	0	0	0	0	0	0	8.61	0	0.12	0
Muribaculaceae	0	0.2	0.32	0.13	0.17	0.21	0	0.02	0.81	0.18
Bacillaceae	0	0.01	1.17	54.74	0.16	0.07	0	0.4	0	0.12
Neisseriaceae	0	0	5.71	0.44	0.03	0	0	0	0	0
Enterobacteriaceae	0	0	0	1.34	0.01	0.89	79.22	0.36	0	0
Pseudomonadaceae	0	0	0	0.77	0.26	0	0.62	2.04	0	0
Others	0.61	0.39	1.3	0.95	1.72	0.34	1.04	2.33	1.91	0.59

Appendix 17

Microbial genera (presence in %) detected in *H. dromedarii* adult ticks from ten locations in Al-Ain, UAE in 2019.

Genus	Aw	AY	BF	BS	DR	DS	MQ	OM	RH	SW
Francisella	99.1	94.1	89.5	27.4	89.3	98	0.2	62.7	95.5	97.6
Corynebacterium	0.16	5	0.11	0.44	7.9	0.26	0.28	0.33	0.8	0.1
Staphylococcus	0.1	0.2	0.3	0.3	0.4	0.23	0.1	0.4	0.45	1.4
Moraxella	0.04	0.1	1.1	0.1	0	0	0	0	0	0
Uruburuella	0	0	5.7	0.44	0.03	0	0	0	0	0
Bacillus	0	0.01	1.11	45.84	0.16	0.05	0	0.06	0	0.12
Muribaculum	0	0.2	0.32	0.13	0.17	0.21	0	0.02	0.81	0.18
Pseudomonas	0	0	0	0.77	0.26	0	0.62	2.04	0	0
Acinetobacter	0	0.01	0	13.42	0.04	0	1.87	15.1	0.17	0.02
Escherichia	0	0	0	1.34	0.01	0	48.41	0.02	0	0
Siccibacter	0	0	0	0	0	0.89	30.81	0.33	0	0
Lysinibacillus	0	0	0	8.8	0	0.02	0	0.34	0	0
Psychrobacter	0	0	0	0.01	0	0	1.19	6.51	0	0.02

Flavobacterium	0	0	0	0	0	0	6.89	9.04	0	0
Clostridium	0	0	0	0	0	0	8.61	0	0.12	0
Others	0.6	0.38	1.86	1.01	1.73	0.34	1.02	3.11	2.15	0.56

Appendix 18

Correlation matrix showing pairwise Pearson's r correlations between genera (bottom) and their associated significance (top).

Genera	*Francisella*	*Acinetobacter*	*Bacillus*	*Corynebacterium*	*Escherichia*	*Flavobacterium*	*Lysinibacillus*	*Moraxella*	*Muribaculum*	*Pseudomonas*	*Psychrobacter*	*Staphylococcus*	*Uruburuella*
Francisella		0.028888	0.2016	0.01161	0.042961	0.3991	0.39019	0.21176	0.62658	0.069064	0.82388	0.13323	0.61401
Acinetobacter	-0.49		0.051909	0.90693	0.91299	0.96279	0.79911	0.76339	0.96784	0.073076	0.54225	0.95475	0.79837
Bacillus	-0.3	0.44		0.47827	0.56815	0.60233	0.0088522	0.58148	0.61607	0.89166	0.57541	0.49103	0.4529
Corynebacterium	-0.55	-0.03	-0.17		0.92492	0.5946	0.98558	0.090979	0.44631	0.93551	0.36876	0.011999	0.40722
Escherichia	-0.46	-0.03	-0.14	-0.02		0.13692	0.50345	0.52328	0.17241	0.037759	0.76825	0.85305	0.47266
Flavobacterium	-0.2	0.01	-0.12	-0.13	0.34		0.70582	0.54796	0.46223	0.38783	0.33889	0.73006	0.55458
Lysinibacillus	-0.2	0.06	0.57	0	-0.16	-0.09		0.86654	0.789	0.75434	0.81548	0.9468	0.49989
Moraxella	-0.29	0.07	0.13	0.39	-0.15	-0.14	0.04		0.98666	0.38345	0.62304	0.65708	0.016011
Muribaculum	0.12	0.01	-0.12	0.18	-0.32	-0.17	-0.06	0		0.42652	0.43271	0.34805	0.84366
Pseudomonas	-0.41	0.41	0.03	-0.02	0.47	0.2	0.07	0.21	-0.19		0.018532	0.95822	0.67982
Psychrobacter	0.05	0.14	-0.13	-0.21	-0.07	0.23	-0.06	-0.12	-0.19	0.52		0.72568	0.64318
Staphylococcus	-0.35	-0.01	-0.16	0.55	0.04	-0.08	-0.02	0.11	-0.22	0.01	-0.08		0.82718
Uruburuella	-0.12	0.06	0.18	0.2	-0.17	-0.14	0.16	0.53	0.05	0.1	-0.11	0.05	

Appendix 19

Molecular identification of *Francisella* sp. isolated from *H. dromedarii* collected from camels in Abu Dhabi, UAE based on DNA similarity between 16S rRNA gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**
Uncultured *Francisella* sp. clone FraApaulo	MN998649.1	98.59	100	0.0	*Amblyomma paulopunctatum*	France
Francisella endosymbiont	JQ764629.1	98.59	100	0.0	*Dermacentor auratus*	Thailand
Ornithodoros moubata symbiont	AB001522.1	98.59	100	0.0	*Ornithodoros moubata*	Japan
Uncultured *Francisella* sp. clone FraAscul	MN998650.1	98.47	100	0.0	*Amblyomma sculptum*	France
Francisella endosymbiont	JQ764628.1	98.47	100	0.0	*Dermacentor auratus*	Thailand
Uncultured *Francisella* sp. clone FraAhume3	MN998638.1	98.35	100	0.0	*Amblyomma humerale*	France
Uncultured *Francisella* sp. clone FraAhume2	MN998637.1	98.35	100	0.0	*Amblyomma humerale*	France
Uncultured *Francisella* sp. clone FraAhume1	MN998636.1	98.35	100	0.0	*Amblyomma humerale*	France
Uncultured *Francisella* sp. clone 627HBF ZOTU 13a	MN088353.1	98.35	100	0.0	*Haemaphysalis bancrofti*	Australia
Uncultured *Francisella* sp. clone 297HBF ZOTU 13a	MN088349.1	98.35	100	0.0	*Haemaphysalis bancrofti*	Australia
Francisella endosymbiont of *Ornitnodoros porcinus*	AF166257.1	98.35	100	0.0	*Ornitnodoros porcinus*	USA
Uncultured bacterium clone he_23	KX465127.1	98.35	100	0.0	Tick	China
Uncultured bacterium clone he_17	KX465125.1	98.35	100	0.0	Tick	China

Uncultured bacterium clone he_6	KX465118.1	98.35	100	0.0	Tick	China
Uncultured bacterium clone he_5	KX465117.1	98.35	100	0.0	Tick	China
Uncultured bacterium clone he_1	KX465115.1	98.35	100	0.0	Tick	China
Francisella endosymbiont isolate XJ-S3	KX852466.1	98.35	100	0.0	*Hyalomma asiaticum*	China
Francisella endosymbiont isolate XJ-S1	KX852464.1	98.35	100	0.0	*Hyalomma asiaticum*	China
Uncultured *Francisella* sp.	MN998635.1	98.24	100	0.0	*Amblyomma goeldii*	France
Uncultured *Francisella* sp.	MN998634.1	98.24	100	0.0	*Amblyomma goeldii*	France
Uncultured *Francisella* sp.	MN998633.1	98.24	100	0.0	*Amblyomma goeldii*	France
Uncultured *Francisella* sp. clone FraAvari	MN998648.1	98.24	100	0.0	*Amblyomma varium*	France
Uncultured *Francisella* sp. clone 297HBF ZOTU 13a	MN088357.1	98.24	100	0.0	*Haemaphysalis bancrofti*	Australia
Francisella endosymbiont of *Hyalomm marginatum*	AF166257.1	98.24	100	0.0	*Rhipicephalus bursa*	Italy
Francisella-like endosymbiont	MH645205.1	98.24	100	0.0	*Hyalomma aegyptium*	Turkey
Francisella-like endosymbiont	MH645204.1	98.24	100	0.0	*Hyalomma aegyptium*	Turkey
Francisella-like endosymbiont	MH645203.1	98.24	100	0.0	*Hyalomma aegyptium*	Turkey
Francisella-like endosymbiont	MH645202.1	98.24	100	0.0	*Hyalomma aegyptium*	Turkey
Francisella-like endosymbiont	MH645201.1	98.24	100	0.0	*Hyalomma aegyptium*	Turkey

Appendix 20

Molecular identification of Uncultured *Rickettsia* sp. isolated from *H. dromedarii* collected from camels in Abu Dhabi, UAE based on DNA similarity between *ompA* gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**
Uncultured *Rickettsia* sp. clone C269_18	MK522488.1	99.80	100	0.0	*Amblyomma parvum*	Brazil
Candidatus Rickettsia andeanae clone Caxias	KY628370.1	99.80	100	0.0	*Amblyomma parvum*	Brazil
Candidatus Rickettsia andeanae haplotype BQ-RS	KX434737.1	99.80	100	0.0	*Amblyomma tigrinum*	Brazil
Candidatus Rickettsia andeanae clone 4	KX158267.1	99.80	100	0.0	*Amblyomma maculatum*	USA
Candidatus Rickettsia andeanae clone 3	KX158266.1	99.80	100	0.0	*Amblyomma maculatum*	USA
Candidatus Rickettsia andeanae isolate Agripino Enciso	KF179352.1	99.80	100	0.0	*Amblyomma parvum*	Paraguay
Candidatus Rickettsia andeanae isolate Ap	KF030932.1	99.80	100	0.0	*Amblyomma parvum*	Brazil
Uncultured *Rickettsia* sp. clone ALAIN-001-2011	KF156874.1	99.80	100	0.0	*Hyalomma dromedarii*	UAE
Rickettsia endosymbiont of *Amblyomma maculatum* strain SH_B4	JX134638.1	99.80	100	0.0	*Amblyomma maculatum*	USA
Candidatus Rickettsia andeanae isolate At2	MT968426.1	99.80	100	0.0	*Amblyomma tigrinum*	Brazil
Candidatus Rickettsia andeanae isolate At2	JQ690599.1	99.80	100	0.0	*Haemaphysalis* sp.	USA
Candidatus Rickettsia amblyommii isolate 61A	JQ690625.1	94.78	98	0.0	*Haemaphysalis* sp.	USA
Candidatus Rickettsia andeanae isolate G614	KX576678.1	99.76	83	0.0	*Amblyomma tigrinum*	Argentina

Candidatus Rickettsia amblyommii isolate GP4A	KM652487.1	94.27	99	0.0	Hard ticks	Panama
Candidatus Rickettsia amblyommii isolate 23B	JQ690590.1	94.76	97	0.0	*Haemaphysalis* sp.	USA
Uncultured *Rickettsia* sp. clone SH_MG7	JQ914762.1	94.08	99	0.0	*Amblyomma maculatum*	USA

Appendix 21

Molecular identification of *T. annulata* isolated from *H. anatolicum* collected from cows in Sharjah, UAE based on DNA similarity between ssrRNA gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**
Theileria annulata	MT341858.1	99.62	99	0.0	*Bos taurus*	Italy
Theileria annulata	MT341857.1	99.62	99	0.0	*Bos taurus*	Italy
Theileria annulata isolate T178	MT318160.1	99.62	99	0.0	Ruminants	Pakistan
Theileria annulata isolate T79	MT318159.1	99.62	99	0.0	Ruminants	Pakistan
Theileria annulata isolate T33	MT318158.1	99.62	99	0.0	Ruminants	Pakistan
Theileria annulata isolate Ticks, No 46	MN227669.1	99.62	99	0.0	Ticks	Egypt
Theileria annulata isolate Ticks, No 45	MN227668.1	99.62	99	0.0	Ticks	Egypt
Theileria annulata isolate Ticks, No 44	MN227667.1	99.62	99	0.0	Ticks	Egypt

Theileria annulata isolate Ticks, No 24	MN227666.1	99.62	99	0.0	Ticks	Egypt
Theileria annulata isolate 355	MN223736.1	99.62	99	0.0	Unpublished data	Egypt
Theileria annulata clone 5-31	AY508465.1	99.62	99	0.0	Cattle	Turkey
Theileria annulata isolate Turkey 4	AY508464.1	99.62	99	0.0	Cattle	Turkey
Theileria annulata isolate Turkey 3	AY508463.1	99.62	99	0.0	Cattle	Turkey
Theileria annulata	EU083801.1	99.43	99	0.0	*Bos taurus*	China
Theileria annulata	M64243.1	99.43	99	0.0	Bovine	USA

Appendix 22

Molecular identification of *T. ovis* isolated from *H. anatolicum* collected from goats in Sharjah, UAE based on DNA similarity between ssrRNA gene and GenBank species using NCBI BLAST.

Best match species	**Accession number**	**Sequence Identity %**	**Sequence coverage %**	***E*-value[a]**	**Host**	**Country**
Theileria ovis isolate HBOY1	MN394810.1	99.81	99	0.0	*Bos grunniens*	China
Theileria ovis isolate HXTS1	MN394809.1	99.81	99	0.0	Tibetan sheep	China
Theileria ovis isolate HBTS1	MN394808.1	99.81	99	0.0	Tibetan sheep	China
Theileria ovis isolate SH. T1	MN712508.1	99.81	99	0.0	Sheep	Iraq

Theileria ovis isolate SH. T5	MN704656.1	99.81	99	0.0	Sheep	Iraq
Theileria ovis isolate THOD2	MN625903.1	99.81	99	0.0	Donkey	Egypt
Theileria ovis isolate THOB2	MN625887.1	99.81	99	0.0	Buffalo	Egypt
Theileria ovis isolate THOSH5	MN625886.1	99.81	99	0.0	Sheep	Egypt
Theileria ovis isolate SH.S2	MN544931.1	99.81	99	0.0	Sheep	Iraq
Theileria ovis isolate 3kz7	MN493111.1	99.81	99	0.0	Sheep	Turkey
Theileria sp. Iwate 141 gene	AB602888.1	99.81	99	0.0	*Cervus nippon*	Japan
Theileria sp. Iwate 276 gene	AB602887.1	99.81	99	0.0	*Cervus nippon*	Japan
Theileria sp. Iwate 228 gene	AB602886.1	99.81	99	0.0	*Cervus nippon*	Japan
Theileria sp. Iwate 194 gene	AB602885.1	99.81	99	0.0	*Cervus nippon*	Japan
Theileria sp. Iwate 169 gene	AB602883.1	99.81	99	0.0	*Cervus nippon*	Japan